# QUANTIFICATION AND CHARACTERIZATION OF THE MOTION AND SHAPE OF A MOVING CELL

by

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# ABSTRACT

The main function of a blood cell's surface is to receive information from the environment. Recently, experiments have indicated that the cell membrane plays a vital role in the life, development, and regulation of cells. However, there is no existing method to quantify the observable changes in membrane shape that occur in locomotion. To achieve this objective using automatic techniques of digital image processing, the main goal of this research is to develop an image interpretation system capable of analyzing the structural changes in the morphology of a non-rigid moving object from a sequence of pictures.

A model for a general dynamic scene analysis system is described. It consists of three basic entities: dynamic data, static data, and a collection of analysis processes. Based on this model, we have implemented a rule-based image interpretation system for moving cells.

The system describes the dynamic behaviour of a moving cell using symbolic terminology which is meaningful to individuals working in cell biology. With the aid of this system, the global changes in the cell structure and pseudopod kinetics are analyzed. Hence, a subpart of the cell is classified as being either "pseudopod" or "cell body". A pseudopod is described as "growing", "contracting", or "stationary". Furthermore, other aspects of the global behaviour of the cell are characterized and described. For

example, the "domination" of a pseudopod in contributing to the locomotion of the cell.

The system is also applicable to the study of the dynamics of other white blood cell types. Ultimately, this type of study could allow the detection of abnormalities, and the efffects of drugs, if any, in the locomotory responses of leucocytes.

# RESUME

La principale fonction de la membrane d'un globule est de recevoir de l'information de son environnement. Recemment, des expériences ont démontré que la membrane joue un role primordial dans la vie, le développement et la regulation des globules. Toutefois, il n'existe pas de méthode permettant de quantifier les changements observables de la forme de la membrane au cours de la locomotion. Afin d'atteindre cet objectif tout en utilisant des techniques automatiques de traitement des images digitales, le but principal de cette recherche est de concevoir un système d'interprétation d'images capable d'analyser les changements structuraux de la morphologie d'un objet non-rigide en mouvement à partir d'une séquence d'images.

Un modèle de système général d'analyse de scènes dynamiques est décrit. Il se compose de trois entités de base: données dynamiques, donnees statiques et ensemble de processus d'analyse. En nous fondant sur ce modèle, nous avons réalisé un système "rule-based" d'interprétation d'images de globules en mouvement.

Ce système décrit le comportement dynamique d'un globule en mouvement à l'aide d'une terminologie symbolique qui est familière à ceux qui oeuvrent dans le domaine de la biologie cellulaire. A l'aide de ce systeme, les changements globaux de la structure cellulaire et de la cinétique des pseudopodes sont analysés. Ainsi, on classifie les parties d'un globule

comme étant soit "pseudopode", soit "corps du globule". Un pseudopode peut être "en expansion", "en contraction" ou "stationnaire". De plus, d'autres aspects du comportement global du globule sont charactérisés et décrits. Par exemple, la "domination" d'un pseudopode dans sa contribution à la locomotion du globule.

Le système s'applique aussi à l'étude de la dynamique d'autres types de globules blancs. Ultimement, ce genre d'études permettrait la detection d'anomalies des réactions locomotrices des leucocytes, et s'il y a lieu, des effets de certaines drogues sur celles-ci.

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# CHAPTER 1

# INTRODUCTION

## 1.1 BIOMEDICAL PROBLEM

Cell movement is a fundamental process of some importance to aspects of cell biology as diverse as migration of cells in embryological development and to host defense mechanisms. Advances have been made recently in the characterization of locomotory paths taken by cells in vitro and how these are affected by various substances. The internal mechanisms for cell locomotion are also reasonably well understood. However, progress has been much slower as to how the cell monitors external substances in order that internal mechanisms might be regulated. This interaction between external factors and cell internal processes has to occur at or within the cell membrane.

The thirty-eighth Symposium of the Society for Developmental Biology, held in Vancover B.C. in June of 1979, was mainly devoted to summarizing the current status of knowledge about cell surface (plasma membrane). At the conclusion of the symposium, Wessells stated [Wessells, 79]:

> "It has become increasingly evident that the cell surface plays a truly pivotal role in the life, development, and regulation of cells. On one hand, the surface functions in the transmission of information from the environment to the cell, and here I mean not only molecular signals, but also mechanical forces stemming from adhesions and junctions that affect the cytoskeleton and so intracellular activations. The surface is also in a real sense an expression of the cell's genetic information and developmental state. Embryologists and developmental biologists must pay increasing heed to the cell surface and to its changing properties."

Development in muticellular eukaryotes(*) must depend on mechanisms that extend beyond the usual notions inherent in our concepts of sequential gene activation. For example, development of an embryo requires that cells know where they are and where they should be. There must be mechanisms that regulate this social behaviour of cells and more than intuition informs us that the cell membrane is involved both as the donor and receptor of such social signals [Branton, 80].

The important activities of the cell membrane have now

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(*)Organisms made up of cells with nuclei bounded by nuclear envelopes.

been documented in terms of transport function, receptor function, and mechanical function. Study of receptor function and mechanical function have led to the realization that components of the cell surface are mobile in the plane of the membrane but that this lateral mobility is subject to regulation [Singer and Nicolson, 1972; Nicolson, 1976a]. Such regulated, lateral mobility has been the basis for many hypotheses on the molecular mechanisms mediating cell recognition and growth control [Edelman, 1976; Nicolson, 1976a,b]. In particular, it is often suggested that the interaction of extracellular ligands(*) with their cell surface receptors alters the distribution of transmembrane elements that can bind to motility-related proteins such as actin or tubulin at the cytoplasmic surface of the plasma membrane [Edelman, 1976; Bourguignon and Singer, 1977].

Although many observations give credence to such hypotheses, it is only recently that a direct chemical demonstration of the binding between membrane and cytoplasmic components has been demonstrated. If transmembrane elements can interact with components at the cytoplasmic surface of the membrane, one would like to know the precise nature of the binding sites, the affinities and specificities of the interaction, and the manner in which

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(*) A molecule that will bind to a complementary site on a given structure.

these affinities are regulated. Some progress has been made in this direction with the polymorphonuclear leucocytes (PMN) in that binding studies with the chemotactic agent, N. formylmethionineleucinephenylalanine(*), have been carried out [Williams et al., 77]. The locomotory organ of these cells is the pseudopod and changes in pseudopod activity are the first morphological events visible as the cells respond to chemotactic agents. Similarly, lymphocytes also show pseudopod activity during locomotion [Lewis and Webster, 21], and many studies have been done on membrane bound ligand interactions in this cell type. Little is known of the pseudopod kinetics of these cells during locomotion.

## 1.2 OBJECTIVES

As indicated in the previous section, pseudopod formation is an important property of locomoting cells, yet presently, there is no existing method for quantifying the observable changes in the membrane shape that occur during locomotion. Consequently, it is difficult to study the interaction, at the membrane level, between the cell internal processes and the external factors which modify cell locomotion. Therefore, what is required is a system to analyze a sequence of images of a moving cell to provide a quantification and description (numerically and

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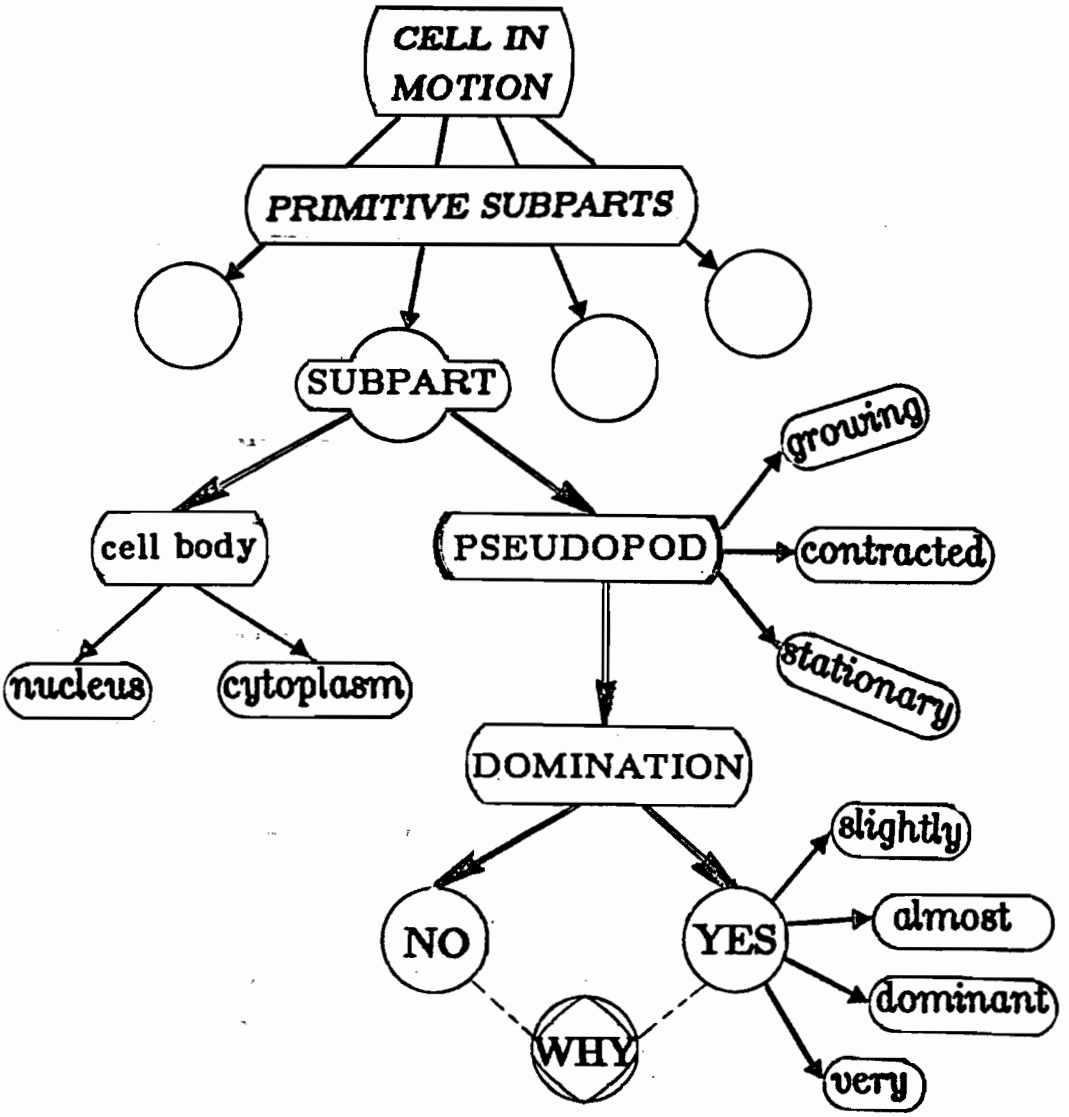
(*) Sometimes abbreviated as: f Met-Leu-Phe

symbolically) of the dynamic changes in membrane shape. For example, this study would prove useful in assessing whether leucocytes sensed a chemotactic gradient by a "temporal" or "spatial" mechanism [Zigmond et al., 81; Gerisch and Keller, 81]. Such a study would be of relevance to the understanding of the role of the cell membrane in the mechanisms which regulate the social behaviour of the cell, i.e directed cell migration.

To achieve this objective using automatic techniques of digital image processing, the main goal of this research is to develop an image interpretation system capable of analyzing the structural changes in the shape of a non-rigid moving object from a sequence of pictures. To do this, the system would have to be able to: recognize the various image patterns, segment and interpret the desired object, and detect the significant (global) changes in the location, shape, and structure of the moving object.

Using this system to analyze the dynamics of blood cell motion, biologists can obtain the quantifications and descriptions of the data necessary to understand or answer questions pertaining to the cell behaviour. Figure(1.1) illustrates certain concepts for analyzing the observable changes in the membrane shape in order to understand its role in regulating and modifying the cell locomotion. This figure refers to a moving cell under observation, and a biologist might be interested in the answers to three basic questions pertaining to the cell's dynamic behaviour.




*Figure(1.1)Characterization of the observable changes in the cell membrane of a moving cell, in order to understand the nature and distribution of "receptors" on or within the membrane. The properties of these receptors might explain the interaction between external factors and cell internal process, and how these mechanisms regulate the social behaviour of the cell.*

First, how can one recognize a subpart developed on or by the membrane as a "pseudopod"? And if it is a pseudopod, is it stationary? growing? or contracting? Second, is a pseudopod "dominant" (the dominant pseudopod is one which contributes to the cell locomotion) or not? If so, what is its degree of domination? Finally, if all this information is at hand, the third question would pertain to the interpretation of this data. For example, why is a specific pseudopod dominant and another not?

Our system has successfully provided all the quantification, description, and characterization information whereby the first two cited basic questions can be answered. Analysis of the data obtained may provide the answer to the third question.

In these studies we have looked at the pseudopod responses of PMN during random and chemotactic motion primarily because at the commencement of this study we had ample knowledge of chemotactic agents for PMN but very little was known of lymphocyte chemotactic agents. Another advantage of commencing this work using PMN is that this cell exhibits complex shape changes and therefore any technique devised to quantitate these changes can readily be applied to relatively simpler shape forms such as the lymphocyte.

## 1.3 RELEVANT AREAS OF STUDY

Constructing and implementing a system which has the capabilities of accomplishing the objectives cited in the preceding section, requires and represents a merging of four different disciplines in computer vision and image processing. They are: (a) Automatic Processing of Microscopic Images, (b) Image Sequence Analysis, (c) Shape Analysis and Description, (d) Knowledge-Based Systems.

In the automatic digital image processing of cell images, most of the work has dealt with static pictures of blood smears (or frozen cells) for the purpose of classification or counting. Previous work applied to the study of moving cells has concentrated on tracking cell paths only, rather than studying cell interaction characteristics. Furthermore, there is no existing system which concerns itself with the analysis of the structural changes in the cell shape. Consequently there is no method extant which quantifies and characterizes the observable changes in the cell membrane shape occuring during locomotion. Indeed, a terminology for these descriptions is not even available, and an objective of this research was to develop one in conjunction with physiologists.

Even if we extend our consideration to the general problem of processing dynamic images, this field has been largely restricted to motion detection, recognition, and tracking. Most of the previous research has attempted to

analyze an image sequence by considering the multitudinous data representing the incremental changes that occur between each two sequential frames from the sequence. This data mainly pertained to the changes in locomotion of an object, rather than the dynamic alterations in its shape. This is because most of the work in this field has dealt with the motion of rigid objects, having static three-dimensional shapes. Thus, two main issues have been ignored by most of the past research: (a) shape and structural changes of a non-rigid moving object, and (b) motion understanding and description. These issues are among the aspects addressed in this thesis.

In our current work we are dealing with non-rigid objects, which change their shape and structure randomly due to physical properties. Thus, we are considering three kinds of changes with time: locomotion, shape, and structure. All alter randomly from frame to frame and interact with each other. This randomness in images recording cell locomotion is partially due to the fact that the chamber simulating the environmental conditions of the live cells under the microscope permits the cells to move to some extent in three-dimensions. Consequently, the two-dimensional images resulting from filming these cells are actually recordings of three-dimensional changes.

To achieve the objectives of motion understanding and description, it is not enough to merely determine the incremental movements or changes that occur between consecutive images [Tsotsos et al., 80]. What is required is a system which abstracts a description of the global motion characteristics from the static and incremental data. Development of such a system represents the approach being taken in our present research in image sequence analysis.

The problem of shape discrimination is a central one to pattern recognition and as such has received considerable attention in most papers dealing with recognition of characters, waveforms, chromosomes, cells, machine parts, etc. Most of the work in the perception of shape has used numerical descriptors in terms of feature measurements such as sides, angles, moments, curvature, color, texture, etc. In our current research, besides the general difficulties of describing an arbitrary shape in a specific image, we are facing the following problems: (a) Estimating the incremental change in the shape and structure of a non-rigid moving object. (b) Detecting and characterizing the structural changes in the morphology of a non-rigid moving object over a period of time from a sequence of pictures. (c) Presenting all of the above descriptions in a meaningful terminology to the user.

We have developed a procedure which produces a meaningful symbolic description of the shape and its changes. We have also developed an expression for measuring the complexity of an arbitrary shape pattern. This expression is based on a group of selected shape properties which are independent of location, translation, rotation, or scaling. Another shape property is introduced to measure the degree of curvature regularity (angle or side regularity) of the shape of an object. This property is shown experimentally to play a considerable role in shape discrimination and is used to describe membrane shape.

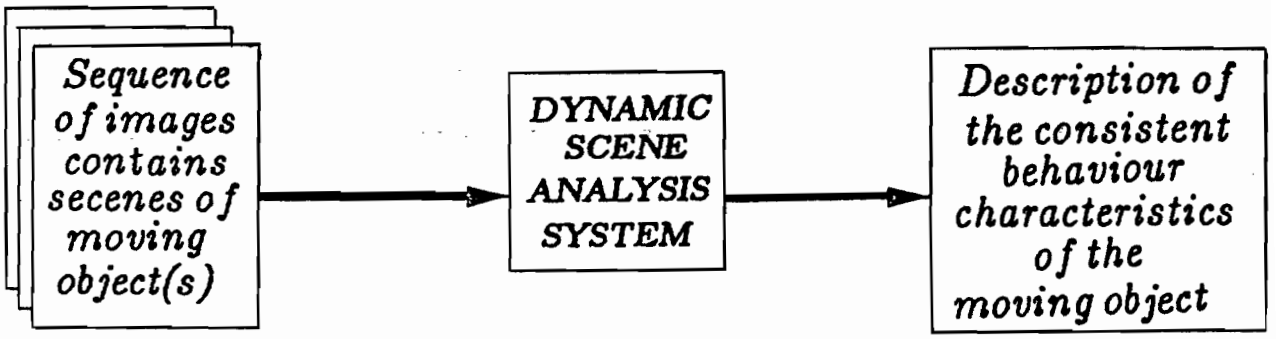
An understanding system, either as a computer vision system for static scene interpretation, or visual motion description from a sequence of images, requires the construction of a knowledge-based system. This system should utilize the knowledge from diverse sources of information, consisting of multiple levels of analysis, and to be supported by an efficient control structure mechanism. The progress towards the development of knowledge-based systems for visual motion understanding is slow, and the work which has been done is very limited. In our research we have utilized the most advanced strategies of computer vision interpretation of static images, merging them with the experience gained in image sequence analysis to construct a visual motion understanding system.

From a philosophical point of view, the present structure is motivated by the computer vision framework proposed by Levine [Levine, 78]. The latter has been revised and implemented by Levine and Shaheen for general static scene analysis and interpretation [Levine and Shaheen, 81]. Low level segmentation has been reported in [Levine and Nazif, 82]. The approach is based on independent processes that cooperate through a common database structure. The system and data structure model for a general motion understanding system developed in our research is described in Chapter 3 and briefly reviewed in the following section.

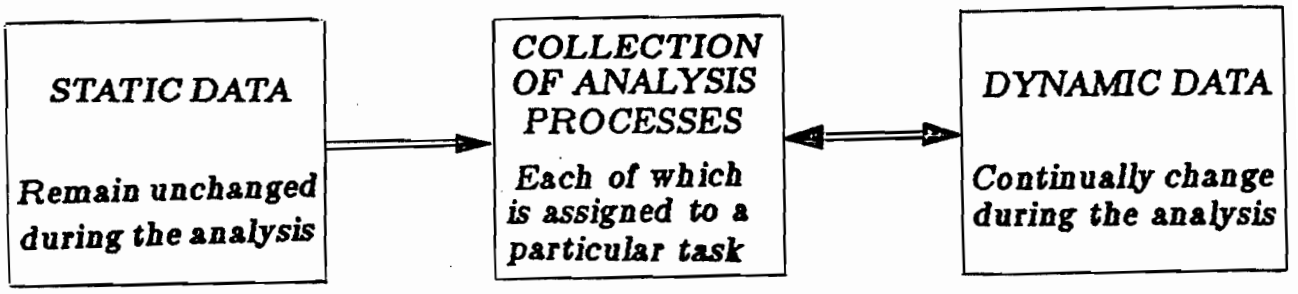
## 1.4 MODEL STRUCTURE AND SYSTEM IMPLEMENTATION

The main input for any image sequence analysis system is a series of two-dimensional digital images representing the variation in a specific scene along a third-dimension. The function of the system is to generate a description of the consistent characteristics and behaviour of the moving object(s) recorded within the sequence (see Figure 1.2). A theoretical model for a general dynamic scene analysis and motion understanding system has been developed. It involves three basic entities (Figure 1.3):

(a) Static (constant) data: The data which remains unchanged during the course of analysis. They contain constraint knowledge pertaining to the class of scenes and motion under analysis, as well as the control




*Figure(1.2) The main input/ouput data of a dynamic scene analysis system.*




*Figure(1.3) Basic structure units of a dynamic scene analysis system.*

information describing the pertinent computational processes.

(b) A collection of analysis processes each of which is assigned to a particular task.

(c) Dynamic data: The data which are continually changing as a result of the functioning of the different analysis processes.

The different types of data which may be manipulated by the system have been classified by the model into:

(a) Sequence of images: A set of two-dimensional images which represents the main input of the system. Each element of this set is a static image of the scene at a specific time.

(b) Set of objects: Each image of the temporal sequence may be segmented into a set of objects. This set may also be divided into two subsets according to whether the object is moving or stationary. Objects have complex shapes, and it is usually necessary to decompose them into primitive subparts. The result of this decomposition is a collection of subobjects associated with each object.

(c) Set of features: A set of objects is described by static features, which define the different properties of shape, structure, or motion of the objects and subobjects to be measured or analyzed by the system.

(d) Group of symbolic descriptors: A set of symbolic descriptors or qualifiers is used to classify and describe the numerical values of the different properties of the moving objects.

(e) Group of characteristics: Each characteristic is the description of a group of features which cooperate to define a specific type of behaviour. These may be based on the global changes in the cell's shape, structure, motion, combination of more than one change, and/or the effect of the environment on the cell's behaviour.

(f) Set of rules: The model which represents the knowledge contains two basic types of data: constraint knowledge and rules. The latter may be further classified into representational and control rules. The representational rules are responsible for generating the different descriptions and characteristics according to the numerical measurements of the different features. The control rules account for the activation and scheduling of the different system processes.

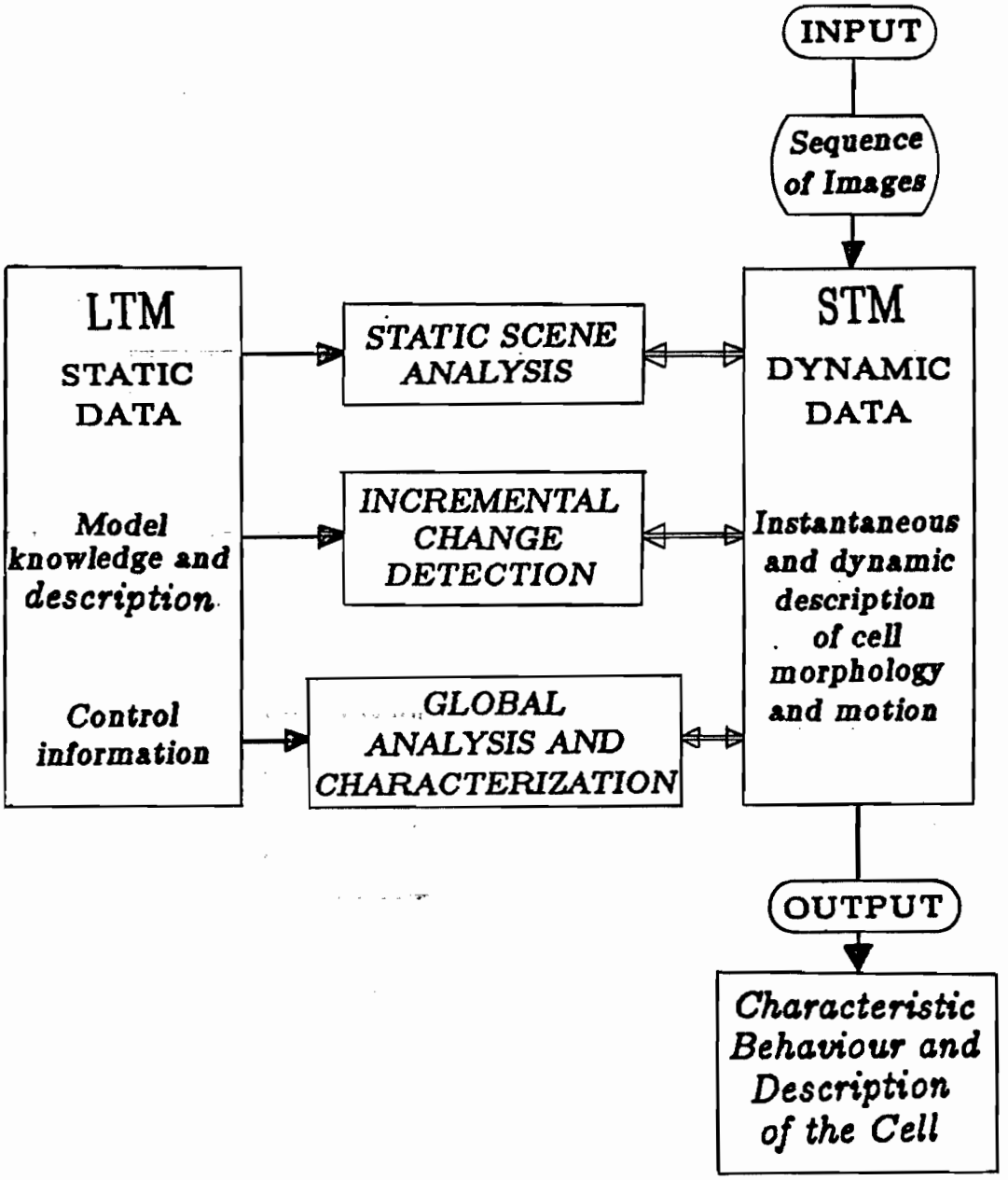
The dynamic and static data are designed as two associative data memories: a Short Term Memory (STM) and a Long Term Memory (LTM). All the analysis processes can communicate with both the STM and LTM. Both the STM and LTM are implemented as a relational database. The STM contains

a record of the instantaneous cell motion, shape, and structural changes, as well as, the current description of the cell behaviour. The LTM contains the general model of the morphology of the cells under analysis, as well as control information describing the pertinent computational processes.

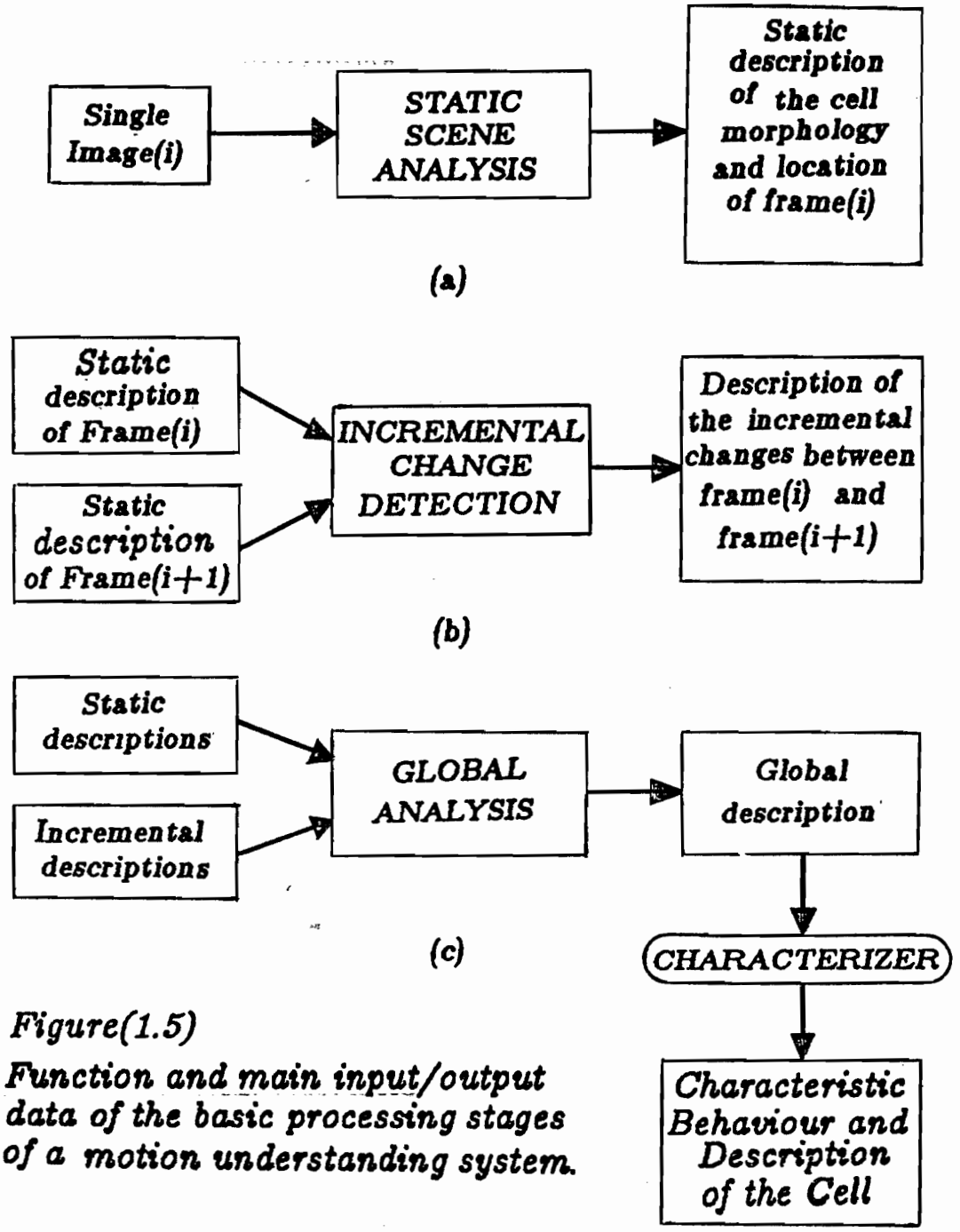
The system consists of different computational processes which are designed to execute through a hierarchial structure consisting of three basic levels: static, incremental, and global (see Figure 1.4).

a)Static Scene Analysis: This step is similar to a conventional image processing system in that the input is a single digital image, and the output is a description and interpretation of the scene. However, in the analysis of dynamic images, the information extracted from the previous frames of the same sequence may also be used as knowledge to assist in the analysis of the current frame. The main objective of this stage is to identify the desired moving object, segment it, and describe it in each frame of the sequence (see Figure 1.5a).

(b) Incremental Change Detection: This is an intermediate step between the static and the global. The main objective is to detect and describe the incremental changes in shape, structure, and motion of the moving object (see Figure 1.5b).




*Figure(1.4) Main processing stages and data structure of a motion understanding system*




*Figure(1.5)*

*Function and main input/output data of the basic processing stages of a motion understanding system.*

(c) Global Analysis: This is the highest level in the hierarchy of the dynamic scene analysis system. The goal is to analyze the static and incremental data in order to detect and describe the global observable changes within the sequence of frames. In this way, the characterization of the consistent dynamic behaviour of the cell may be obtained (see Figure 1.5c).

From the above discussion, we can see that the separation of the knowledge and control information in the LTM from the analysis processes will allow for the application of the system to different classes of scenes or motion. Storing the dynamic data in an associative memory (STM), completely separate from the analysis processes, was necessary in order to achieve the complete independence of the processes. This means that each process will communicate data to and from the STM and not to the other processes, a fact which enhances the consistency of the overall system data. Moreover, the complete independence of the analysis processes allows for the modularity and extensibility of the system.

In conclusion, a system for quantifying and characterizing the motion and structural changes in the shape of a non-rigid moving object has been developed and applied to analyze the cell's dynamic behaviour.

# 1.5 SUMMARY AND GENERAL OVERVIEW

## 1.5.1 Summary

The main function of a blood cell's surface is to receive information from the environment. Recently, experiments have indicated that the cell membrane plays a vital role in the life, development, and regulation of cells. However, there is no existing method to quantify the observable changes in membrane shape that occur in locomotion. To achieve this objective using automatic techniques of digital image processing, the main goal of this research is to develop an image interpretation system capable of analyzing the structural changes in the morphology of a non-rigid moving object from a sequence of pictures.

A model for a general dynamic scene analysis system is described. It consists of three basic entities: dynamic data, static data, and a collection of analysis processes. Based on this model, we have implemented a rule-based image interpretation system for moving cells. The system consists of different cooperating computational processes, which interact with two common memories, a Short Term Memory (STM) and a Long Term Memory (LTM). The STM contains a dynamic record of the instantaneous cell motion, shape, and structural changes, as well as the current global description of the cell behaviour. The LTM data are static, and are implemented as rules. These describe the general

model of the morphology of the cells under analysis, as well as control information pertinent to the computational processes. The latter are activated by the control rules throughout the three hierarchical analysis stages: static, incremental, and global. They interact through the STM using the information stored in the LTM, until a complete description of the dynamic cell motion and morphology is obtained.

It is of interest to describe the dynamic activity of the cell using a symbolic terminology which is meaningful to the concerned biologist. With the aid of the global observable changes in the cell locomotion, one of the main behavioural characteristics is mathematically quantified and described, namely, the chemotaxis behaviour. This refers to the directional locomotion of the cell with respect to the directional effect of an external factor. Consequently, the effectiveness of an external factor on modifying the cell locomotion is quantified. Also, a mathematical expression for measuring the complexity of an arbitrary shape pattern is developed and demonstrated in describing the membrane shape and quantifying its observable changes. The global changes in the cell structure are also analyzed; hence, a subpart of the cell is classified as "pseudopod or cell body", and a pseudopod is described as "growing, contracting, or stationary". Furthermore, some aspects of the global behaviour of the cell are characterized and described. For example, the "domination" of a pseudopod in

leading the locomotion of the cell.

This study might provide clues to the nature and distribution of "receptors" on or within the membrane which would be a vital link in the interaction between the external factors and cell internal processes. Also, it might lead to the understanding of the roles cell membrane play in the mechanisms which regulate the social behaviour of the cell.

It is interesting to note that this technique is also applicable to other similar problems. Examples are the visual monitoring of the behaviour of rats under the influence of various drug protocols, or the quantification and analysis of the changes of growing plants in different soils or under the effect of different fertilizers.

## 1.5.2 Review Of Chapters

The thesis consists of nine chapters. Chapter 1 is the introduction which briefly describes all the aspects and objectives of our current research. Chapter 2 is a review of the significant work which has been done in the relevant areas of study. These are: (a) Image Sequence Analysis, (b) Shape Analysis and Description, (c) Knowledge-Based Systems, and (d) Automatic Processing of Microscopic Images. The critiques which are presented in this chapter are aimed at: (a) A brief review of the significant work in each of the cited fields, focusing attention on those related to our

research. (b) Analyzing and summarizing the progress in each of these fields as a gained experience. (c) Comparing our current work in this research to that which has preceded it, and demonstrating the possibility and advantages of integrating the experience from the different fields in order to utilize it in our present study. Although, we have restricted our review in this chapter only to the work that is relevant to our research, the resulting critique is lengthy. Therefore, the busy reader may wish to only refer to Sections 2.2.4, 2.3.11, 2.4.7, and 2.5.7, which represent summaries of the progress in each of these fields and the contribution of our work in each.

The system and data structure of a theoretical model for a general dynamic scene analysis and motion understanding system is discussed in Chapter 3. The chapter includes five sections. Section 3.1 is an introduction and general overview of the present structure. The data structure and knowledge representation is presented in Section 3.2. This section defines all the types of data which may be manipulated by the system and the basic elements for knowledge representation, as well as their mathematical definitions. Section 3.3 describes the structure of the rules that are responsible for knowledge representation, and Section 3.4 describes those associated with the control structure of the system. The last Section 3.5 is a summary.

Chapter 4 describes the lowest level of analysis, that is, static scene analysis. The main objectives of this stage are to identify the desired moving object, segment it, and describe it in each frame of the sequence. This stage consists of the four main processes shown in Figure (4.1). The first process is concerned with the extraction of the cell under analysis from the input image; this is described in Section 4.2. An algorithm for generating the polygonal approximation of the cell boundaries is described in Section 4.3. Section 4.4 presents the approach for decomposing the cell into its primitive subparts. A discussion pertaining to the selection of the properties to be measured, and their theoretical definition is given in Section 4.5. Finally, Section 4.6 details a process which summarizes the cell morphology to generate a description of the cell in the current frame.

Given the location and geometric features of the cell in two different frames, Chapter 5 presents processes for detecting, qualifying, and describing the incremental changes in the location, shape, and structure of the cell and its subparts between the two frames (see Figure 5.1). The discussion in this chapter is given in the three main sections 5.2, 5.3, and 5.4. They describe the changes in the location, shape, and structure, respectively. In each of these sections, the different aspects associated with incremental change detection, qualification, and description is discussed. Section 5.5 is a summary.

Chapter 6 describes processes for global locomotion analysis and description (see Figure 6.2). The input data for this stage are the static location of the cell in each frame, the incremental displacement, and the direction of motion between two sequential frames. The output is a description of the cell locomotion behaviour. First, the automatic cell tracking to construct its path, and extract the path parameters is described in Section 6.2. This includes a description of methods for reconstructing a smooth and simple cell path in order to retain a record of the significant changes. Also, techniques for detecting and removing any artifact of cell movement due to noise or undesirable experimental conditions are demonstrated. Then, the motion analysis and description is discussed in Section 6.3. Based on measurement parameters of the cell path, a quantification and symbolic description of the chemotaxis behaviour of the cell is provided in Section 6.4. In this way, the effectiveness of an external factor on the global cell locomotion is quantified and described. Finally, in Section 6.5, a summary of this chapter is presented.

A quantification and symbolic description of the observable changes in the cell shape is given in Chapter 7. Figure (7.1) shows the main processes and data structures used in shape analysis and description. The input to this stage of the system consists of the static and incremental description of the shape properties in and between each

frame. The output consists of a summary describing the changes of the cell shape and their characterization. The methodologies and processes which are used to accomplish this are discussed in Chapter 7 in the following order. The basic methodologies and techniques for detecting the global changes from the static and incremental data are described in Section 7.2. Description of the global changes in each of the main shape properties is given in Section 7.3. In Section 7.4, we show experimentally that an individual shape property is not sufficient to describe an arbitrary shape. Also in the same section, we discuss the development of a mathematical expression for the membrane shape measure. In this way, the global changes in the cell membrane shape can be characterized and described. Finally, Section 7.5 is a summary of the chapter.

Two main issues are addressed in Chapter 8, global structural analysis, and characterization of global dynamic behaviour. The objective of the first issue is to analyze the static and incremental structural descriptions in order to generate a summary of the global structural changes. This analysis includes techniques to detect any false decompositions of the cell in the low level processes, due to irrelevant change, noise, error in the segmentation, or experimental conditions. Using the LTM rules, we show how to reconstruct the cell structure by modifying the low level decomposition using high level information for feedback. Besides generating the description of the global observable

changes in the cell structure, a description of the morphology and locomotion of each individual subpart is given for the period of time it appears. In this way, a subpart of the cell is classified as pseudopod or cell body. Then a pseudopod is described as stationary, growing, or contracting.

The second part of Chapter 8 is concerned with the integration of the three global aspects pertaining to locomotion, shape, and structure, in order to understand and characterize the dynamic behaviour of the cell. In this high level process, the symbolic description of the observable changes in the cell shape, structure, and motion are the essential data to characterize the consistent dynamic behaviour of the cell. The last section of Chapter 8 discusses one of the basic questions related to understanding the role the cell membrane plays in the mechanisms which regulate its social behaviour. That is, the domination of a pseudopod in the global locomotion of the cell. Thus, a characterization of the degree of pseudopod domination is defined.

Finally, in Chapter 9, the research is summarized and discussed. Different experimental conditions and results are also presented. An evaluation of the computer analysis and characterization is given based on a comparison to those obtained by a physiologist. The possiblity of different applications and future work conclude the thesis.

Two appendices have been added at the end of the thesis: Appendix (A) presents a short mathematical proof of the formulas that are used for measuring angle and side regularity of the contour of an arbitrary shape. Appendix (B) describes the laboratory methods and facilities, including blood cell preparation and the computational facilities.

# CHAPTER 2

# A CRITIQUE OF THE LITERATURE

## 2.1 INTRODUCTION

The main objective of our research is to design and implement a knowledge-based system capable of analyzing, understanding, and describing the visual motion of a non-rigid moving object. Using this system for quantifying and characterizing the structural changes in the morphology of a moving cell from a sequence of pictures, we may be able to understand and describe the cell dynamic behaviour. Constructing and implementing a system which has these capabilities requires and represents a merging experience at the intersection of four different disciplines in computer vision and image processing. They are:

(a) Knowledge-based Systems,

(b) Image Sequence Analysis,

(c) Shape Analysis and Description,

(d) Automatic Processing of Microscopic Images.

The recent rapid growth in each of these fields of study makes an exhaustive survey of any one of them a thesis on its own. Table(2.1) gives some of the significant references and surveys in each field. However, the critiques which are presented in this chapter are aimed at:

(a) A brief review of the significant work in each of the cited fields focusing our attention on those related to our research.

(b) Analyzing and summarizing the progress in each of these fields as a gained experience.

(c) Comparing our current work in this research to that which has preceded it, and demonstrating the possibility and advantages of integrating the experience from the different fields in order to utilize it in our present study.

The review in this chapter is organized as follows: First, the general problem of analyzing a sequence of images is discussed in Section 2.2. This section shows the rapid progress in this field from empirical techniques for change detection between two images based on low-level pixel comparison methods, to the recent trend of using high-level global symbolic descriptors for visual motion understanding. Second, the problem of shape perception is described in Section 2.3 as a composition of two hierarchical processes, shape analysis and shape description.

# TABLE(2.1)

## (1) Image Sequence Analysis:

| | |
|---|---|
| [Martin and Aggarwal, 78] | survey |
| [Nagel, 78;79] | survey |
| [Scacchi, 79] | survey |
| IEEE, Trans. on Pattern Analysis and Machine Intelligence, Vol. PAMI-2, No. 6, Nov. 1980. | |

## (2) Shape Analysis and Description:

| | |
|---|---|
| [Pavlidis, 78;80] | surveys |
| [Meagher, 79] | survey |
| "Structural Pattern Recognition", Springer_Verlag, New York. 1977. T. Pavlidis, (ed.) | book |

## (3) Knowledge-Based Systems:

| | |
|---|---|
| [Levine, 78; Hanson and Riseman, 78a;78b] | articles |
| "Computer Vision Systems" A.R. Hanson and E.M. Riseman, (eds.), | book |
| [Rychener, 81] | Bibliography |
| "Pattern-Directed Inference Systems" D.A. Waterman and F. Hayes-Roth (eds.) | book |

## (4) Automatic Processing of Cell Images:

| | |
|---|---|
| [Preston, 76] | article, survey |
| Digital Picture Analysis in Cytology, in "Digital Picture Analysis" | |
| [Bartels and Wied 77] | survey |
| IEEE, Trans. on Pattern Analysis and Machine Intelligence, Vol. PAMI-2, No. 5, Sep. 1980. | |

Table (2.1) Major references in areas related to research in understanding the dynamic behaviour of non-rigid moving object.

The different techniques and algorithms developed in each stage are discussed, as well as the advantages and disadvantages of each scheme for different applications. Third, in Section 2.4 the art of the "knowledge engineer" is discussed through the different strategies for the structure and implementation of knowledge-based systems. In this respect, two major problems are of interest: knowledge representation, and the control structure which makes an efficient use of this knowledge. Different approaches to these problems are discussed. Also, examples of knowledge-based systems for computer vision and visual motion understanding are reviewed in Section 2.4.

## 2.2 DYNAMIC SCENE ANALYSIS

### 2.2.1 Introduction

The input data to a static image processing system is a digital image which is obtained by quantizing the sensor signal from one or several spectral channels at each grid point of a two-dimensional raster. If this sampling process is extended to include time as a third dimension, the resulting samples are a sequence of images. The dynamic scene analysis system is the system which analyzes this sequence of images by studying the variations from frame (image) to frame due to the motion of an object(s) recorded within the sequence. The objective of this analysis is to describe the observable changes throughout the sequence of images in order to study or characterize the behaviour of

the moving object(s) (see Figure 1.2). The system which attempts to solve this problem thoroughly is attempting to imitate visual motion perception. This perception constitutes a hierarchy of processes which include motion detection, understanding, and description. All of these aspects are addressed by this thesis.

Image sequence analysis differs from scene analysis of one image in that not only must information be extracted from each frame, but in addition, information must be extracted from the sequence as well. This means that the details derived from each image must be integrated into a coherent whole. This integration is not a simple compilation of facts because changes in the scene are continually occuring due to the motion of the sensor or the object(s) in the scene [Martin and Aggarwal, 78].

On the other hand, image sequences provide information which may assist the analysis, so that more efficient results may be obtained. Thus, the results of processing previous images from a sequence may be used to guide segmentation and feature extraction processes of the current image. Also, the results of processing later images may be used to clarify ambiguities (for example, due to object occlusion or poor image quality) in previous images [Yachida et al., 78].

Ulstad [Ulstad, 73] has related the history of this problem to the work done in 1920 by Stillman which was concerned with the detection, using analog methods, of small changes between two photographs. However, most of the work that has been undertaken on image sequence analysis is relatively recent, depending, as it does on digital image processing. Attention to the analysis of image sequences as a field of study in its own right was actually only started in 1979 when the first international workshop took place. The work on sequence image analysis in the last few years has been quite extensive. This fact can be realized from the recent survey by Nagel [Nagel, 79]. In this survey, he reviewed some (not all) of the aspects of sequence image analysis; 517 articles are cited by 683 different authors.

In order to focus our attention on the main trends of our research, the review of the previous work in image sequence analysis will be restricted to three sections. First, the major recent surveys in this field will be discussed [Martin and Aggrawal, 78; Nagel, 78;79; Scacchi, 79]. Second, the individual work which is directly related or employs similar techniques to those we are using in our current research is reviewed. In the third section, conclusions and general remarks are given, as well as a comparison of our current research with the previous work done in sequence image analysis.

## 2.2.2 REVIEW OF SURVEYS

### 2.2.2.1 [Martin And Aggarwal, 78]

The first survey to review the work in dynamic scene analysis was presented by Martin and Aggarwal [Martin and Aggarwal, 78]. They first discuss the perception of motion in biological systems. Then, in the remainder of their report, they review the different literature in dynamic scene analysis under two classes: motion detection, and motion analysis. The following is a brief review of this paper.

In the perception of motion in biological systems, Martin and Aggarwal define two phases of visual perception: peripheral and attentive processes. The peripheral process must be able to detect motion and direct the attentive process to it, while the latter must be able to track the movement and attend to the details of the object in motion [Chien and Jones, 75]. It is claimed that these two phases of perception are not cognitive processes. A higher level cognitive process is probably required to relate both of these processes to the current "psychological set" of the person, his knowledge, and expectations. In this discussion of motion perception in a biological system, Martin and Aggarwal refer to the work in [Johansson, 75; Hubel and Wiesel, 59; Lettvin et al., 59; Barlow and Hill, 63; Mackay, 61; Schouten, 67].

In terms of the structure of the biological vision systems, Martin and Aggarwal define and describe the dynamic scene analysis system and its role. They address two basic functions: (a) to associate "semantically identical" images, (b) to solve the occlusion problem. The different techniques in dynamic scene analysis are classified into two main classes: motion detection (peripheral process), and motion analysis (attentive process). The methodologies which have been used for motion detection were mainly concentrated on cross-correlation and image differencing; whereas in motion analysis, the centroid matching and shape analysis techniques were used.

In the cross-correlation technique, the "cross-correlation coefficient" is computed (using the FFT) for each pairing of pixels in a section of an original picture to a candidate one in the second picture. The candidate section which yields the maximum coefficint is chosen as the match. Most of the systems which have used cross-correlation techniques were originaly designed for estimating cloud motion. Among these is the work by [Leese et al. 70; 71; Smith and Phillips, 72; Lo and Parikh, 73; Arking et al., 75].

The image differencing techniques are based on determining areas of change between two different images of the same sequence. The areas of change are found by a simple subtractive process. Therefore, the images must be carefully normalized, with respect to both spatial

coordinates and intensity value. Thus, these types of techniques do not attempt to recognize any particular feature in either of the two images; consequently, they cannot describe any of the motion features. Examples of systems which have used this type of technique were presented in the work of [Lillestrand, 72; Ulstad 73; Limb and Murphy, 75; Nagel, 76].

In the centroid matching techniques, the objects are reduced to centroids, making spatial location the only feature of the objects. While this approach makes the solution to the tracking problem rather simple, it destroys all the features needed for shape analysis or solving occlusion problems. Centroid coordinates as descriptors of objects are used in the tracking approaches of [Endlich et al., 71; Greaves, 75; Levine and Youssef, 78; Levine et al., 81].

In summary, the review by Martin and Aggarwal discusses two low level supposedly non-cognitive processes to analyze a sequence of images, motion detection (peripheral) and motion analysis (attentive). Their study leads to the conclusion that additional research is needed to derive systems which use both levels of analysis, as well as employing higher level cognitive processes to exploit the parallelism inherent in the visual process. The first step taken in this direction was by Badler [Badler, 74] in generating a scenario from a sequence of two-dimensional images.

### 2.2.2.2 [Nagel, 78]

The first comprehensive survey to discuss the problem of motion analysis based on a digital image sequence was presented by [Nagel, 78]. In this survey, the analysis of image sequences has been emphasized in two main aspects: First, the different experience in image sequence analysis is evaluated according to specialized application areas. Second, the schema within which the different approaches are organized, according to the techniques used for interframe comparison. The different approaches for interframe comparison are classified by Nagel into the following six categories:

(1) No Interframe Comparison: The approaches included in this category use only interframe image processing techniques to derive a sequence description which is subsequently evaluated by different means such as human perception or non-pictorial data processing. Since the frames of a sequence are not compared with each other, no dissimilarity function is required. For example, the sequence description of Tasto [Tasto, 73; Tasto et al., 78] consists of a series of ordered lists of coordinate pairs. Each list represents a left vertricular contour at each frame of the sequence. Other approaches by [Jones, 74; Chien and Jones, 75] can also be included in this category.

(2) Indirect Interframe Comparison: In this category, the comparison is performed indirectly by detecting the change in a specific measured feature from sequential images or windows of the images. For example, Uno [Uno et al., 76] inspected the displacement of an object moving horizontally from the center of a fixed window. This displacement is assigned a positive or negative value, depending upon whether the object passed the window center or not. Then, an interframe comparison is performed by detecting the sign change in the subsequent frames. The indirect interframe comparison in the work of Tasto [Tasto, 73;74] is performed by finding an initial estimate for a left ventricular contour, which is then tracked in the subsequent frames.

(3) Dissimilarity Grading: The approaches which belong to this category require that images from the sequence are registered with respect to each other, as in video frame sequences from stationary cameras. These approaches are mainly based on detecting the changes in the grayvalue of the corresponding raster position between two sequential images. The number of raster points (pixels) which is used as a neighborhood for determining the change are defined by [Nagel, 78] as the ORDER of the descriptors for the dissimilarity grading. He discusses different approaches, using a different ORDER of descriptors. In general, most of the algorithms which assign the different components of an image of a

sequence as stationary or non-stationary, use techniques which can be categorized as dissimilarity grading. For example, in the approach of Hogg [Hogg, 76; 77], the non-stationary image components are determined by change detection with respect to a reference frame. The descriptor involved in the dissimilarity grading is the pixel grayvalue (ORDER = 1). The velocity determination for video images of moving objects is demonstrated by Nagel [Nagel, 78]. His technique is based on several approaches [Limb and Murphy, 75a; 75b; Cafforia and Rocca, 76; Fennema and Thompson, 78].

(4) Similarity Search followed by Dissimilarity Grading: The interframe comparison techniques which belong to this category use a similarity search procedure in order to find the best match between the substructures from the two images to be compared. Once sufficient correspondence is achieved, both images can be registered on a single raster in order to determine the difference (dissimilarity grading). The early approaches in this category are rarely made explicit. Nor do they even provide, in a formalized manner, the appropriate substructures, their descriptors, and the way the similarity search is guided [Nagel, 78]. The similarity search between the two images to be compared by Price [Price, 76; and Price and Reddy, 77] is performed by comparing the description (group of selected features) for each region from one image with

the description of each region from the other. Then, the dissimilarity grading may be performed in more than one way. One alternative consists of comparing the actual feature values of best-match regions. In fact, the approach of Price has, to some extent, a common basis with our technique for incremental change detection; therefore, a detailed description of his work is presented in a later section of this chapter.

(5) Dissimilarity Grading, followed by Similarity Search: In the approaches assigned under this category, first, a dissimilarity grading procedure is used to detect areas where changes have occurred (non-stationary components). Then, these non-stationary components may be tracked from frame to frame (using a similarity search) in order to gather more information pertaining to the moving object. Consequently, a complete description of the object dynamic behaviour may be generated. Dissimilarity grading is illustrated in the work of [Yachida et al., 78] by selecting non-stationary image components which are subsequently tracked from frame to frame using several descriptors. Technical details of Yachida's work will be given in a later section of this chapter. Another example of approaches belonging to this category is in the work of [Nagel, 78]. Two subsequences (A, B) were chosen from a series of TV-frames so that the image of a moving object in a frame from subsequence B never overlapped the object

image in the corresponding frame of subsequence A. Then, a dissimilarity function was defined. It was based on the likelihood ratio for the hypotheses that the grayvalues observed in the overlap of two regions from different frames had been sampled from the same or from two different normal graylevel distributions. Finally, the similarity search is performed using the cross-correlation of segment edges for candidate images from adjacent frames.

(6) Similarity Search: Most of the approaches which are concerned with tracking moving objects through a sequence of images can be assigned to this category. The similarity search procedure is frequently applied using cross-correlation techniques [Leese et al. 70; 71; Smith and Phillips, 72; Arking et al., 75]. Another set of approaches assigned to this category, are those which are based on centroid coordinates as descriptors for tracking moving objects [Endlich et al. 71, Greaves, 75; Levine and Youssef, 78].

The survey of [Nagel, 78] discusses the different aspects of image sequence analysis through a review of the different approaches and techniques which have been used. These are classified and discussed according to: (a) their application areas, (b) the methodologies they use for interframe comparison. Discussing the different approaches according to their application areas is the main objective of an extensive recent survey by Nagel [Nagel, 79]. This

survey is reviewed in the next sction. The techniques used for interframe comparison are classified into the six categories listed above. The purpose of this classification is to demonstrate the wide variety of possibilities for interframe comparison at different levels; from comparing individual pixels from different images to comparing symbolic descriptors.

Aspects like segmentation and velocity determination of moving objects, in terms of interframe coding, are discussed by Nagel [Nagel, 78]. The survey concludes with remarks on the overall field. Specifically, the question, " What conclusion may be drawn from observable variations in descriptor values for the image of a moving object?". In other words, can a global description be generated from the observable changes?. To achieve this type of description, problems such as the loss of a tracked object due to occlusion, or poor image conditions, must be investigated on a global level. In this respect, Nagel suggests that the research in the analysis of image sequences could contribute to the task of modeling the dynamic environment (model-based systems).

A final, noteworthy, remark by Nagel is that researchers working on the analysis of image sequences should be aware of the increasing diversity of applications as a gained experience. Based on this suggestion, he has made a considerable effort to gather the accumulated individual experience and investigations from different

applications and has presented them in a coherent review. This application-oriented survey by Nagel [Nagel, 79] is briefly discussed in the next section.

### 2.2.2.3 [Nagel, 79]

A thorough discussion of the analysis of image sequences is given in the most recent survey by Nagel [Nagel, 79]. In this survey, more than five hundred articles have been listed from a wide scattering of application-oriented journals, conference proceedings, periodicals, texts, and technical reports. This well-documented article will appear as a chapter in the book entitled "Image Sequence Analysis" and edited by T. S. Huang. The different areas of application are organized by Nagel to include: coding of image sequences, processing image sequences from airborne and satellite sensors, medical applications (image sequences of the human body), biomedical, behavioural studies, object tracking in outdoor scenes (traffic monitoring and target tracking), industrial automation and robotics, and spatial image sequences.

The main objective of the survey is to demonstrate the parallels between the different application areas, so that the commonalities in basic problems, processing techniques, and underlying concepts may become discernible. This investigation might facilitate the transfer of solution approaches from one application to another. Another advantage is that the development of some application areas

may have a future analog in certain other areas. Furthermore, the survey reviews the literature in sufficient detail to enable a non-specialized reader to decide whether or not an approach is relevant to him in the context of current or future development. The survey is documented with a comprehensive bibliography, augmented by an author index which facilitates access to the literature.

The significance of this survey by Nagel is that it summarizes the current status of knowledge on image sequence analysis, the shortcomings in the present approaches, and what the future work should offer. This may be realized from Nagel's concluding remark:

> "A theory for the evaluation of image sequences will have to offer a rather wide spectrum of concepts and descriptive tools in order to cope with available input data. In view of this consideration, it appears questionable whether we know already enough to sketch the structure of "intelligent systems" which should be capable of analyzing image sequences. Such schemas are useful because they attempt to chart a vast area and thus provide a preliminary framework within which detailed research results can be assessed. It is only argued here that one should be careful with any explicit or implicit claims about presenting the definite structure. The discrepancy between the information contained in image sequences and the fraction which we know to extract automatically is still too great".

From the above review of Nagel's survey, one can realize the huge gap between current experience, the capability of the present techniques, and the amount of information which is contained in and can be extracted from image sequences. However, the development of general models rooted in a knowledge-based system is more promising. Such

a system is referred to by Scacchi [Scacchi, 79] as an "intelligent system", which is the subject of our discussion in the next section.

### 2.2.2.4 [Scacchi, 79]

The review of [Scacchi, 79] is different from both that of [Martin and Aggarwall 77;78] and [Nagel, 78;79], in that it is not survey-oriented, but is mainly concerned with the structure of an "intelligent" system for visual motion perception, analysis, and understanding.

Scacchi has built his discussion on the literature applicable to visual motion perception reviewed from topics which include scene analysis, hardware-based vision systems, computer animation, artificial intelligence, and human motion perception. From the review of this literature, he has attempted to define the attributes and internal structure of an intelligent system which would simulate (model) human visual motion perception. His study led to the conclusion that the solution to achieving this objective is implicit in the design of a knowledge-based computer vision system.

Thus, Scacchi's study concerns two related subjects, visual motion analysis (image sequence analysis), and the structure of a knowledge-based system capable of performing this analysis. Both subjects are the main concern of our current research. Therefore, Scacchi's report will be

reviewed in this section, from a motion analysis point of view, and in Section 2.4 it will be discussed as a reference for a knowledge-based system structure.

Scacchi's brief review of the work in dynamic scene analysis does not differ from our discussion of the same subject in the preceding sections. However, an exception, is his pointing out the impracticality of using symbolic descriptions. This is attributed to the computational difficulties associated with real-time analysis. He describes these difficulties as follows: ..."the generation and implied processing of high-level descriptions of objects is impractical for each image arriving at real-time frame rates". From our point of view, we do not agree with Scacchi on this aspect, since the problem does not represent a theoretical difficulty. This is essentially a technological problem which could be solved either by parallel computer processing, or by a multilevel analysis. An alternative possibility (implemented in our system), is a low-level process, concerned with feature extraction and measurements, which may be easily performed in real time. The analysis and symbolic descriptions may later be computed by a higher level stage of the system (see Chapter 3).

Discussing computer animation techniques, Scacchi demonstrates that most of these are complementary to those in dynamic scene analysis. Both are based on the use of internal image descriptions and representational organization. In dynamic scene analysis, the system

transforms motion into descriptions. Whereas, in computer animation, the system translates the descriptions into motion. One of the most common techniques used in the production of animated films is "key frame" interpolation [Burtynk and Wein, 78]. It is worth mentioning here that some steps in our approach to the extraction of the changes in static and incremental data, are similar to this technique.

According to Scacchi, visual motion analysis by an intelligent system requires different analysis strategies depending upon the viewing situation. He defined three different viewing situations according to the "motion vantage perspective": (a) stationary observer and moving objects, (b) moving observer viewing stationary objects, and (c) moving observer viewing moving objects. Based on these, he discussed the knowledge and control requirements for a visual system (see Section 2.4).

An intelligent vision system should exploit the visual knowledge embedded in a coherent image sequence, the so-called "object motion coherence". The latter is defined in [Scacchi, 79] as a low-level property of visual motion knowledge. This knowledge is similar to that which humans use when viewing an image sequence; that is, if an object is recognized at a given moment (frame of the sequence), it is still the same object in sight until the scene changes (in a later frame). Thus, an intelligent system should rely more on high-order knowledge, description, representations,

and processing to recognize the key frames, those in which object features change unexpectedly. We observe that a high-level of recognition should be supported by such a low-level processing procedure. Thus, when new object features appear, or when the existing features change, a key frame is initiated (see Section 7.2.1 for more detail on this methodology).

In this section, we have briefly reviewed the study of Scacchi in visual motion analysis, and his view of the function of an intelligent system for that purpose. The remainder of his report discusses the basic requirements for constructing such an intelligent system, to be reviewed in Section 2.4. However, in conclusion, Scacchi suggests that a system for visual motion analysis and understanding must be organized around descriptive multi-level knowledge sources, whose interactions are directed by continually emergent, distributed control processes.

Certain recent empirical approaches which follow this new trend and/or are related to our research, have been selected for discussion in the following sections.

## 2.2.3 Review Of Relevant Work

### 2.2.3.1 [Yachida Et Al., 78]

Yachida et al. [Yachida et al., 78] presented a system that detects and tracks live moving objects. They observed fishes swimming in a vat, in order to study their behaviour under a variety of stimuli, such as lights or tones. A sequence of 20-250 frames recorded for periods of between 2-30 seconds (8 frames per second) were obtained through either video tape (connected to an overhead camera) or cine film. Yachida et al. addressed three main problems to be solved by their system: (a) developing an efficient procedure to process a large number of frames, (b) to solve the difficulty in boundary detection due to blurred images, and (c) to solve the occlusion problem.

They successfully designed a sophisticated image sequences analysis system which has the following features: (a) identification of the moving objects from images which are often motion-blurred, (b) using the analysis results of previous frames to guide a feature extraction process in a current frame, (c) employing shape prediction in order to disambiguate situations where one object occludes another, and (d) using the results of later frames, to reanalyze previous frames, where uncertainty existed.

In general, the system of Yachida et al. is an efficient one for motion detection and tracking moving objects. However, their work does not include global motion analysis, or shape change detection, quantification, and analysis; these aspects are considered in our present work.

### 2.2.3.2 [Price And Reddy, 77]

As discussed previously in Section 2.1.2.2 regarding change detection, Price and Reddy [Price and Reddy, 77] describe a technique for symbolic registration and change analysis.

The two images to be compared are segmented partially or completely using a region splitting algorithm. The segmented regions are described by features including size, intensity, location, circularity (perimeter squared divided by area), orientation, elongation (length-to-width ratio), as well as a combination of these features, and relations between a region and its neighbors. The feature based description of the segmented image constitutes the symbolic representation of the image. A similarity search is performed by comparing the symbolic representation of the two images in order to determine the corresponding regions in the two images. These results are used to analyze changes in the corresponding regions (dissimilarity grading) which occurred between the two images.

It is important to note that the work of Price and Reddy provides explicit means for introducing domain and task knowledge to each stage of the processing. For example, in the similarity search, each feature difference is multiplied by a strength factor which could be chosen by the user to reflect specific knowledge on the relevance of this feature for the current domain, task, or state of search. This methodology for introducing outside knowledge and task description, is a step towards the development of a more general system, rather than just solving a specific problem. However, the use of a general analysis system introduces the problem of specifying and incorporating task knowledge, which is not encountered in a special purpose system.

In conclusion, the research of Price and Reddy represents an initial effort towards the development of a general system for symbolic change detection and analysis. Although they concentrated their work on the symbolic description of the change between two aerial or satellite images, their results provided important incentives to adapting them for use in image sequence analysis.

Our current work, concerned with incremental change detection and analysis (an intermediate stage between the static and global analysis), is to some extent similar to the work of Price and Reddy. This similarity rests on the use of symbolic descriptions and analysis. Also, the knowledge and task description are expressed explicitly for

each stage of the processing. However, in their report, they neither specify the type of symbolic descriptors they employ, nor the method of quantifying the changes symbolically. These aspects are addressed in our current work (see Section 4.6).

### 2.2.3.3 [Tsotsos, 76]

An extensive and significant work on motion detection, representation, understanding and description has been carried out over the last few years by Tsotsos at the University of Toronto. In an early work by Tsotsos [Tsotsos, 76], he describes a scheme for recognizing the motion of an object from a sequence of images in order to describe them symbolically. This work has its roots in the research of Miller [Miller, 72] and Badler [Badler, 75].

Miller has analyzed the English motion verbs and directional prepositions. He provides a classification of English motion verbs using a hierarchy of primitive motions. Badler was the first to use such symbolic components as descriptors in temporal scene analysis, although he did not work with real images. He considered image sequences when attempting to provide English descriptions of object movements. Badler's work provides precise definitions for directionals and adverbs, and he also outlines ideas on the representation of the semantic components of the verb, as defined by Miller.

Based on Miller's linguistic analysis and Badler's symbolic representation of motion, the main objective of Tsotsos' work [Tsotsos, 76] was first to find representations for the concepts associated with motion. Secondly, he outlined an appropriate structure and implementation of the system. The motion terms used in this description are verbs, directionals, and adverbs. These were built upon lower level concepts such as trajectories, locations and velocities. They are defined in hierarchial levels of description, starting with the changing object location and ending with the motion verbs. In this early work of Tsotsos, the type of motion to be recognized and described by the system is restricted to translation.

The representation and description of motion concepts in Tsotsos, 76 was the basis of the development of a general framework for the abstraction and understanding of motion concepts from a sequence of images [Tsotsos, 80].

The main goal of Tsotsos' framework is to find suitable representations for motion descriptions, at different levels of abstraction, and appropriate control structure strategies for using these levels for recognition. This framework is based on the assumption that the design of an expert system for the performance of certain tasks requires the representation and use of knowledge relevant to that task. Based on this assumption, Motion Description Formalism (MDF) has been defined as a domain expert which can create a knowledge-base of motion concepts which apply to a

particular problem domain.

Tsotsos has defined two main difficulties with his specific application, the analysis of cinecardioangiograms: (a) the huge number of images to be analyzed, (b) the poor quality of the individual images due to X-ray dosage limitations. Also, he has defined and classified the important aspects of motion understanding as follows :

(1) Computer Vision : (a) image segmentation and object recognition (b) object description (c) motion detection (d) motion tracking (e) interimage movement description.

(2) Representation of Knowledge : (a) general temporal concept representation (b) problem domain motion concept representation (c) recognition biased knowledge organization.

(3) Recognition Control Structure : (a) integration of descriptive and visual concepts (b) change and focus of attention mechanisms (c) temporal segmentation (d) disambiguation due to object occlusion (e) goodness-of-fit measures (f) generation of low-level guidance (g) scene sampling rate considerations (h) artifactual motion handling, i.e., "temporal noise" (i) generation of sequence spanning descriptions.

The motion concepts are represented in a hierarchial structure. Each level down the hierarchy provides a more detailed form of description for the motion concept, spanning all the levels between the most abstract motion terms to the picture elements in the image. In this hierarchy, each motion concept is represented within a "frame". Each frame has an arbitrary number of "slots" that form its parts. Each slot has an associated "type" that refers to another frame, thus defining a "PART-OF" hierarchy of description.

From the above review of Tsotsos' work, we may conclude that based on the liguistic analysis of motion verbs by Miller, and the symbolic description of movement of an object by Badler, Tsotsos was the first to develop a methodology for representing and describing motion concepts from a sequence of images. Thus, he has achieved a significant advance toward the development of a general framework for motion understanding, based on knowledge representation and a symbolic analysis and description of the semantic motion concepts. This framework is being tested through an ongoing project called ALVEN ( A Left Ventricular Wall Motion Analysis Consultant). The objective of ALVEN is to analyze films of the human left ventricle in order to generate a conceptual description of the shapes and motion exhibited by the left ventricular wall, noting abnormalities and unusual occurances.

In spite of the major differences in the technicalities of Tsotsos' work, and the work developed in our current research, theoretically there are two similarities in both approaches: First, both are based on a knowledge-based scheme. Secondly, the extraction and symbolic description of the global changes are common objectives.

The differences lie in the following factors:

(1) Data Structure: In Tsotsos' model, knowledge is represented through a hierarchial data structure consisting of frames and slots, whereas our model structure is a rule-based system.

(2) Control Structure: In Tsotsos' framework, the control structure is based on hypothesis and prediction, which depends on the existing knowledge of the model (model-driven). Our control structure is based on condition-action rules, which depend on the occurence of observed events (data-driven).

(3) Application Domain: Tsotsos' framework is restricted to certain problem domains in which motion concepts are definable, such as human gait patterns or heart wall motion. These restrictions do not apply to our system. Thus, the knowledge pertaining to the class of scenes under analysis is represented in the LTM as a set of local (primitive) constraints related to the physical properties of the object and their motion capabilities. Examples are maximum, minimum, or average dimensions of the object in

pixels, and maximum possible displacement during a given time period. Moreover, the constraint knowledge may be generated by the system using a training set of data (see Section 3.3). The representational rules use these constraints and the STM dynamic data (generated from an analysis of the input sequence of images) to generate the motion description.

A major problem which has been ignored by all previous work, including Tsotsos, is the recognition, quantification, and description of the structural changes of a moving object. Tsotsos [Tsotsos, 76] has commented on this problem as follows: "A problem which Badler and others (including myself) seem to ignore in their designs, but which became apparent on consideration of several examples, is the recognition and description of object construction.". In our current work, we analyze the dynamic changes of a non-rigid moving object. Consequently, the problem of recognition and description of the object construction changes has been studied in detail.

## 2.2.4 Summary

In the preceding sections, we have briefly reviewed the progress in the analysis of a series of two-dimensional digital images representing the variation in a specific scene along a third-dimension, that is, time. The previous techniques and approaches for this analysis have been presented in the literature under three titles: Dynamic

Scene Analysis, Image Sequence Analysis, and Visual Motion Perception/Understanding. First, the title "Dynamic Scene Analysis" was used to address the initial survey in this area of study by Martin and Aggarwal [Martin and Aggarwal, 78]. Nagel [Nagel, 78], in order to generalize the field to include spatial image sequences as well, published the second survey using the title "Analysis of Image Sequences". The third title "Visual Motion Perception/Understanding" indicates the recent trend in the current research in this area of study, which seeks to develop a general "inteligent" system for the understanding and description of visual motion.

The first survey in image sequence analysis was presented by Martin and Aggarwal [Martin and Aggarwall, 78]. In this review, they first discuss the perception of motion in a biological system. Second, in terms of the biological system structure, they divide the work in dynamic scene analysis into two main classes, motion detection (peripheral) and motion analysis (attentive). These were initially applied to the automated detection and measurement of cloud motion from satellite pictures. Most of the resulting computer programs used cross-correlation and image differencing techniques. In motion analysis, the work was based on two techniques, centroid matching and shape analysis.

Further significant progress in image sequence analysis was carried out by Nagel. Besides his own research, he has presented the two most extensive surveys in image sequence analysis. In Nagel's first survey [Nagel, 78], he first reviewed the different experiences in image sequence analysis, discussed according to specific application areas. Second, he made a very thorough analysis of the different comparison techniques, the so-called "interframe comparison". In this study, he listed six categories within which the different comparison approaches may be organized: (a) no interframe comparison, (b) indirect interframe comparison, (c) dissimilarity grading, (d) similarity search, followed by dissimilarity grading, (e) dissimilarity grading, followed by similarity search, (f) similarity search. Furthermore, he discussed aspects, such as segmentation and velocity determination of moving objects, in terms of interframe coding.

The second survey by Nagel [Nagel, 79] is a rather application-oriented review. He cited more than five hundred articles, gathered from diverse applications. Throughout this review, attention has been drawn to the interaction between the evaluation of image sequences and the importance of quantitative models in describing complex phenomena in the application domain. The parallels between different approaches in different applications is demonstrated. This investigation might facilitate the transfer of experience between different applications. Of

particular interest in this survey is that it shows the wide gap between the amount of information current techniques can extract from an image sequence, and the information potential of the sequence itself. Also, it shows that present experience is not enough to achieve what we can call an "inteligent" system which can simulate human "visual motion perception".

In general, most of the previous research has attempted to analyze an image sequences by considering the multitudinous data representing the movements or changes that occur between each two sequential frames from the sequence. This incremental data may be generated by several techniques of comparison. This approach to image sequence analysis has resulted in the development of sophisticated computer systems for motion detection, recognition, and tracking and has yielded an enormous number of applications. Motion understanding and description have been ignored by most of the past research. To achieve the objectives of motion understanding and description, it is not enough to merely determine the incremental movements or changes that occur between consecutive images [Tsotsos et al., 80]. What is required is a system which abstracts a summary description (in natural language) of the global motion characteristics from the multitude of static and incremental data. Development of such a system represents the new direction being taken in the current research in image sequence analysis.

This recent trend in current research is directed towards the development of a system which has the capability of motion detection, understanding, and description in a global manner similar to human visual motion perception. The first step in this direction was taken by Badler; his achievement was to generate a symbolic description for motion concepts. Also, Price and Reddy attempted to use symbolic descriptors and analysis to describe the changes between two different images of the same scene. Yachida et al. [Yachida et al., 78] used high-level global analysis to improve the low-level processing of ambiguous situations in individual frames. Based on Badler's work, Tsotsos then developed a system for motion detection and symbolic description. Recently, Tsotsos has introduced a framework for a general system for motion understanding and description using predefined motion patterns.

Most of the recent approaches are based on modelling the dynamic environment of the motion using knowledge-based systems. However, there are different views pertaining to model construction, data and knowledge representation, and control structure strategies.

In our current work, we have developed a model for a general image sequence analysis system consisting of three basic entities:

(a)Dynamic data: The data which continually changes during the analysis (input, output, and any computational results). These data are stored in a Short Term Memory (STM).

(b)Static (constant) data: The data which remains unchanged during the analysis (description of the class of scenes under analysis and the control information describing the pertinent computational processes. These are stored in a Long Term Memory (LTM).

(c)A collection of analysis processors, each of which is assigned to a particular task.

Both STM and LTM are implemented as a relational database. The STM is designed to work as a communication channel for all of the processes. It contains a record of the instantaneous object motion, shape, and structural changes, as well as, the current global description of the object behaviour. The LTM contains the general model of the morphology of the objects under analysis, as well as control information describing the pertinent computational processes. The different processors are activated throughout three hierarchial stages: static, incremental, and global analysis. They interact through the STM using the information stored in the LTM, until a complete description of the dynamic object motion and morphology is obtained.

In the preceding section we have briefly compared our work to the most relevant recent approach by Tsotsos. From this comparison and the above review, we may claim that our work provides an essential and original contribution to the research in image sequence analysis in two basic ways: First, the construction of our model is a rule-based structure (knowledge representation and control strategy). Within this structure, the dynamic behaviour of the moving object is described using generic knowledge (constraints) and rules; for example, the exact motion pattern of the class of scene under analysis is not defined. Consequently, the system has a much wider application, especially for those sequences containing moving objects whose motion patterns are not known a priori, or which exhibit random motion. Second, in our work we analyze, quantify, and symbolically describe the structural changes of non-rigid moving objects hitherto neglected in all the previous work done in image sequence analysis.

## 2.3 SHAPE ANALYSIS AND DESCRIPTION

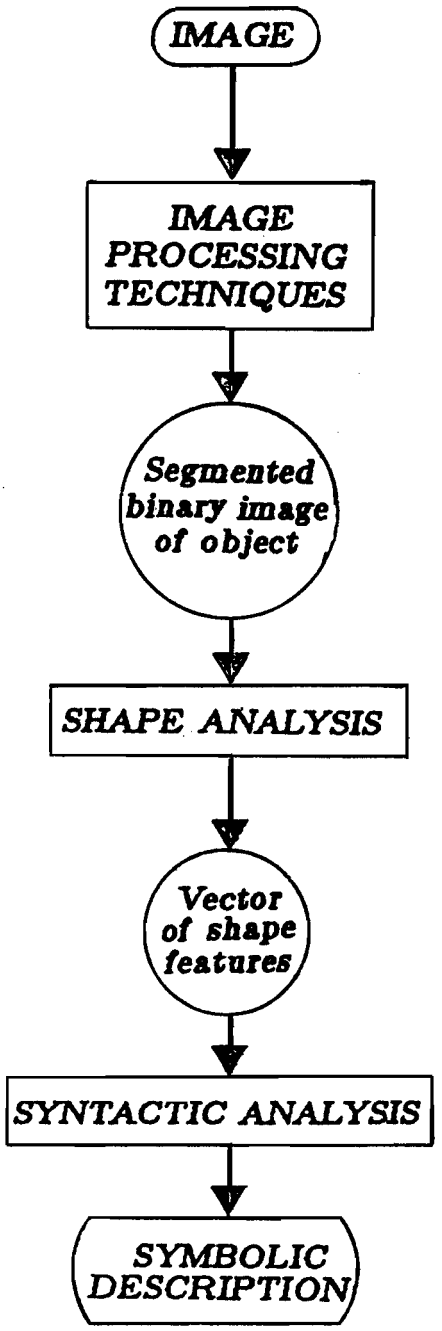
### 2.3.1 Introduction

The problem of shape descrimination is a central one to pattern recognition and as such has received considerable attention in most papers dealing with recognition of characters, waveforms, chromosomes, cells, machine parts, among other applications. This area of study is taking on increasing importance but much work still remains to be

done. Furthermore, shape perception is a common problem in any computer vision, scene analysis, or pattern recognition system. The solution to this problem may be achieved through two stages of processing: shape analysis, and shape description. Figure (2.1) is a schematic diagram which shows the basic steps for shape analysis and description, as well as the input and output data at each step.

Shape analysis: In shape analysis, a digitized image of an object is transformed into a scalar vector whose elements are measurements of some of the shape properties (features or shape descriptors), eg., length, width, elongation, circularity, Fourier descriptors, moments, and other shape features. The second task of shape analysis is to transform the image of an object into a graph. The properties of this graph express the shape and structural properties of the object.

Shape description: Shape description represents the higher level process of shape perception by computer. In this process the scalar vector or graph, the resulting form of the shape analysis, is analyzed by a syntactic analysis methodology in order to generate a text in a natural language (symbolic description). It contains all the relevant information pertaining to the shape of the object.




*Figure(2.1) The basic processes for shape analysis and description.*

Early attention to this area of study is related to [Attneave, 54], which shows that shape analysis has roots in psychology as well as computer vision. In Attneave's work, he discussed human visual perception from an information theory point of view. Of particular interest in his findings is that shape information is concentrated along contours, especially at those points at which its direction changes rapidly (currently called "critical points").

A brief review of current shape analysis and description techniques is presented in this section. The review in this section is restricted to the techniques which analyze two-dimensional objects in a plane. The literature is reviewed in the following order: First, the focus of attention is directed to the most recent surveys in this area by [Meagher, 79; Pavlidis, 81]. Second, the shape analysis techniques which have been used or are directly related to our current work are reviewed under topics which include: (a) curve representation and critical point detection, (b) curve and polygonal approximation, and (c) shape decomposition. Then, more sophisticated shape analysis techniques are briefly reviewed in Sections 2.1.6-2.1.9 inclusive. These include moments, Fourier transforms, thinning and integral geometry, and relaxation. The syntactic analysis and shape description techniques are reviewed in Section 2.1.10. Finally, in the last section 2.1.11, our current work in shape analysis and description is summarized and compared to the previous ones.

## 2.3.2 Recent Surveys In Shape Analysis And Description

The recent survey of Meagher [Meagher 79] presents a brief overview of the literature pertaining to techniques for shape analysis and description. This survey begins with a general short overview of the field. He then considers papers pertaining to curve representation and proceeds to more sophisticated techniques. He reviews the literature under topics which include chain code methodology, polygonal approximation, syntactic analysis, relaxation, Fourier descriptors, and moments. Meagher's survey is a basic review for a neophyte reader as a pointer to the significant work in the field. However, the survey by Pavlidis [Pavlidis, 81], (the last updated version on a series of reviews by the same author), is not only a complete well-documented review, but it may also be considered as a comprehensive study in shape analysis and description.

In the recent review by Pavlidis [Pavlidis, 81], the methodologies used in shape discrimination have been classified according to several criteria. First, he defines "external" and "internal" to refer to local boundary followers and global boundary or area examiners, respectively. Second, he makes another distinction on the basis of "scalar transform" and "space domain" techniques as to whether the process transforms the picture into an array of scalar features or into another picture, as is the case in the medial axis transformation technique. Finally, he defines "information preserving" and "information

non-preserving" techniques, depending on whether or not it is possible to reconstruct the picture from the shape description.

Pavlidis uses the first two criteria to form four categories in order to classify the different techniques of shape analysis as follows:

(1) Internal Scalar Transform

(a) moments and moment invariants,

(b) two-dimensional Fourier Transform (FT) of the binary coded image,

(c) binary masks,

(2) External Scalar Transform

(a) Fourier Transform of boundary (eg., FT of tangent angle versus arc length),

(3) Internal Space Domain

(a) medial axis transformation (MAT)

(b) various thinning algorithms

(c) various integral geometry schemes

(d) techniques using the concepts of convexity and concavity (eg., decomposition into primary convex subsets)

(4) External Space Domain

(a) syntactic description techniques.

Most of the publications on shape have dealt with "information non-preserving" techniques. In particular they have emphasized properties such as symmetry, elongation, angularity, etc. [Kaufman, 67; Kolers, 70; and Langridge, 72]. Such properties give useful information about the shape of simple objects but fail to do so for complicated ones which must necessarily be given in terms of local characteristics of primitive subparts [Rosenfeld, 76].

The different techniques for shape analysis and description will be reviewed in the following sections in groups according to the methodologies defined in Section 2.3.1.

## 2.3.3 Curve Representation And Critical Point Detection

The Freeman chain code may be considered one of the earliest and most famous schemes for representing information pertaining to curve or contour of a digital image. The chain code may also be used for further shape analysis or description. The method of encoding an arbitrary geometric curve via the use of the chain code is presented in [Freeman, 61]. This scheme is further developed in [Freeman, 74; 77; Freeman and Davis, 77], for finding the critical points (corners, maxima curvature, inflection, discontinuities in curvature, etc.) of a curve.

These critical points were used originally for processing line drawings. This method has been used widely, since, by many authors using different techniques for general shape analysis and description. An advanced application of techniques based on chain codes to object recognition is presented in McKee and Aggarwal, 77.

Detecting the critical points from the boundary of a curve or the contour of an object has been the main concern in many publications in this field. From these publications, the work of [Rosenberg, 72] is noteworthy in determining the dominant points of a convex blob. In addition, an algorithm was presented by [Rosenfeld and Johnston, 73] for selecting the curvature maxima. Also the work of [Davis, 77] in shape understanding was carried out mainly for detecting such critical points as the definition of angles and sides of a curve.

In our current application, the contour (boundary points) of an arbitrary shape is represented by number of sides and vertices obtained by a polygonal approximation procedure (a description of these techniques is given below). Then the critical points are extracted at those vertices which exhibit special convexity (see Section 4.3).

## 2.3.4 Curve And Polygonal Approximation

Polygonal approximation is a known technique in shape analysis. In this technique a curve or contour is represented by fitted straight lines. The input to such algorithms can be either the boundary points directly, or their chain code. The output is a list of vertices of the fitted lines. These techniques have the advantage of reducing the noise as well as the amount of data to be manipulated by higher level stages of the system (the number of the vertices is always less than the boundary points). The mathematical aspects of these techniques may be found in [Pavlidis, 77]. In some applications the fitted lines or polygons may be used directly for shape recognition or description, while in others they are an intermediate form of data. For example, in the introductory report for a general dynamic scene analysis system applicable for the characterization of the dynamic behaviour of cell motion by [Levine and Youssef, 80], the output of the polygonal approximation represents the input for the shape decomposition. The latter is further analyzed by higher level processes of the system (incremental and global analysis).

Some schemes for constructing polygonal approximation are concerned with selecting the vertices from the boundary points so as to generate the best fitted polygon. Others may allow the vertices to leave the curve itself if they generate a better fitted polygon. One of the early and most

efficient techniques by Ramer, selected the polygon vertices from the boundary points [Ramer, 72]. The resulting polygons of Ramer's algorithm are not necessarily optimum (minimum number of vertices), but his algorithm is computationally much more efficient than those generating optimum ones. An algorithm for polygonal approximation based on Ramer's scheme is implemented in our current work; therefore, a detailed description of this technique is presented in Section 4.3.

In some schemes for constructing polygonal approximation, the minimum distance (not to exceed a specific threshold) between the segment of the boundary and the fitted line is used as a criterion for selecting the best fitted approximation line (eg. Ramer 72); in others, the fitted line is chosen so as to minimize the area difference between the approximation line and the original curve, eg., [McClure, 77].

Other techniques for extracting the polygonal approximation are based on the chain-encoding of the boundary points. Montanari [Montanari, 70] presented an algorithm for determining the chain code of a contour. Then, by a smoothing operation on the chain code, a minimum perimeter approximating polygon may be generated.

Different from previous work, Pavlidis [Pavlidis, 73] introduced an algorithm for segmenting a waveform in order to generate a piecewise linear approximation. This

algorithm is modified in [Pavlidis and Horowitz, 74] to introduce a split-and-merge algorithm which improves the previous one, in that it is faster and does not require an initial segmentation. This algorithm is currently one of the most popular for curve segmentation, and its application is presented in many papers by different authors. For example, these methods are applied to the recognition of handwritten numbers in [Pavlidis and Ali 75].

## 2.3.5 Shape Decomposition

Another set of shape analysis techniques is based on decomposition of complex shapes into simpler ones. These methodologies are prime examples of structural pattern recognition and shape analysis. They are based on the assumption that shape perception is a hierarchical process [Pavlidis, 68;72]. In these techniques the original figure is expressed as the union of some of its subsets (primitive components). The shape of the latter may be simpler, and therefore, some of the less complex descriptions may be applicable. Most of the subsequent schemes emphasize the concept of convexity and assume polygonal approximation of the original object. One of these requires the decomposition of the object into primary convex subsets [Pavlidis, 68;72], giving an output which can be expressed through a juxtaposition graph [Pavlidis, 72a;72b; and Feng and Pavlidis, 75].

A decomposition technique based on a graph-theoretic clustering method is developed by [Shapiro and Haralick, 79] to transform a two-dimensional shape into a binary relation whose clusters represent the simple parts of the shape. This method first determines dense regions, which are local regions of high compactness and then forms clusters by merging those dense regions having a high enough overlap. Maruyama [Maruyama, 72], suggested a decomposition of shapes into angularly simple regions. Each angularly simple region has at least one interior point which can "see" its entire boundary. Schachter [Schachter, 78] presented a method for decomposing polygons into convex sets, based upon a Delaunay tessellation of the polygon. It is implemented as a divide-and-conquer technique.

With these decomposition methods, the shape of an object is represented as a graph. The results have a number of desirable features which are also shared to some extent by the MAT :

(a) They are translation and rotation invariant and insensitive to registration.

(b) To a large extent they are size invariant.

(c) They usually produce an "anthropomorphic" description.

(d) THey give data structures which are particularly appropriate for syntactic or structured pattern recognition.

The main disadvantage of this methodology is that the programs required to implement it tend to be quite complex. In our system the cell shape is decomposed into its primative parts at the convex angles, in a fashion similar to [Feng and Pavlidis, 75]. However, in our algorithm the decomposition procedure is governed by the rules dependent on the physical structure of the cell (see Section 4.4).

## 2.3.6 Moments And Moment Invariant Techniques

In the category of "internal" scalar transform techniques, the method of moments was the earliest one used [Alt, 62; Giuliano et al., 61; Hannah, 71; Ledley, 64]. In these techniques the moments of a digitized object in a plane are defined as in geometry and mechanics as follows:

$$m(u,v) = \mathrm{SUM}\ (\ X^{u}\ Y^{v}\ ) \qquad u,v = 0,1,2,\ldots$$

where m = is the moment,

X,Y = the different points of the object.

It can be shown that:

m(0,0) = the area of the object,

m(1,0)/m(0,0) = the X coordinate of the center of gravity,

m(0,1)/m(0,0) = the Y coordinate of the center of gravity.

Higher order moments and linear combinations of moments ("moment invariants") possess various invariant characteristics under a number of object transformations. An early work using moments for pattern recognition is presented by Alt [Alt, 62]. He showed that moments of order

5 or below are sufficient to discriminate between 35 printed characters, whereas moments of higher order are found to be increasingly sensitive to noise. Another early work in pattern recognition which was based on moments was [Giuliano et al., 61]. Ledley [Ledley, 64] has used the moments methodology for shape analysis in biomedical pictures. A more recent application of moments to aircraft identification is presented in [Dudani et al., 77].

## 2.3.7 Fourier Transform (FT) Techniques

Other significant approaches to shape description compute the two-dimensional Fourier transform (FT) of the characteristic function of the object or use binary masks to extract features conveying the shape information [Nagy, 64]. We note that the one-dimensional FT of the boundary can be profitably assigned as an "external" scalar transform technique [Zahn and Roskies, 72; and Rosenfeld and Kak, 76]. Due to the fact that most of the shape information of an object is concentrated along its contour, the one-dimensional FT has been the concern of many researchers. In this methodology, the Fourier transform of the boundary is computed, and the resulting coefficients are used as features (shape descriptors) for shape discrimination.

An early work in shape analysis using FT was presented by Borel [Borel, 65]. He used the tangent angle versus arc length to detect the curve segment of maximum curvature. Then a matching (using cross correlation) between the

section of maximum curvature of an unknown contour and known shape is performed. Systems and algorithms for shape analysis which are based on FT schemes involve many applications. Examples are handprinted character recognition [Granlund, 72] and aircraft identification [Richard and Hemami, 74].

A reference which contains a considerable amount of information on Fourier descriptors is [Zahn and Roskies, 72]. In a more recent study by Persoon and Fu [Persoon and Fu, 77], the work described in [Zahn and Roskies, 72] and [Granlund, 72] is extended. The paper also includes a review and general discussion of the subject, as well as experimental results.

## 2.3.8 Thinning Algorithms And Integral Geometry

The Medial Axis Transformation (MAT) or skeleton was the earliest and most widely studied method among the "internal" space domain techniques [Montanari, 69; Mott-Smith, 70; Philbrick, 66; Rosenfeld and Weszka 76]. The skeleton may be used to derive information on the shape of the original figure, but its computation can be quite time-consuming and very sensitive to noise [Rosenfeld and Weszka, 76]. These difficulties may be reduced by first obtaining a polygonal approximation of the original contour.

A number of techniques related to integral geometry have been proposed by various authors [Klinger et al. 71; Nakimoto et al., 73; Pavlidis, 68; Rutovitz, 70; Spinrad, 65; Wong and Steppe, 69]. In these techniques, the object is intersected by a number of chords whose length statistics can be used for shape description. For example, Rutovitz [Rutovitz, 70] has used radial chords, all passing through a common point, to describe the shape of chromosomes.

## 2.3.9 Relaxation

Other methodologies which may be used in shape analysis and interpretation are based on relaxation techniques. They use the context of a scene or section of the scene to reduce the ambiguity in the labeling of a set of objects or subparts of an object. For example, in an indoor scene interpretation system, the fact that a roof is always above the walls, or in a face recognition system, a nose is always spatially above a mouth, could be used to eliminate some erroneous labeling attempts. Four models of relaxation processes are presented in [Rosenfeld et al. 76]. The authors showed how these models are used to reduce the uncertainity in a situation before processing by a semantics-based analyzer. Further, they demonstrated that by using contextual information, the more powerful forms of relaxation may be used to adjust the probabilities of labels assigned to parts of an object.

## 2.3.10 Syntactic Analysis And Shape Description

The main goal of shape perception either by human or computer is to translate pictorial data of an object into symbolic descriptions containing the relevant information pertaining to the object. This goal may be achieved through two processes: shape analysis and shape description. In the above sections we reviewed different techniques for shape analysis. In this section, we will discuss shape description, and briefly review the limited work which has been done in this subject.

Shape analysis processes produce information related to the shape of an object in the form of scalar vectors or a graph. The main objective of shape description is to analyze this data using a syntactic methodology in order to generate a symbolic description of the object.

To describe the shape of an object, the scalar vectors are not very helpful unless the features of their components have well-defined physical meanings. Moreover, in the case where the shape of the object is complex, the description cannot be accomplished in terms of such scalar measures unless some simplifying transformation is used first. The techniques which achieve this are those which transform the object into a graph, so that its shape properties can be expressed through the properties of that graph. Such a transformation can be performed by one of two basic techniques. The first is thinning, where the object is

reduced into a line drawing graph (a skeleton), eg., MAT. The second is the decomposition methodology, (both types of techniques are reviewed in the above sections). The thinning techniques are more appropriate for the description of filamentary-like shapes, whereas the decomposition techniques are applicable to any types of shapes.

In order to generate a symbolic description of an object from its graph, it is necessary to develop a "graph language" and "graph grammar". Then, a given graph could be parsed according to this grammer to generate sentences in a natural language describing the shape.

Although the work done in this area is rather limited, Pavlidis [Pavlidis, 75; 76; 77] provides a considerable amount of information on this subject. He demonstrates the results of syntactic analysis in various applications. Examples are: Chinese character recognition and handwritten numerals. A theoretical discussion of the advantages and disadvantages of various syntactic techniques in shape description is presented in [Pavlidis, 77]. The recent work of Shapiro [Shapiro, 80] is also an example of syntactic analysis. First, she used the method described in [Shapiro and Haralick, 79] to decompose a shape into a set of its primitives. Then, using these primitives, their properties, and their interrelationships, a matching procedure to find mappings from a prototype shape to a candidate shape is performed. Her model gives a favorable result on hand-printed character data.

One of the few systems which has used a symbolic description from everyday natural language is the work of [Hollerback, 75]. He developed an approach towards shape description, based on prototype modification and generalized cylinders. The emphasis throughout his work has been to develop useful, qualitative descriptions which bring out the significant features of pottery and polyhedra.

### 2.3.11 Summary And Review Of Our Current Work

The perception of shape plays a prominent role in both human and computer vision. Shape perception by computer may be achieved through two stages of processing, shape analysis and shape description. Algorithms for shape analysis have been briefly reviewed and classified under two categories: whether they examine only the boundary or the whole area, and whether they describe the original pictures in terms of scalar measurements or through structural descriptions. Most studies of shape and pattern recognition are based on global feature measurements which then constitute a feature vector used for the shape representation.

In spite of the difficulty of addressing the general problem of shape description, the solution is more promising with the use of syntactic analysis. Therefore, more recently, there has been interest in syntactic pattern recognition techniques which analyze patterns by a parsing process of hierarchial decomposition. The advantages of such an approach suggest that it might be appropriate to

study hierarchial shape representation in more detail as a vehicle for cell shape description, as well as the global structural and membrane shape changes which occur during locomotion.

In our current research, besides the general difficulties of describing an arbitrary shape in a specific image, we are facing the following problems:

(a) Estimating the incremental change between two different images in the shape and structure of a non-rigid moving object.

(b) Detecting and characterizing the global structural changes in the morphology of a non-rigid moving object over a period of time from a sequence of pictures.

(c) Presenting all the above descriptions in a meaningful terminology to the user.

We have developed the procedures which produce a meaningful symbolic description of the shape and its changes. Also, we have developed a mathematical expression for measuring the complexity of an arbitrary shape pattern. This expression is based on a group of selected shape properties which are independent of translation, rotation, or scaling. Another shape feature is introduced through our work in shape analysis, to measure the degree of curvature regularity (angle and/or side regularity) of the shape of an object. This feature is shown experimentally to play a

valuable role in shape descrimination. This study in shape analysis is demonstrated by describing the membrane shape and quantifying its observable changes.

## 2.4 COMPUTER VISION KNOWLEDGE-BASED SYSTEMS

### 2.4.1 Introduction

Any vision system consists of two basic hierarchical processes: a low level, which is concerned with data extraction from the perceived scene, and a high level, which is concerned with interpretation and description of that which constitutes the scene. To accomplish this, the system utilizes information from two sources: data from the perceived scene, and the knowledge and expectations (perceptual set) of the observer. Thus, the development of computer vision systems has become a study at the intersection of the neighboring disciplines of image processing, scene analysis, pattern recognition, artificial intelligence, and cognitive psychology.

One of our main concerns in this research is the development of a computer vision system for understanding and describing the visual motion of non-rigid moving objects. The objective of this section is to briefly review the significant work in this field as a gained experience. However, we cannot discuss visual motion understanding without first considering its roots in static scene interpretation and description.

In the past, most computer vision research has dealt with the low level processes associated with this problem. As a result, considerable experience has been gained on how to segment a simple digital image into regions that correspond to objects as preceived by a human observer. However, this stage of progress has not been accomplished yet for complex scene analysis. In complex scene analysis, the low level segmentation results in a larger number of regions than those which can be perceived by a human observer. In order to achieve a meaningful partition of the image under analysis, the system should utilize external knowledge. The latter may be pertaining to the class of scenes under consideration and/or general knowledge about regions, lines, edges, angles, corners,...etc. In this case we may refer to the system as a "knowledge-based system", in other words we may define the knowledge-based system as: a system whose output depends upon the use of external information (knowledge) that is independent of that contained within the input digital image.

In this review, the two following Sections 2.4.2 and 2.4.3 discuss the two basic problems which are associated with the construction of knowledge-based systems, knowledge representation and control structure. Three different methods of introducing the knowledge to the analysis algorithm are then described in Section 2.4.4. Examples of different structures of knowledge-based systems will be briefly reviewed in Section 2.4.5. These include

HEARSAY-II, a speech recognition system, and the work of Levine and his co-workers on image segmentation and interpretation. In Section 2.4.6, a review of the limited work in the development of knowledge-based systems for visual motion understanding is presented. Finally, in Section 2.4.7, the different aspects of this review will be summarized.

## 2.4.2 Knowledge Representation

Knowledge representation is the first problem to face the designer of a knowledge-based system. Most research has concentrated on the development and use of "models" which describe the problem domain at different levels of abstraction [Levine, 78; Riseman and Hanson, 78; Hewitt, 77; Soloway and Riseman, 77]. Thus, models play an important role in the organization of the descriptive information incorporated in a given knowledge representation. However, model construction can be very difficult for ill-defined problem domains [Scacchi, 79].

In the past many knowledge-based computer vision systems have been developed based on using specialized knowledge models of the problem domain under analysis. Zucker et al. have suggested the development of a system capable of analyzing different scene classes, instead of developing different models for different classes [Zucker et al., 75]. In their approach, the knowledge to be utilized by a knowledge-based system is classified into two

categories, scene-independent and scene-dependent. The former includes local features (edges, lines, angles, ...etc) that occur in many different types of scenes, as well as knowledge to coherently group these features. This knowledge may be represented as "general purpose models". The scene-dependent knowledge includes descriptions pertaining to the scene to be analyzed (for example the location, shape, or the existance of specific objects). The models representing such knowledge are "specialized models". Thus, if the scene-dependent knowledge is structurally separated from the scene-independent knowledge, changing the class of scene to be analyzed will only necessitate substitution of the scene-dependent knowledge base [Levine and Shaheen 81; Riseman and Hanson, 78]. General purpose models have the advantage that they can be used in analyzing different types of scenes, even when no a priori knowledge about the scene is available. A prime example of systems based on this strategy can be found in the low level segmentation system developed by Levine and Nazif [Levine and Nazif, 82].

## 2.4.3 Control Structure

The second problem encountered in designing a knowledge-based system deals with the design of an efficient control structure, necessary for an effective use of the knowledge organized at different levels. Thus, a knowledge-based system which employs different levels of

processing, and uses diverse knowledge sources organized at multiple levels, needs a control structure mechanism to focus its attention on which processing task to be activated, and on which knowledge is to be chosen. This attention focusing mechanism may be directed by one of two control structure strategies: data-driven, and model-driven [Lesser and Erman, 77; Nii and Feigenbaum, 78]. In the data-driven control structure, the systems's processing attention is directed by incoming low level information, ie. the occurence of specific observed events, such as recognizing or characterizing specific object features [Levine, 78; Levine and Shaheen, 81; Levine and Nazif, 82]. In the model-driven control strategy, the system relies on its existing knowledge (scene model) to suggest or hypothesize the occurrence of objects or events [Tsotsos, 80].

## 2.4.4 Knowledge Interaction

Another issue related to knowledge-based systems is the interfacing of the knowledge with the analysis processors. This interfacing may be accomplished through one of three possibilities:

(a) by incorporating the required knowledge directly into the analysis processors,

(b) through interaction with the user (man/machine interaction),

(c) by storing in a properly designed database.

Early work in computer vision included the knowledge (scene model) within the analysis processes. These systems achieved satisfactory results with limited classes of scenes, such as the blocks world [Shirai, 75] or office scene [Garvey and Tenenbaum, 74]. Shaheen concludes that the disadvantages of this methodology rest in the inhibiting of the flexibility, extensibility, and the capability of experimentation with the system [Shaheen, 79]. It was precisely this factor that motivated Levine to use the interaction method in the early version of his reported computer vision system structure [Levine, 78].

Introducing the world knowledge through an interactive process is necessary to gain the experience required to build the "intelligent system". Experimentation with different types of knowledge at different levels of abstraction for different scene classes will lead to an efficient design of the database. Examples of interactive knowledge-based systems can be found in [Ariki et al., 78; Levine, 78; Tsotsos, 76; Futrell and Speckert 78; and Potel and Sayre, 76].

Accessing the knowledge from a properly initialized database is an essential factor in the automation of an image understanding system. However, construction of such a database is not easy, especially in the case of multiple level knowledge representation. Therefore, systems using this method of knowledge/process communication should be supported by an efficient control structure strategy as

described above. In the case of a general interpretation system, the complexity of the database structure is proportional to the generality of the system (the extent of different scene classes to be analyzed by the system).

In this section we described some (not all) aspects related to the structure of a knowledge-based system as a tool for a computer vision system. The literature which will be reviewed in the following section represents but a fraction of the accumulated work and experience gathered in this field and for purposes of this thesis the most closely related to our current research.

## 2.4.5 Examples Of Knowledge-Based Systems

### HEARSAY-II

[Lesser and Erman, 77]

In HEARSAY-II a "blackboard" global data structure was introduced as a means of communication and interaction between the different sources of knowledge. This knowledge structure serves as a short term database for storing active data generated at different levels. A scheduler is used as a mechanism to focus the system attention to which chunks of information are useful for a given task, and which task should be activated.

In general, HEARSAY-II has initiated a new generation of complex systems. It illustrated how a system performance may be affected by its knowledge engineering. Because of the potential of the HEARSAY-II application to other research domains, it has influenced other approaches by many researchers. It had an impact on their work and ideas for different fields, especially in computer vision. The role of the STM in Levine's vision system is very similar to that of the blackboard in HEARSAY-II. The global blackboard structure, together with the attention focusing schemes, provide a working control strategy for an understanding system that analizes multiple knowledge sources organized at different levels. It seems that a knowledge-based vision understanding system could be developed along these lines.

## An Image Segmentation and Interpretation System

[Levine, 78; Levine and Shaheen, 81; Levine and Ting, 1981]

The concept of cooperating independent sources of knowledge which operate on a global data structure has motivated the framework for a computer vision system proposed by Levine [Levine, 78]. This framework laid the foundation for much advanced work in computer vision carried out in the last few years by Levine and his co-workers at McGill University [Levine, 78; Levine and Shaheen, 81; Levine and Youssef, 80; Levine and Nazif, 82; Levine and Ting, 1981]

The main objective of Levine's work was to develop an interactive computer vision system to experiment with different picture strategies. The system consists of three hierarchical levels. The objective of the low level process is to segement the image into regions possessing similar primary features such as intensity, hue, saturation, and texture. The result of this process may be described as complete or partial segmentation, depending on whether the segmented regions correspond to object as perceived by a human observer or not. A complete segmentation is possible for a picture which contains nonoverlapping objects on a uniform background, such as blood cells; whereas partial segmentations result from the processing of normally complex pictures, which contain objects that exhibit depth, occlusion, shadows, and highlights, such as for example, outdoor scenes.

The resulting regions from the low level partial segmentation may be the input to an intermediate level processing [Levine and Ting, 1981; Ting, 791. At this level, the model features and topological structure are used in two stages, local and global. A local template matching is used to match regions against object prototypes, followed by global optimization using dynamic programming. Other optimization techniques could also have been used. The result of this intermediate level is a group of merged regions, each of which is assigned a set of region interpretations. At the highest level of the hierarchy, a

vision production system, in conjunction with a relational database, is used to complete the analysis.

The implementation and experimentation results of the high level computer vision interpretation system proposed in [Levine, 78], have been reported recently [Levine and Shaheen, 81]. The results show that the performance of the high level processing stage and consequently the final interpretation of the scene depends strongly on the initial segmentation (low level processing). Therefore, the objective of the ongoing research by Levine and Nazif [Levine and Nazif, 82] is to develop a general purpose low level rule-based system to improve the initial segmentation.

## Rule-Based System for Low Level Image Segmentation

[Levine and Nazif, 82; Nazif and Levine, 82]

The objective of this work is to design a low level rule-based segmentation system in order to test different segmentation strategies and compare their results to those obtained by a humans. The approach is based on using general knowledge about low level properties of the image in the form of condition-action rules, in order to decide, for example, if a specific region(s) should be merged or split. Thus, for an arbitrary initial segmentation, they test different strategies which employ different sets of rules, in order to find a set of condition-action rules which leads to the best segmentation. For example, in one of the methodologies they use, the process begins with an initial

segmentation, and then iterates by merging and splitting the different regions until the final segmentation is achieved. The action, merge or split, is decided by the incoming low level data conditions (region features, neighborhood in the picture) and a set of condition-action rules. The latter is a representation of the general knowledge pertaining to regions, lines, and groups of both.

The potential of Levine and Nazif's low level segmentation system rests on the following factors:

(a) The system accepts any level of initial segmentation, for example, the entire image may be considered as one region or each pixel as a region,

(b) The system facilitates experimentation with different rules and sets of rules to examine the different segmentation strategies,

(c) Scene context knowledge is not required, therefore the system is applicable to any class of scenes (general purpose model).

(d) The control structure is that of a production system, in which control rules are used in addition to the knowledge rules.

(e) The output of the system provides a description of the image in terms of regions (uniform neighborhoods in the picture), lines (major discontinuities in features), and areas (large regions or groups of regions and lines

which represent textured areas),

(f) Early results obtained using this system indicate promising results for image segmentation which are very close to the low level output expected by a human observer.

In this section we reviewed a few examples of knowledge-based systems; however our discussion was restricted to computer vision systems for static scene analysis. In the following section we shall discuss the same topics for visual motion understanding from sequence of images.

## 2.4.6 Knowledge-Based Systems For Visual Motion Understanding

From the discussion in the preceding section we may realize the difficulty in designing a computer vision system for static scene analysis. For motion understanding from a sequence of images, the task is more difficult. This is because we face all the difficulties of static scene interpretation, as well as the problems of motion understanding and description. Tsotsos has commented on this difficulty by stating that "motion understanding is a monstrously large problem" [Tsotsos, 76].

Similar to our discussion in the previous sections with regard to image interpretation, visual motion peception consists of two hierarchical processes, motion detection and motion understanding. These aspects were discussed in Section 2.2.2.1, referring to the study by Martin and Aggarawal, and in Section 2.2.3, we reviewed the aspects and the related work that have been done in image sequence analysis in general. In this section, we will focus our attention on the aspects of visual motion understanding by using knowledge-based systems.

Most of the work that has been done in image sequence analysis has been restricted to the fairly low level processing part, which is concerned with motion detection. The few approaches which considered an intermediate level (motion interpretation) and/or high level part (motion understanding) have been based on interactive systems [Badler, 75; Price and Reddy, 77] and specific domain applications [Tsotsos, 80].

The first attempt in constructing a knowledge-based system for motion understanding may be related to Badler [Badler, 75]. He used the motion verbs as models to describe the motion concepts. Based on Badler's approach, Tsotsos has adapted the problem of motion understanding to his research. Tsotsos's recent work, in spite of its restrictions, may be considered the first actual knowledge-based system employing high level knowledge (models) to describe the motion concepts.

In order to develop a visual motion understanding system, we should make use of the experience gained in computer vision of static images (this is our philosophy in the current research). Scacchi reported an outline for future work towards the development of an "intelligent system" for visual motion perception. His report is based on emerging experience from computer vision, hardware-based vision systems, computer animation, artificial intelligence, and human motion perception [Scacchi, 79]. Considering the different relative viewing situations, Scacchi specifies five requirements for developing a visual motion perception and understanding system:

(a) a "long-term" memory to model the features and spatial relationships of known or observed static objects,

(b) an "intermediate" memory where the focusing mechanism (control structure) can interact with both the knowledge-base (database) and incoming data (dynamic data),

(c) a "low level" visual memory to support hardware-based capabilities for extracting the features from the scene,

(d) an attention-directed "retina" to observe object features within the image, which acts as a knowledge-base process,

(e) the different processes and system structure should interactively work in no predetermined order (data-driven).

The system which Scacchi outlined and seeks is an integrated system (he claims) similar to HEARSAY-II, which has "the ability to analyze, understand, and react to a bounded though not necessarily predictable range of situations arising from conflicting, competing, and cooperating interactions" [Scacchi, 79]. This integration may be achieved through a distributed control structure of the different knowledge sources and processing activities, directed by knowledge-based transactions.

As a concluding remark, one can see that the outline for visual motion understanding, to a large extent, shares the same philosophy of the recent trend in computer vision of static images which was proposed by many approaches [Riseman and Hanson; 78, Levine, 78]. This philosophy is very closely related to our approach in this research which is described in the next section.

## A System for Understanding the Dynamic Behaviour of a Moving Cell

[Levine and Youssef 81]

In our current research, we have designed a knowledge-based system for understanding and describing the dynamic behaviour of non-rigid moving objects. From a

philosophical point of view, the present structure is motivated by the computer vision framework proposed by Levine [Levine, 78]. The latter has been revised and implemented by Levine and Shaheen for general static scene analysis and interpretation [Levine and Shaheen, 81; Levine and Nazif, 82]. The structure consists of independent analysis processes cooperating through a common database structure.

Our proposed structure for a general motion understanding system involves three basic entities: dynamic data, static data, and a collection of analysis processes, each of which is assigned a particular task. Conceptually, two different memories are used, a Short Term Memory (STM) and a Long Term Memory (LTM). The dynamic data are continually changing as a result of the functioning of the different analysis processes. They are stored in the STM, which is designed to work as a communication channel for all of the processes. Each process can read from and write into the STM. It contains a record of the instantaneous object motion, shape, and structural changes, as well as the current global description of the object dynamic behaviour. The static data in the LTM remains unchanged during the course of analysis, and contains constraint knowledge pertaining to the class of scenes and type of motion under analysis, as well as the pertinent computational processes. The system also consists of different computational processors, which are designed to execute through a (loose)

hierarchical structure consisting of three basic levels: static, incremental, and global (see Figure 1.4).

## 2.4.7 Summary

The objective of computer vision systems is to interpret and describe the contents of a given digital image. In image sequence analysis this objective includes the understanding and description of the motion recorded within the sequence. In order to accomplish this, the system should utilize information from two sources, the input image, and external knowledge pertaining to the class of scenes and/or type of motion under consideration. The system which utilizes this type of external knowledge is refered to as a knowledge-based system. Currently, the structure and use of knowledge-based systems represents a topic of broad interest within the computer vision and AI community.

One of the basic and major problems in the structure of knowledge-based systems is the representation and efficient use of this knowledge. A common and powerful paradigm suggested by many approaches, is to represent the knowledge through "models" describing the problem domain under consideration. Most of the early computer vision systems are based on using models of specialized knowledge describing the class of scenes under analysis. The recent trend in the structure of knowledge-based systems is directed to the use of a general purpose models, which can

be used for a wide variety of scene classes [Zucker et al., 75; Levine and Nazif, 82].

Knowledge-based systems which employ different levels of processing that use multiple sources of knowledge organized at different levels of description, need an efficient control structure mechanism. The function of the control structure is to regulate the social behaviour (activities) of the system, that is to decide what and when specific knowledge should be used, which processor should be activated, and the general communication between the knowledge and the analysis processes. Two types of control structure have been defined, model-driven and data-driven. The knowledge pertaining to the control structure is also represented in the system by the models.

In the preceding sections, we have reviewed some of the related approaches which propose a basic knowledge-based structure. The foundation of most of the recent approaches were found in HEARSAY-II, the speech recognition system. The computer vision system proposed by Levine [Levine, 78] has the same philosophy as that proposed by Riseman and Erman [Hanson and Riseman, 75]. Both structures are based on independent processes that cooperate through a common database structure.

The progress towards the development of knowledge-based systems for visual motion understanding is slow, and the work which has been done is very limited. The first step in

this direction was taken by Badler [Badler, 75], who used motion verbs as models to describe motion concepts. The application was to the generation of line drawing images. Tsotsos has been engaged in the problem of motion understanding; his recent report [Tsotsos et al., 81] may be designated as the first knowledge-based system for visual motion understanding, in spite of its restrictions (see Section 2.2.3.3). Scacchi has presented an outline of the structure of a knowledge-based "intelligent" system for visual motion understanding [Scacchi, 79]. His outline is based on emerging experience from computer vision (scene analysis), hardware-based vision systems, computer animation, artificial intelligence, and human motion perception.

Considering the previous discussion, we may conclude that an understanding system, either as a computer vision system for static scene interpretation, or visual motion description from a sequence of images, requires the construction of a knowledge-based system. This system should utilize the knowledge from diverse sources of information, consisting of multiple levels of analysis, and to be supported by an efficient control structure mechanism.

In light of the above discussion on the aspects and structure of knowledge-based systems and the structure developed in this thesis, we may claim that our research has successfully utilized the most advanced strategies of computer vision interpretation of static images, merging

them with the experience gained in image sequence analysis to construct a visual motion understanding system.

## 2.5 AUTOMATIC PROCESSING OF MICROSCOPIC IMAGES

### 2.5.1 Introduction

The early history of the automatic image processing of cell images can be traced to the 1950's, and is directly related to the development of the so-called television microscope [Preston, 76]. Most of the work which has been done in this field has been concerned with the feature extraction and analysis of cell images for theoretical study and research. However, in the practical field, there have been studies aimed at the automation of the recognition, classification, and counting of the cells in a blood smear. Significantly, little of this work was addressed to the tracking and study of cell locomotion. Recently, experiments have indicated that the cell membrane plays a vital role in the mechanisms that regulate the social behaviour of the cell, of which locomotion is an important component. However, there is no existing work that attempts to quantify the observable changes in the membrane shape that occur in locomotion.

In that which follows, we shall present a concise description of the important work done in each of these directions. First, a brief review of the progress in the studies of cell morphology is traced in Section 2.5.2.

Second, the significant work which has been done in automatic processing of cell images for counting and classification purposes is reviewed in Section 2.5.3. Third, the early techniques for cell tracking are reviewed in Section 2.5.4. The fourth Section 2.5.5, discusses some of the advanced approaches for the quantification and analysis of cell locomotion, focusing attention on projects which have been under study at McGill University. The recent interest in the role that cell membrane plays in locomotion is discussed in Section 2.5.6. Finally, in Section 2.5.7 a conclusion of this section, as well as the contribution of our research to the field of automatic processing of cell images, is presented.

## 2.5.2 Theoretical Study Of Cell Morphology

The initial effort in the processing of cell images was concerned with the study of cell morphology, primarily for the purposes of clinical diagnostic application. Early in 1952, Young and Robert developed a flying-spot scanner for use in particle size analysis [Young and Robert, 52]. By 1961, a special-purpose computer and television microscope had been constructed as reported by Izzo and Coles [Izzo and Coles, 62]. Its initial use was in the research of the feasibility of screening blood smears for rare types of blood cell whose occurence appeared to be significant as an indicator of low levels of radiation damage to humans.

A major program in chromosome picture processing, using a photomicrograph scanner rather than a television microscope as the input device to the general purpose computer, was established at the National Research Foundation (Washington, D.C.) and the new England Medical Center (Boston), as described by Ledley et al. in 1965 [Ledley et al., 65]. In 1968, the first general-purpose system for the digital analysis of cell images was realized by Wied et al. [Wied et al., 68]. In 1971, Ledley described three different methods for analyzing cell images [Ledley, 72]. In the first method, intercommunicating programmable cursors are utilized for the detection of boundary and area features. The second is concerned with an evaluation of the curvatures of boundary segments to isolate the cells. The third method imbeds the picture data in a non-Euclidean coordinate system as an aid to the ensuing analysis.

As recent as 1975, Bacus reported on a novel method which required that the cell image be digitized through two different colour filters [Bacus, 76]. He then used a whitening transformation technique to produce two separate, transformed "colour" and "density" images. Based on this type of preprocessing, he developed a scene segmentation technique for blood cell neutrophils [Mui et al., 76].

## 2.5.3 Cell Counting And Classification

In order to develop a practical analysis system, it was necessary to study pattern recognition and classification of cells. The automatic classification of peripheral blood leukocytes has also been the subject of considerable study since the 1960's [Bacus, 70; Ingram et al., 68; Ingram and Preston, 70; Mendelsohn et al., 68; Prewitt and Mendelsohn, 66].

In one of the earliest pieces of work, Prewitt and Mendelsohn attempted to classify blood cells into five categories, utilizing an optical density histogram representation of the digitized cellular image [Mendelsohn et al., 68; Prewitt and Mendelsohn, 66].

During the period 1969-1972, Young at MIT simulated a system to perform an automated leukocyte differential count through the measurement of nucleus and cytoplasm color and size [Young, 69]. He classified the leukocytes into one of five basic types: neutrophil, cosinophil, basophil, lymphocyte, and monocyte. Ingram and Preston worked with larger data bases but classified cells into only three categories [Ingram et al., 68; Ingram and Preston, 70].

Later in 1972, Bacus et al. dealt with the difficult interclass problem to classify the peripheral blood leukocytes into eight categories [Bacus and Gose, 72]. They used as morphology measurements, the nuclear size, nuclear shape, nuclear and cytoplasmic texture, cytoplasm colour,

and cytoplasm texture. The leukocytes were classified into the following categories: "small lymphocytes, medium lymphocytes, large lymphocytes, band neutrophils, segmented neutrophils, cosinophiles, basophils, and monocytes".

Although more recently, several private companies have begun the development, and in some cases the marketing, of practical classification devices, the majority of the reported research has been of a very preliminary nature. For example, the investigations were carried out with extremely small data bases, usually with cells from only one person, and with either no independent testing set, or an extremely small one. Some of the investigations have purposely not included "difficult" or "nontypical" cells [Causley and Young, 53].

## 2.5.4 Early Work On Cell Tracking

With regard to the analysis of cell movement, Parpart was one of the first to examine this problem in 1951 [Parpart, 51]. He used television to study the movement of blood cells. During the period 1953-1957, Causley and Young [Causley and Young, 53], Hawksley [Hawksley et al., 54], and Barer [Barer, 57] used television techniques to study living cells.

In 1975 Greaves reported on an interactive on-line computer-television system for studying the behaviour of moving organisms [Greaves, 75]. Parameters relating to this

behaviour, such as for example velocity and rate of change of direction, were extracted with the aid of an interactive graphics system. In 1976, Green and Barnes designed an electromechanical instrument to automatically follow the movements of an individual white blood cell [Greene and Barnes, 76]. Youssef [Youssef, 77] discusses this literature in detail.

## 2.5.5 Quantification Of Blood Cell Locomotion

The movement of blood cells and the factors which affect their locomotion are of importance to the understanding of the role these cells play in host defense mechanisms. A major project which concerns the study of blood cell movment, quantifying and characterizing the different factors controlling their dynamic behaviour, has been carried out during the last few years in the Computer Vision and Graphics Laboratory (CVaGL), at McGill Universty. Four projects have been under study:

(i) Quantification of blood cell movement by automatic image processing methods.

(ii) A real-time laboratory device for tracking and quantifying blood cell movement.

(iii) An automatic picture processing method for extracting genealogical information from proliferating cell cultures.

(iv) Quantification and characterization of the motion and shape of a moving cell.

The first two projects have been completed and their results have been reported [Levine and Youssef, 78a; 78b; Knoll, 79]. The third project is discussed in [Ferrie, 79; Ferrie et al., 82; Ferrie and Levine, 81]. The proposed approach adopted by the fourth project has been presented by Youssef and Levine [Levine et al., 79; Youssef and Levine, 80]. Recently this work has been completed and the results are presented in this thesis. A brief review of the objectives and achievements of the above projects is presented below.

(i) Quantification of Blood Cell Movement by Automatic Image Processing Methods

In order to facilitate the study of cell movement, Levine and Youssef introduced a new automatic picture processing method for tracking and quantifying the dynamics of blood cell motion [Youssef, 77; Levine and Youssef, 78; Levine et al. 80]. The input to the program is a 16 mm cine film of the cell culture viewed in a steady state. Under the circumstances, the global directional movement of the group of cells under consideration can be characterized by a Markov chain model [Boyarsky, 77]. By observing and quantifying the cell paths, it is possible, using this model, to determine the probability that the cell population

is moving in a particular direction. This information might be of interest in the study of the effect of substances on cell movement, defects in white blood cell migration, and the general interaction among cells.

The above approach has many desirable features over previous work. The system has successfully achieved the objective of automatically tracking a group of live blood cells. It quantifies the cell path data, and computes the steady-state probabilities in order to predict the direction in which the cells will ultimatly move. Besides obtaining accurate steady-state results, the system can also be used to obtain a path for each tracked cell. This data is essential to the study of the characterization of locomotion for different types of cells under varying environmental influences.

(ii) A Real-time Laboratory Device for Tracking and Quantifying Blood Cell Movement

Based on the approach described above, Levine and Youssef presented a design for an integrated practical laboratory system which facilitates the tracking and quantifying of the cell movement in a real-time environment [Levine and Youssef, 78b]. The construction of this device is based on observing the cell movement directly (in real-time) via a microscope using a TV camera connected to a digitizer. The practical implementation of this decice was reported by Knoll [knoll, 79]. Another desirable advantage

of this device is that besides tracking the cell in real-time, it also reduces the noise in the input image which results from recording the motion on a cine film. Other possible applications might be in the study of the effect of drugs on cell movement, defects in WBC migration, and general interaction among cells.

Two common restrictions characterizing all the tracking approaches described thus far (including the approach of Levine and Youssef) are: (a) the cells being tracked must remain well isolated from one another (no occlusion or overlapping), (b) most of the approaches are based on centroid matching techniques. Thus, the changes on the cell shape that may occur between the sequential frames have been ignored. In order to develope a tracking system which considers these factors, the system should utilize world knowledge about the class of scene and the objects to be tracked (see Section 2.4 on knowledge-based systems). The first tracking system which considered these restrictions was developed in Japan at Koyoto University by Ariki et al. [Ariki et al., 78]. They proposed an interactive image modeling system for tracking moving objects from a sequence of images. They accomplished this objective by constructing interactively, models of the object to be tracked. Using these model patterns, they can measure the changes in specified features (intensity, location, and shapes). The first system which achieved the same objectives automatically was developed by Ferrie and Levine [Ferrie and

Levine, 81]. A brief description of this approach is presented below.

(iii) An Automatic Picture Processing Method for Extracting Genealogical Information from Proliferating Cell Cultures

The study of Ferrie and Levine was concerned with extracting genealogical information from proliferating cell cultures by using automatic image processing techniques [Ferrie and Levine, 81]. Their tracking approach is based on updating the description of the cell in the current frame (t) by matching the feature vectors of each candidate in this image to the feature vector of the cell in the previous frame (t-1) (the static model of the cell). Two cases arise: (a) the cell is located, then the static model is updated and the tracking is continued, (b) the match failed indicating that the cell has undergone a state transition. In this case the process computes the hypothesis of the different possible state transitions. By testing each hypothesis individually, the system selects the one that best matches its supporting evidence.

In this approach, in order to solve the correspondence problem, explicit models of the different state transitions of the cell are used. However, the random morphological changes, especially with a phase-contrast objective, are very difficult to model. Consequently, the prototype models of the cell transitions are not sufficient to solve the correspondence problem in these cases. Of particular

interest, is the fact that an "expert" human can keep tracking the cell despite these artifactual problems. Thus, he uses cues from the visible morophlogy of the cell to distinguish those which are artifactual due to shadows or phase-contrast.

The system which simulates this "expert" process is the objective of the current research by Levine and Ferrie [Ferrie and Levine, 81]. They propose to accomplish this objective by modeling the set of visual cues and response of the expert which he uses to identify the location of a cell. This knowledge will be represented in the form of implicit models which may be used through inferential mechanisms where syntactic knowledge (explicit models) are not sufficient.

## 2.5.6 Membrane Shape Changes

The main function of a cell's surface is to receive information from the environment. It has become increasingly evident that the surface plays a pivotal role in the life, development, and regulation of cells. The mechanisms that regulate this social behaviour of cells, of which locomotion is an important component, are not well understood. Recently, experiments have indicated that the cell membrane plays a vital role in these mechanisms. However, there is no existing method to quantify the observable changes in membrane shape that occur in locomotion.

The work of Lewandowska et al represents an empirical approach to analyzing the locomotion of leukemia cells by a computer image processing method [Lewandowska et al., 81]. They measure the location and shape of specific types of cells (leukemia), using a group of simple features, such as area, perimeter, ratio of perimeter to area, and elongation. These features are computed for a few frames (10 frames are reported) selected interactively from a sequence of images recording the cell locomotion. This interactive selection is based on choosing those frames in which a significant change in the cell's shape has occured. Then, the computed features of the cell in each of these frames are compared with ten simple prototype models describing the different patterns which may be exhibited by the cell.

For obvious reasons we are not going to compare our current work to the approach of Lewandowska et al., however we may point out the restrictions of their work by the following points. Their method is applicable to lymphocyte cells (leukemia) which exhibit relatively simple shape patterns (which are easy to model) during their locomotion, compared to PMN. The system computes only the location and general shape (using shape features useful only for simple shapes) in a few static frames, ignoring the structure of the cell. The system does not deal with any problems of image sequence analysis, such as the quantification and description of incremental or global changes. Another major problem in image sequence analysis is finding the frames in

which significant changes have occured (key frames) and which is solved in this system interactively (manually).

On the other hand, in spite of the simplistic methodologies and technicalities of Lewandowska et al, this work is still worth mentioning for two reasons: First, it is an attempt towards the solution of a difficult problem, namely quantifying and characterizing the observable changes in the cell membrane. Second, this work is rooted in a completely independent local research experience (they do not make use of or refer to any of the previous work in the related areas). For example, they refer to the ratio of the area over the perimeter as the "Malinowska factor". This feature is now established in the computer vision community as "circularity". Another example, is that the method they use to generate the polygonal approximation of the cell shape, has no theoretical basis. Therefore, in many cases the polygon may represent a completely different shape from the cell.

### 2.5.7 Summary

From the previous brief discussion, it is evident that research on the analysis of cells using computers has been carried on for over twenty years. This has more recently led to the development of experimental and also practical systems whose performance in many cases equals that of the human technologist.

The major efforts in automatic image processing of cell images have been focussed on the analysis of chromosomes, blood smears, and cervical smears. In general, the majority of the work which has been done in this area has been directed towards the recognition and classification of cells on a blood smear. The study of cell movement and the characteristics of cell interaction have been largely ignored. However, the recent work at the Computer Vision and Graphics Laboratory at McGill University offers the possibility of quantifying and characterizing the cell dynamic behaviour.

The structural changes in the cell morophology that occur during locomotion have not been reported in the literature. Furthermore, in spite of the importance and great interest in understanding the role that the cell membrane plays in locomotion, there is no existing method for quantifying and analyzing the observable changes in the membrane shape.

In our current research we have developed an image interpretation system capable of quantifying, analyzing, and describing the structural changes in the morophology of a moving cell. Thus, we see that the contributions of our research in the automatic processing of cell images lie in the following:

(a) With the aid of the global observable changes in the cell locomotion, one of the main behavioural characteristics is mathematically quantified and described; namely, the chemotactic behaviour (the directional locomotion of the cell with respect to the directional effect of an external factor). Consequently, the effectiveness of an external factor in modifying the cell locomotion is quantified.

(b) A mathematical expression for measuring the complexity of the cell shape pattern has been developed and used to describe the membrane shape and its observable changes.

(c) The global changes in the cell structure are also analyzed; hence, a subpart of the cell is classified as being a "pseudopod" or "cell body", and a pseudopod is described as "growing", "contracting", or "stationary".

(d) Furthermore, some aspects of the global behaviour of the cell are summarized and described; for example, the "domination" of a pseudopod in leading the locomotion of the cell.

(e) We describe the dynamic activity of the cell using a symbolic terminology which is meaningful to the biologist.

This computer study might provide clues to the nature and distribution of "receptors" on or within the membrane, which is a vital link in the interaction between the external factors and cell internal processes. Also, it might lead to a better understanding of the role that the cell membrane plays in the mechanisms which regulate the social behaviour of cells.

# CHAPTER 3

# SYSTEM AND DATA STRUCTURE

## 3.1 INTRODUCTION

The main goal of our research is to design and implement an image understanding system capable of analyzing the locomotion and structural changes in the shape of a non-rigid moving object from a sequence of pictures. Any system which tries to solve this problem thoroughly is attempting to imitate human visual motion perception. The latter constitutes a hierarchy of processes, which includes motion detection, understanding, and description. All of these aspects are addressed by this research.

From a philosophical point of view, the present structure is motivated by the computer vision framework proposed by Levine [Levine, 78]. The latter has been revised and implemented by Levine and Shaheen for general static scene analysis and interpretation [Levine and Shaheen, 81]. A rule-based system for low-level image segmentation is described in [Levine and Nazif, 1982].

Our proposed structure for a general motion understanding system involves three basic entities: dynamic data, static data, and a collection of analysis processes, each of which is assigned a particular task. The dynamic data are continually changing as a result of the functioning of the different analysis processes. They are stored in the the STM, which is designed to work as a communication channel for all of the processes. Each process can read from and write into the STM. It contains a record of the instantaneous object motion, shape, and structural changes, as well as the current global description of the object dynamic behaviour. The static data in the LTM remains unchanged during the course of analysis, and contains constraint knowledge pertaining to the class of scenes and motion under analysis, as well as the pertinent computational processes.

The system also consists of different computational processors, which are designed to execute through a (loose) hierarchial structure consisting of three basic levels: static, incremental, and global (see Figure 1.4). These may be described as follows (Figure 1.5):

Static Scene Analysis:
==========

This step is similar to a static image processing system. The input is a single digital image, examples of which are shown in Figure (4.3). The output is a description and interpretation of the scene. However, in image sequence analysis, the information extracted from the previous frames

of the same sequence may be used to assist the analysis of the current frame. The main objective of this stage is to identify the desired moving object, segment it, and describe it in each frame of the sequence. Figure (1.5a) shows a block diagram of this stage of analysis, and Figure (4.1) shows the processes and data structure. The segmentation output can be seen in Figure (4.21a). In (4.21b), the result of static shape analysis is presented, in which the cell is represented by a labeled star graph. Description (4.1) is typical of those generated by the system. How this is obtained is described in Chapter 4.

Incremental Change Detection:
==========

This stage is an intermediate step between the static and the global. The main objective is to detect and describe the incremental changes in the shape, structure, and motion of the moving object between two sequential frames (see Figure 1.5b). Figure (5.1) shows the processes and data structure of this stage, and Description (5.1) gives an example of a summary of the incremental changes between two sequential frames. This stage of analysis is described in Chapter 5.

Global Analysis:
==========

This presents the highest level in the hierarchy of the system. The goal is to analyze the static and incremental data in order to detect and describe the global observable changes within the sequence of frames (Figure 1.5c). Two basic steps are involved. The first is concerned with the

global description of three aspects pertaining to the cell dynamics; that is, locomotion, shape, and structure. The second integrates these different global descriptions into a coherent characterization of the behaviour of the cell. The main steps of this stage are shown in Figure (6.1). The methodologies and implemented techniques pertaining to this stage are described in Chapters 6-8, inclusive.

The different processes interact through the STM using the information stored in the LTM, until a complete description of the dynamic cell motion and morphology is obtained. Examples of these descriptions are given in Descriptions (6.1), (7.1), and (8.3).

## 3.2 DATA STRUCTURE AND KNOWLEDGE REPRESENTATION

The different types of data which may be manipulated by the system are classified into: a sequence of images, a group of objects and subobjects, a set of features, a group of symbolic descriptors, a group of characteristics, and a set of rules. The latter may be further divided into representational and control rules. In this section, the definition of these data will be presented. Also, we will demonstrate how they can be used for knowledge representation and model construction at any level of abstraction.

## 3.2.1 Basic Elements For Knowledge Representation

The main input for any image sequence analysis system is a series of two-dimensional digital images representing the variation in a specific scene along a third-dimension. These images may be obtained in various ways; for example, cine film, video tape, or a TV camera observing a scene in real time, whereby the images are captured at specific time intervals. Whatever the input device, the actual data for the analysis system is a set of two-dimensional images {I}, where each element of this set represents a static (single) image of the scene at a specific time ti. The latter are elements of {T}, the set of sample times at which the frames are obtained:

$$\{T\} = \{t1, t2, t3, \ldots, ti, \ldots, tn\}, \quad (3.1)$$

and Iti is the image of the scene at time ti. The period of time between two frames (i) and (k) is given as Tik.

Each image Iti of the temporal sequence may be segmented into a set of objects {OBJECTS} so that :

$$\{OBJECTS\} = \{O1, O2, O3, \ldots, On\} \quad (3.2)$$

This set may also be divided into two subsets according to whether the object is moving or stationary:

$$\{OBJECTS\} = \{MOVING\ OBJECTS\}, \{STATIONARY\ OBJECTS\} \quad (3.3)$$

$$\{MOVING\ OBJECTS\} = \{MO1, MO2, \ldots, MOm\} \quad (3.4)$$

$$\{STATIONARY\ OBJECTS\} = \{SO1, SO2, \ldots, SOk\} \quad (3.5)$$

where MOi is moving object(i) and SOi is stationary object(i).

Objects have a complex shapes, and it is usually necessary to decompose them into primitive subparts. The result of this decomposition is a collection of SUBOBJECTS associated with each object. Thus, for example, a moving object MOi may be decomposed into :

$$MOi = \{SMOi1, SMOi2, \ldots, SMOs\}, \quad (3.6)$$

where s is the number of subobjects of object MOi. If MOi = {0}, this indicates that the object MOi is simple and cannot be decomposed. Figure (4.12b) shows an example of a sequence of cell boundaries, Figure (4.15) indicates their polygonal approximations, and Figure (4.17) illustrates the cell decompositions. In our case, the object is a cell whose decomposition is represented by a star graph. The central node indicates the body of the cell, while the others symbolize the "bumps" on the cell membrane. It is these protrusions that ultimately grow into the pseudopods discussed earlier. An example is given in Figure (4.18).

A set of objects is described by static features. The latter define the different properties of shape, structure, or motion of the objects and subobjects to be measured or analyzed by the system:

$$\{FEATURES\} = \{PROPERTY, NUMERICAL\ VALUE\} = \{P, V\} \quad (3.7)$$

$$\text{where } \{P\} = \{P1, P2, \ldots, Pi, \ldots\}, \quad (3.8)$$

$$\text{and } \{V\} = \{V1, V2, \ldots, Vi, \ldots\}. \quad (3.9)$$

A property name in the set {P} is given by Pi. It may take on any value in the set {V}. Furthermore, we may write V(Pi) to define the numerical value of a specific

property Pi. If the value is given at a certain time tj, this is indicated by V(Pi, tj).

The set of properties may be divided (grouped) into subsets; for example:

$$\{PROPERTIES\} = \{SHAPE, LOCATION, STRUCTURE, MOTION\}$$
$$= \{SP, LP, RP, MP\} \quad (3.10)$$
$$\{SHAPE\} = \{SP1, SP2, \ldots\} \quad (3.11)$$
$$\{LOCATION\} = \{LP1, LP2, \ldots\} \quad (3.12)$$
$$\{STRUCTURE\} = \{RP1, RP2, \ldots\} \quad (3.13)$$
$$\{MOTION\} = \{MP1, MP2, \ldots\} \quad (3.14)$$
$$\{P\} = \{SP\} \cup \{LF\} \cup \{RF\} \cup \{MF\} \quad (3.15)$$

where SF, LF, RF, and MF are the group of properties which define the shape, location, structure, and motion, respectively.

Table (3.1) gives examples of the different cell properties under each group heading (shape, structure, motion, and location). This fixed list is stored in the LTM. On the other hand, the numerical values (dynamic data) of the specified properties are stored in the STM, and may be updated at any time throughout the analysis, by any process.

## 3.2.2 Symbolic Qualifiers

A set of symbolic descriptors or qualifiers {Q} is used to classify and describe the numerical values of the different properties of the moving cells. The number of

defined qualifiers may be less than the number of properties, because more than one property may be described using the same symbolic descriptor. For example, all properties based on measurements of distance between two points (length, base-line, connective-line, displacement) may be described as SHORT, LONG,...,VERY LONG. Also, as already indicated for the properties, the set of symbolic qualifiers may be grouped into subsets according to the nature of the properties they describe. Thus:

$$\{Q\} = \{SQ, RQ, MQ, LQ\} \quad (3.16)$$

$$\{SQ\} = \{SQ1, SQ2, \ldots\} \text{ , shape qualifiers,} \quad (3.17)$$

$$\{RQ\} = \{RQ1, RQ2, \ldots\} \text{ , structural qualifiers,} \quad (3.18)$$

$$\{MQ\} = \{MQ1, MQ2, \ldots\} \text{ , motion qualifiers,} \quad (3.19)$$

$$\{LQ\} = \{LQ1, LQ2, \ldots\} \text{ , location qualifiers.} \quad (3.20)$$

Each property is specified by a subset of symbolic qualifiers; thus:

$$\{Q(Pi)\} = \{Q1(Pi), Q2(Pi), \ldots, Qk(Pi)\}, \quad (3.21)$$

where Q(Pi) is the subset of qualifiers describing the property Pi. Table (3.2) gives a list of examples of different cell properties and their multiple levels of description. This type of information is stored in the LTM, whereas the symbolic descriptors of the actual image under analysis are stored in the STM. For the latter, the assigned qualifier for a specific property Pi at a given time $t_j$ is given as $Q(Pi, t_j)$.

Finally, in order to summarize the global cell behaviour, we have defined the so-called cell characteristic. Each cell characteristic is the description of a group of features which cooperate to define a specific type of behaviour. These may be based on the global changes in cell shape, structure, motion, combination of more than one global change, and/or the effect of the environment on the cell behaviour. The general form of this set of characteristics,

$$\{CH\} = \{CH1, CH2, \ldots, CHi, \ldots\}, \quad (3.22)$$

is given by

$$\{CHARACTERISTICS\} = \{PROPERTY, OPERATOR, QUALIFIER\} \quad (3.23)$$

$$\{CHi\} = \{Pi, Oi, Qi\}, \quad (3.24)$$

where Pi, Oi, Qi are elements of the sets {P}, {O}, {Q}. The sets {P} and {Q} have already been defined, and the set

$$\{O\} = \{AND, OR, LT, GT, EQ, LE, GE, NE, AE, GTT, LTT, MST, LST\} \quad (3.25)$$

specifies the relationship between the property and its qualifier.

Table (3.3) gives the definition of the different elements of {O}.

As indicated in the preceding sections for the properties and qualifiers, the set of characteristics may also be grouped into subsets, according to the nature of the behaviour they describe.

## 3.3 KNOWLEDGE REPRESENTATION RULES (LTM)

Knowledge representation and model construction are probably the most important aspects of the structure of a knowledge-based system. In the preceding section, we described the different types of data which represent the basic elements of the knowledge representation for a motion understanding system. In this section, we will demonstrate how these basic data elements can be used to create a world model.

In our proposed structure, the model which represents the knowledge contains two basic types of data, constraint knowledge and rules. The latter may be further classified into representational and control rules. The representational rules are responsible for generating the different descriptions and characteristics according to the numerical measurements of the different features. The control rules account for the activation and scheduling of the different system processors. This type of data will be described in Section 3.4.

In the human understanding system, the interpretation and/or description of a specific situation is accomplished by an inference process which utilizes the perceived data, as well as a priori knowledge and experience. The latter is modeled by the constraint knowledge. It is classified according to two basic criteria: first, whether it is scene-dependent or -independent. Second, according to the

nature of the cell properties.

Scene-independent knowledge holds for any class of objects in motion. For example, the descriptions of a circular or elongated shape. Similar examples, related to motion, are the descriptions constant velocity, positive acceleration, or negative acceleration. Conversely, scene-dependent knowledge only pertains to a specific class of scene or motion pattern. Thus, the required knowledge which may be used for the understanding of the blood cell motion is different from that which characterizes the behaviour of vehicle drivers on an urban highway, an animal under a drug protocol, or a growing plant. Both of these kinds of constraints are involved in the design of the representational rules which embody the world model.

The function, classification and construction of the rules pertaining to the image model will be discussed in this section. Rules involving the control of the analysis will be considered in Section 3.4. All the LTM rules may be described as condition-action relations. They consist of predefined situations {C} and descriptions of actions {A} to be taken when the specified situations occur. The general form of these rules is:

R U L E : if CONDITIONS ==then==>> ACTIONS
=======

{RULES} = { {CONDITIONS}, {ACTIONS} } (3.26)

{R} = {{C},{A}} (3.27)

where {C} = {C1, C2, . . . , Cj}, (3.28)
and {A} = {A1, A2, . . . , Ak}. (3.29)

Typical situations (conditions) and actions may be summarized as follows:

Situations:

(a) Completion of the execution of a specific process.

(b) Occurrence of a specific situations through or after the execution of a specific process.

(c) Specific property values.

(d) Object or subobject descriptions.

Actions:

(a) Starting and stopping the system.

(b) Activation of a specific process.

(c) Generation of a set of features, descriptions, or characteristics.

(d) Data presentation for output.

Both general and "expert" knowledge are represented as production rules. These may be classified into three groups according to their function, as follows:

(a) To translate the measured numerical values V(P) of the different cell properties into symbolic qualifiers Q(P).

(b) To generate the static, incremental, and global symbolic descriptions of the motion, shape, and structure of the moving cell.

(c) To integrate the multitude of observable changes and global descriptions into a coherent whole, in order to generate the characteristic behaviour of the cell.

In the following, the structure and examples of each of these different groups of representational rules are described.

Qualifiers {Q} (group (a) above) are the elements of natural language which humans use for describing objects. In our system, they are computed by a set of Qualifier Rules {QR}. The function of the latter is to choose among a set of symbolic descriptors the appropriate one for a given numerical value associated with a specific property. Thus we have:

{QUALIFIER RULES} = {CONDITIONS} ==then==> {ACTIONS} (3.30)

{CONDITIONS} = {NUMERICAL VALUES, OPERATORS}, (3.31)

{ACTIONS} = {QUALIFIERS} (3.32)

{QR} = {QR1, QR2, . . . , QRi, . . .} (3.33)

{QRi} = {V(Pi), O, Ej} (3.34)

where Ej is an elemment of {E}, a set of thresholds stored in the LTM. The complete set of qualifiers {Q} and operators {O} used by this set of rules was defined in Section 3.2. The numerical value V(Pi) may be defined by a measurement or a constraint pertaining to the specified property.

Two issues are of significance. How many levels, and how should the thresholds be chosen?. A thorough study of these issues is presented in Chapter 4. With regard to the first, the number of classes for a symbolic descriptor is controlled by the accepted level of approximation related to the desirable description. Specifically, the greater the number of classes (levels), the more precise the description. On the other hand, the smaller the number of classes defined, the more insensitive will be the process of analysis to noise. Obviously, more data compression is also obtained. The question arises as to how these conflicting factors can be reconciled.

The simplest method for dividing a subclass is to organize it into two groups. This is similar to transforming a gray-level image into a binary one (black and white). Thus, using a single threshold, each numerical value can be assigned to one of SHORT, LONG or FAST, SLOW, and so on. It is generally accepted that the human ability to categorize into subclasses is indeed limited. In recognition of this, we have defined the number of subclasses to be five, in the following order: VERY LOW, LOW, MEDIUM, HIGH, VERY HIGH. Table (3.2) gives examples of the different qualifiers for some of the main properties measured. In some special cases, the nature of the property to be described requires a different form of quantification. For example, in describing the direction of motion of a moving object, eight levels are defined (EAST, EAST-NORTH,

NORTH, WEST-NORTH, WEST, WEST-SOUTH, SOUTH, EAST-SOUTH).

The second issue involved in describing a set of numerical values symbolically, is how to define the thresholds in order to divide this set. This problem seems easier than it is; it involves clustering theory, human psychology, and problem domain knowledge. A thorough study of this subject has been made by Denofsky at MIT [Denofsky, 76]. The title of his study, "HOW NEAR IS NEAR?" is, in fact, a good definition of the problem. In order to design a suitable quantification to be used for different classes of objects and motion, normalization of the dynamic data is important. Thus, if the numerical values of a specific property are normalized to range between zero and one, a general rule may then be used for assigning the qualifiers. For example:

RULE(3.1):

IF V(Pi) .GE. E1, .AND. .LT. E2, ==then==> Pi<--Q1,

where V(Pi) is the normalized value of property Pk, Q1 is the chosen symbolic qualifier, and E1, E2 are classification threshold values which define the boundaries of the qualifier Q1. The action Pi<--Q1 is interpreted as assigning the qualifier Q1 to the property Pi, thereby generating Q1(Pi). An example of a complete specification is:

RULE(3.2):

(a) IF V(area) .GE. E0, AND .LT. E1, =then=> AREA <-- V.SMALL

(b) IF V(area) .GE. E1, AND .LT. E2, =then=> AREA <-- SMALL

(c) IF V(area) .GE. E2, AND .LT. E3, =then=> AREA <-- MEDIUM

(d) IF V(area) .GE. E3, AND .LT. E4, =then=> AREA <-- LARGE

(e) IF V(area) .GE. E4, AND .LE. E5, =then=> AREA <-- V.LARGE

The threshold values E1,E2,...,Em,...,En are stored in the LTM as constraint knowledge.

The second set of rules (group (b) above) is responsible for creating meaningful symbolic descriptions of the static, incremental, and global changes of the object. Each of these processes may involve rules related to shape, structure, or motion. We note that all of the data manipulated at this stage will be in symbolic form.

The function of these rules in the motion understanding system is similar to that of constructing a sentence in the natural language. Examples are:

```
RULE(3.3): IF    SMALL(RELATIVE-AREA) .AND. SHORT(BASE-LINE),
           =then=> DESCRIPTION .EQ. PSEUDOPOD CANDIDATE.

RULE(3.4): IF    VERY-SHORT(DISPLACEMENT(i,i+1))
           =then=> DESCRIPTION .EQ. STATIONARY.

RULE(3.5): IF    VERY-LONG(DISPLACEMENT(i,i+1))
           =then=> DESCRIPTION .EQ. ARTIFACT.
```

These examples demonstrate the use of the representational rules at the static and incremental levels of description. In corporating these, another function is to generate the global features of the cell and its motion. This task may be accomplished in two steps. The first is concerned with the detection of all the frames in which a change has occurred. These are referred to as "key frames" (KF) [Burtynk and Wein, 78], and are described by rules based on

symbolic qualifiers. The set of key frames may be defined as:

$$\{KF\} = \{KF1, KF2, \ldots, KF_j, \ldots\}. \qquad (3.35)$$

Thus, for example, if a specific property has the same qualifier for a sequence of images, then that property may be described by the same global descriptor. The frames in which the qualifier has changed (beginning and end), are assigned as key frames. This strategy may be modeled by the following rule:

RULE(3.6):

IF Q(Pi,t1) .EQ. Q(Pi,t2) .EQ. ... Q(Pi,tk) .NE. Q(t(k+1)) =then=> Q(T1k) .EQ. Q(t1) .AND. $KF_j$ .EQ. k.

Note that T1k is the period which includes the samples {t1,t2,...,tk} and $KF_j$ is the key frame number j which occurs at time tk. As with other variables, T1k may be normalized and described symbolically as VERY SHORT, SHORT, MEDIUM, LONG, or VERY LONG.

The above is an example of a simple rule, based only on the dynamic data of the different properties. The second step in providing a useful global description, is concerned with distinguishing the significant changes from the irrelevent or noisy ones. In this case, a more sophisticated approach would utilize the dynamic description of the different properties, in conjunction with logical knowledge constraints. For example, an inference process could eliminate the very short events. These are usually

caused by noise or random changes due to undesirable experimental conditions. Thus, suppose a specific property Pi is described through three sequential periods Q1(Pi,T01), Q2(Pi,T12), and Q3(Pi,T23). If the time T12 is very short relative to both T01 and T23 (T01,T23 >> T12), then an inference process might eliminate T12 by one of two actions: first, by merging all three periods T01,T12,T23 into one (T03), or second, by merging T12 either to T01 or T23. Thus, if the cell was described in three sequential periods as SMALL, VERY SMALL, and SMALL, then a merging of these periods would result in SMALL as the description. This may be achieved by the following rule:

RULE(3.7): IF (T01 .GTT. T12 .LTT. T23) .AND.
(Q1 .EQ. Q3 .AND. Q1 .NE. Q2),
=then=> MERGE THE THREE PERIODS INTO Q13(Pi,T03)

where T03 = T01 + T12 + T23, and Q13 = Q1 = Q3.
Figure (7.5a) illustrates the function of this rule.

A second action, in which T12 may be merged to either T01 or T23, is based on logical inference. For example, if the length of the cell was described in three sequential periods as being SHORT, VERY SHORT, MEDIUM, then one may logically deduce that it has changed from SHORT to MEDIUM, if the description of the very short period (T12) is the same as that of either the preceding or following period. Thus, if the description of the VERY SHORT period is closer to the SHORT period rather than to MEDIUM period, period T12

may be merged to T01. This logical deduction may be accomplished by the following rule:

RULE(3.8):

IF (T01 .GTT. T12, .AND. T12 .LTT. T23)
.AND. (Q1 .GT. Q2 .AND. Q2 .LT. Q3 .AND. Q1 .LT. Q3),
.OR. (Q1 .LT. Q2 .AND. Q2 .GT. Q3 .AND. Q1 .GT. Q3),
=then=> MERGE PERIODS T01 AND T12 INTO Q12(Pi, T02)

where T02 = T01 + T12, and Q12 = Q1. Figure (7.5b) illustrates the function of this rule. Other examples of similar rules are shown in Figures (7.5c-7.5g).

The third set of rules (group (c) above) is responsible for integrating these different descriptions into a final coherent characterization. In this sense, its task is similar to that of a human. The latter studies and analyzes the data by observing the cell in order to understand and characterize its dynamic behaviour. Below are examples of this set of rules, which mainly depend on the expert model. The general format is given by

RULE : IF DESCRIPTION ==then==> CHARACTERISTIC

For example, if the total displacement of the cell in the main directions (EAST, EAST-NORTH, ...) is given as:

{TOTAL DISPLACEMENTS} = {TD1, TD2,..., TDi,..., TDm}, (3.36)

where TDi is the total displacements in direction (i), then we have:

RULE(3.9): IF TDi .GTT. TDJ (J=1,2,...,m)

=then=> CHARACTERISTIC . EQ. DIRECTIONAL LOCOMOTION.

RULE(3.10): IF DIRECTIONAL LOCOMOTION
. AND. CELL DIRECTION . AE. BACTERIA LOCATION
=then=> CHARACTERISTIC . EQ. POSITIVE CHEMOTAXIS.

RULE(3.11): IF PSEUDOPOD IS GROWING
. AND. PSEUDOPOD DIRECTION . EQ. CELL DIRECTION
=then=> CHARACTERISTIC . EQ. DOMINANT PSEUDOPOD.

The above show how the global cell locomotion may be characterized as positive chemotaxis, or how a pseudopod may be described as dominant. Of course, different descriptions are also possible. For example, the global cell locomotion may be described as being VERY STRONG NEGATIVE CHEMOTAXIS to VERY STRONG POSITIVE CHEMOTAXIS, or a pseudpod may be classified as being NOT DOMINANT or VERY DOMINANT. An example of the global locomotion description is given in Description (6.1), Description (7.1) summarizes the global changes in the cell membrane shape, and Description (8.3) presents the final characterization of some pseudopods.

## 3.4 CONTROL STRUCTURE RULES

Constructing a knowledge-based system with different levels of processing, which utilize multiple sources of knowledge organized at different levels of description, requires an efficient control structure. Two strategies for defining control structure have been discussed in the

literature: model-driven and data-driven. In the first, system activities are controlled by the existing knowledge, whereas in the second, it is the data that dominates. The mechanism we are proposing for this motion understanding system is a rule-based structure which is both model and data-driven.

The model-driven control rules are responsible for activating, deactivating, and scheduling the different processes and representational rules, as well as starting and stopping the system according to a predefined (by the model) hierarchical or sequential order. These rules are executed hierarchically, and in sequential order within the same level of the hierarchy.

The high level rules in the hierarchy are concerned with starting and stopping the system, as well as activating the main analysis stages. For example;

| CONDITIONS | ===then===> | ACTIONS |
|---|---|---|
| Completed Stage | | Stage to be Activated |
| START | ---> | STATIC SCENE ANALYSIS |
| STATIC SCENE ANALYSIS | ---> | INCREMENTAL CHANGE DETECTION |
| INCREMENTAL CHANGE DETECTION | ---> | GLOBAL ANALYSIS |
| GLOBAL ANALYSIS | ---> | CHARACTERIZATION |
| CHARACTERIZATION | ---> | STOP |

The intermediate level control structure rules are divided into groups corresponding to the main analysis stages of the system. Each group is executed in a special order to activate the different computational processes required for the corresponding stage. For example, STATIC

SCENE ANALYSIS is given by the following:

| CONDITIONS<br>Completed Processes | ===then===> | ACTIONS<br>Process to be Activated |
|---|---|---|
| START | ---> | INITIALIZATION, DIGITIZATION |
| DIGITIZATION | ---> | SEGMENTATION |
| SEGMENTATION | ---> | BOUNDARY AND MAIN FEATURES |
| BOUNDARY AND MAIN FEATURES | ---> | POLYGONAL APPROXIMATION |
| POLYGONAL APPROXIMATION | ---> | DECOMPOSITION |
| DECOMPOSITION | ---> | STATIC DESCRIPTION |
| STATIC DESCRIPTION | ---> | EXIT |

Finally, low level control rules are associated with each computational process. They specify and schedule the different processors (subroutines) that are neccessary to complete the analysis of the current process. For example, segmentation is defined as a set of actions by the model-driven rules, to be executed in sequential order:

A(0): START SEGMENTATION
A(1): GET THE CURRENT FRAME
A(2): LOCATE WINDOW
A(3): COMPUTE HISTOGRAM
A(4): SELECT THRESHOLD
A(5): COMPUTE BINARY WINDOW
A(6): FILTER BINARY WINDOW
A(7): RECOGNIZE TRACKED CELL
A(8): TRACK BOUNDARY
A(9): COMPUTE MAIN FEATURES
A(10): RETURN

The model-driven rules which control the execution of the above computational processes may be described as:

RULE: IF A(i) completed ==then==> activate A(i+1)

Note that at this low level of analysis, most of the control rules are data-driven, since the next action to be taken is usually dependent on the result of the preceding computation.

The predefined model-driven structure we discussed above may be interrupted at any time during the analysis on the occurrence of specific situations. Examples are mentioned in Section 3.3. This type of control strategy is essential for any multiple level understanding system. For example, the results of higher level processes may be used to remove ambiguous situations, or to improve the analysis results of lower level processes. This is of particular importance in motion understanding, where the dynamic analysis may be utilized to improve object representation in the static images.

Communication between the different processes (at different levels) of the system could be achieved through direct feedback links from the higher levels to the lower ones. However, the direction of this feedback is not known a priori, since it depends on the evaluation of different properties of the images. Therefore, the basic function of the data-driven control structure rules is to direct the flow of the control between the different processes of the system.

The direction of the control may be achieved by specifying the action to be taken, such as the next process or the representational or control rules to be activated. It is important to note that the data-driven rules take precedence over the flow of the analysis. Thus, if a model-driven rule should posses the same conditions as a data-driven rule, the action specified by the former would

have priority. For example, during static scene analysis, the following model-driven rule specifies the next process to be activated after the polygonal approximation is completed:

RULE(3.12): IF POLYGONAL APPROXIMATION IS COMPLETED,
=then=> ACTIVATE POLYGON DECOMPOSITION.

However, the following data-driven rule could change this action:

RULE(3.13): IF POLYGONAL APPROXIMATION IS COMPLETED,
.AND. NCA .LE. 1,
=then=> ACTIVATE STATIC DESCRIPTION.

(NCA refers to the number of convex angles. See Table 3.1).

Thus, by using data-driven control rules, the predefined flow of analysis (POLYGONAL APPROXIMATION --> POLYGON DECOMPOSITION), is changed to (POLYGONAL APPROXIMATION --> STATIC DESCRIPTION. This occurred because the cell in the current frame had a simple shape which could not be decomposed. Another important function of data-driven rules is to influence the flow of control between the different stages of analysis. In this way, top-down feedback could remove ambiguous situations at the lower levels of the data hierarchy.

## 3.5 SUMMARY

To achieve the objectives of motion understanding and description, it is not enough to merely determine the incremental movements or changes that occur between consecutive images. What is required is a system which abstracts a summary description of the global motion characteristics from the multitude of static and incremental data. Development of such a system represents the new direction being taken in the current research in image sequence analysis.

A model for a general dynamic scene analysis system has been constructed. It consists of three basic entities: dynamic data, static data, and a collection of analysis processes. The different types of data which may be manipulated by the system have been classified into: a sequence of images, a group of objects and subobjects, a set of object features, symbolic descriptors, global behaviour characteristics (these are functions of groups of features and descriptors used to describe specific behavioural patterns), and a set of rules, which may be classified into representational rules and control rules.

Based on this model, we have implemented a rule-based image interpretation system for moving cells. The system consists of different cooperating computational processors. Conceptually, two different memories are used, a Short Term Memory (STM) and a Long Term Memory (LTM). Both are

implemented as a relational database. The STM is designed to work as a communication channel for all of the processes. It contains a dynamic record of the instantaneous cell motion, shape, and structural changes, as well as the current global description of the cell behaviour. The LTM data are static, and are implemented as rules. These describe the general model of the morphology of the cells under analysis, as well as control information pertinent to the computational processes. The latter are activated by the control rules throughout the three hierarchical analysis stages: static, incremental, and global. They interact through the STM using the information stored in the LTM, until a complete description of the dynamic cell motion and morphology is obtained.

# TABLE (3.1) DIFFERENT TYPES OF PROPERTIES

## SHAPE PROPERTIES

| PROPERTY | SYMBOL | DEFINITION |
|---|---|---|
| AREA | A | Total number of pixels. |
| PERIMETER | P | Length of the contour. |
| LENGTH | L | The distance between the two farthest points on the boundary. |
| WIDTH | W | The maximum extension of the object normal to the length. |
| ELONGATION | E | The complement of the ratio of the width to the length. |
| CIRCULARITY | C | The ratio of the square of the perimeter to the area. |
| AVERAGE BENDING ENERGY | ABE | The rate of change of the Tangent along the boundary. |
| ANGLE REGULARITY | AR | A measurement of the sum of the differences between the angles of a given polygon and a regular one (equal angles) having the same number of sides. |
| SIDE REGULARITY | SR | The same as above, except the measurement involves the sides instead of the angles. |
| NO. OF POLYGON SIDES | NS | Number of sides in the approximation polygon. |
| LENGTH OF POLYGON SIDE | LS | The length between two sequential vertices. |
| INTENSITY (COLOR) | I | The average intensity of the object. |
| NO. OF CONCAVE ANGLES | NCA | Number of vertices which have an internal angle greater than 180. |

## STRUCTURE PROPERTIES

| PROPERTY | | DEFINITION |
|---|---|---|
| NUMBER OF SUBOBJECTS | NSO | Number of primitive subparts. |
| CONNECTIVE LINE | CL | The distance between the cell centroid and that of a subpart. |
| BASE LINE | BL | The adjacent line between the cell and a subpart. |
| SUBOBJECT AREA | ASO | Area of a subpart. |
| SUBOBJECT PERIMETER | PSO | Perimeter of a subpart. |
| RELATIVE AREA | RA | The ratio of a subpart area to the total cell area. |

## LOCATION PROPERTIES

| PROPERTY | | DEFINITION |
|---|---|---|
| BOUNDARY COORDINATES | BX, BY | The X and Y coordinates of the boundary points. |
| CENTROID (X,Y) | CNTX, CNTY | Centroid coordinates. |
| ORIENTATION | OR | The angle between the main axis of the object and the X axis. |
| CONTAINING RECTANGLE | CRX, CRY | The minimum containing rectangle. |
| FITTED RECTANGLE | FRX, FRY | The rectangle whose main axes are the length and width of the object. |
| POLYGON CENTROID | PCNTX, PCNTY | Centroid coordinates of the polygonal approximation. |
| SUBOBJECT CENTROID | SCNTX, SCNTY | Centroid coordinates of a subpart. |

# MOTION PROPERTIES

| PROPERTY | SYMBOL | DEFINITION |
|---|---|---|
| DISPLACEMENT | DIS | Translation distance. |
| DIRECTION | DIR | Direction of motion w.r.t. X axis. |
| VELOCITY | VL | Ratio of displacement to time. |
| ROTATION | RO | Change in the orientation of the cell |

# Table(3.2)

## Examples of Symbolic Qualifiers of Some Properties, and Their Different Levels (Classes) of Qualification.

| PROPERTY | 1st QUALIFIER | 2nd QUALIFIER | 3rd QUALIFIER |
|---|---|---|---|
| ELONGATION | NOT ELONGATED | SLIGHTLY ELONGATED | ELONGATED |
| CIRCULARITY | NOT CIRCULAR | SLIGHTLY CIRCULAR | ALMOST CIRCULAR |
| A. B. ENERGY | VERY JAGGY | JAGGY | ALMOST SMOTH |
| REGULARITY | VERY IREGULAR | IREGULAR | ALMOST REGULAR |
| SIZE | VERY SMALL | SMALL | MEDIUM |
| MEMBRANE SHAPE | VERY COMPLEX | COMPLEX | ALMOST SIMPLE |
| SPEED | VERY SLOW | SLOW | AVERAGE |
| ACCELERATION | HIGH POSITIVE | LOW POSITIVE | CONST. SPEED |
| LENGTH, TIME, DISTANCE | VERY SHORT | SHORT | MEDIUM |

| 4th QUALIFIER | 5th QUALIFIER |
|---|---|
| VERY ELONGATED | FILAMENTARY |
| CIRCULAR | VERY CIRCULAR |
| SMOOTH | VERY SMOOTH |
| REGULAR | VERY REGULAR |
| LARGE | VERY LARGE |
| SIMPLE | VERY SIMPLE |
| FAST | VERY FAST |
| LOW NEGATIVE | HIGH NEGATIVE |
| LONG | VERY LONG |

## Table(3.3)

## A Set of Operators and Their Definitions.

| OPERATOR | DEFINITION |
|---|---|
| AND | Logical AND. |
| OR | Logical OR. |
| LT | Less than. |
| GT | Greater than. |
| EQ | Equal. |
| LE | Less than or equal. |
| GE | Greater than or equal. |
| NE | Not equal. |
| AE | Approximately equal. |
| GTT | Much greater than. |
| LTT | Much less than. |
| MST | Most. |
| LST | Least. |

# CHAPTER 4

# STATIC SCENE ANALYSIS

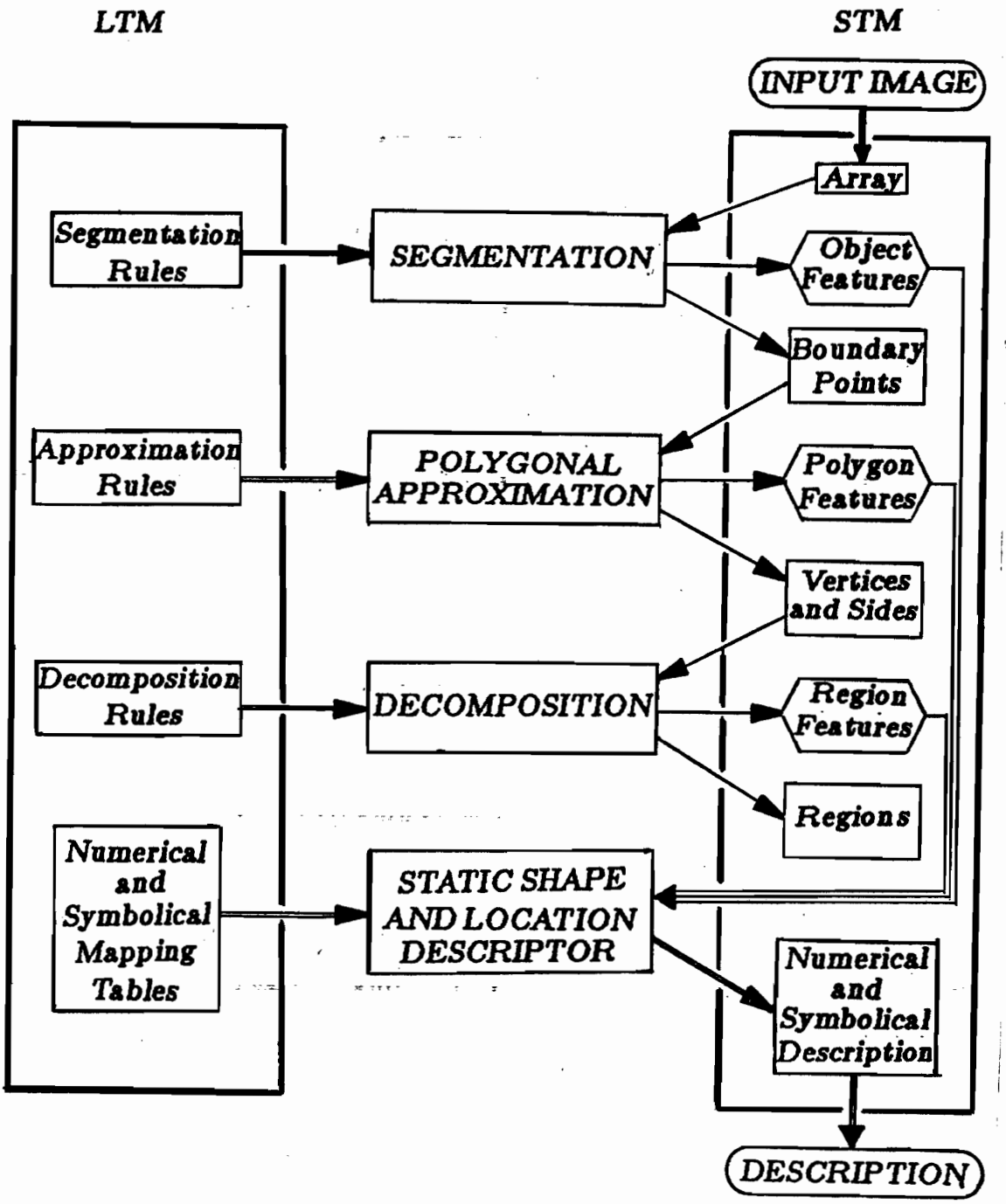
## 4.1 INTRODUCTION

In the preceding chapter, we presented the system and data structure of a system for understanding and describing the dynamic behaviour of a non-rigid moving object. Based on this structure, we have implemented a rule-based system for characterizing the behaviour of a moving cell. The system consists of three basic entities: dynamic data (STM), static data (LTM), and a collection of analysis processes. The latter are designed to perform three stages of analysis: static scene analysis, incremental change detection, and global analysis. This chapter is devoted to describing the input, output, and function of the different processes of the first stage of analysis, that is, Static Scene Analysis.

The static scene analysis stage, in the proposed system, is similar to, and has the same function, as a static image processing system. Thus, the input is a single digital image, and the output is an interpretation and description of the scene. However, in image sequence analysis, the information extracted from the previous frames

of the same sequence may be used to assist the analysis of the current frame. The main objectives of this stage are to identify the desired moving object, segment it, and describe it in each frame of the sequence (see Figure 1.5a). This stage consists of four main processes as shown in Figure (4.1). They include segmentation, polygonal approximation, polygonal decomposition, and description (location and shape of the moving object).

The first process in the static scene analysis stage is concerned with the extraction of the cell under analysis from the input image; this is described in Section 4.2. An algorithm for generating the polygonal approximation of the cell boundaries is described in Section 4.3, as are the advantages of this approximation. Section 4.4 describes the approach of decomposing the cell (which may have a complex shape) into its primitive subparts. A discussion pertaining to the selection of the properties to be measured, and their theoretical definition is given in Section 4.5. Finally, Section 4.6 describes a process which integrates the cell morphology that has been extracted and measured by the previous processes in order to generate a coherent description (numerically and symbolically) of the cell in the current frame.




*Figure(4.1) Processes and data structure of the static scene analysis stage.*

# 4.2 CELL EXTRACTION (SEGMENTATION)

## 4.2.1 Introduction

Segmentation is a basic and common problem in any image processing system. The objective is to divide the input image into regions corresponding to the objects and the background in the scene as perceived by a viewer. Although this problem has different definitions in the literature, all of them have the same argument. For example, Pavlidis has defined it as "the operation of looking at a scene and picking up objects from the background. In such an effort we divide the picture into different parts which have some meaning for the viewer." [Pavlidis, 77]. Whereas, Levine has defined the segmentation stage of a vision system as "At this stage, the input image is divided into regions containing pixels whose primary features such as intensity, hue, saturation, and texture are similar." [Levine, 78].

The final goal of the segmentation is a collection of regions which correspond exactly to the objects in the scene. In some cases, this may be achieved by applying the segmentation operations directly to the input image. In other cases, processes which use external knowledge must be applied to the image after it has been partially segmented. The former is applicable in cases where the image consists of objects superimposed on a uniform background, for example, nonoverlapping blood cells [Youssef, 77]. In this case a simple thresholding process suffices to result in a

complete segmentation of the objects from the background. Utilizing external knowledge is necessary for segmenting complex images, such as a typical suburban scenes, or images containing threee-dimensional effects, i.e. occlusion or shadows. In these cases, the initial segmentation results only in a partial segmentation, where the segmented regions do not necessarily correspond to the objects in the scene. Therefore, the use of a priori knowledge about the scene under consideration is essential.

Segmentation of a scene by the human visual system may be considered as one of psychophysical perception. It involves processes which are not well understood yet [Rosenfeld, 76]. Therefore, there are no criteria to define the successful segmentation and it is not susceptible to a purely analytical solution. However, this problem has been considered in a recent work by [Levine and Nazif, 82].

In image sequence analysis, the segmentation has two main objectives; the first one is to segment the scene into objects and background, and the second is to segment the objects into moving and stationary. Most image sequence analysis systems have accomplished these two objectives separately; segmenting the scene first, then extracting the moving object, using its geometrical properties [Ferrie, 79]. In some cases, the objective was to segment the individual pixels of the image into moving or stationary using pixel to pixel comparison between the sequential frames [Yakimovsky, 75]. Using this methodology, the moving

pixels can be grouped to form regions corresponding to the moving objects [Nagel, ??]. Other techniques in image sequence analysis are based on: first, the initial segmentation of the sequence of frames, then, the utilization of the information extracted from the sequence in order to achieve a complete segmentation of the individual frames. Recently, a knowledge-based system has been developed in order to accomplish both complete segmentation and motion detection simultaneously by utilizing a priori knowledge about the sequence under consideration [Tsotsos, 80].

The input of any motion understanding system is a sequence of two-dimensional digital images recording the object motion to be analyzed. In the case of microscopic cell images, the sequence recording the cell motion may be obtained in various ways; for example, cine film (time-lapse unit), or a TV camera observing live cells in real-time through a microscope, where the images are captured at specific intervals of time. Then the images may be transfered directly to the computer memory, and/or to a video tape. However, the actual data of the analysis processes is a series of two-dimensional images.

Most of the previous techniques for microscopic cell image segmentation are based on the existing contrast between the cells and the background. However, in the case of live cells, the contrast is continualy changing due to two factors: the phase contrast of the cell photography and

the three-dimensional motion of the cell. The latter may also cause a change in the cell intensity distribution (gray-value distribution of the cell region). Thus, the difficulties of segmenting live cells in motion may be summarized as:

(1) Phase contrast photography causes a gradient shadow area between the cell and the background. Consequently, the exact boundary of the cell is hidden within this shadow area. Therefore, an inference process for labelling the pixels of this area as belonging to either the cell or the background may be necessary.

(2) The three-dimensional changes in the cell structure continually cause random changes in the cell intensity distribution over time; consequently, the cell-background contrast will change. Therefore, we cannot use the same threshold(s) for segmenting the cell in the different frames of a sequence. A dynamic thresholding scheme may be necessary.

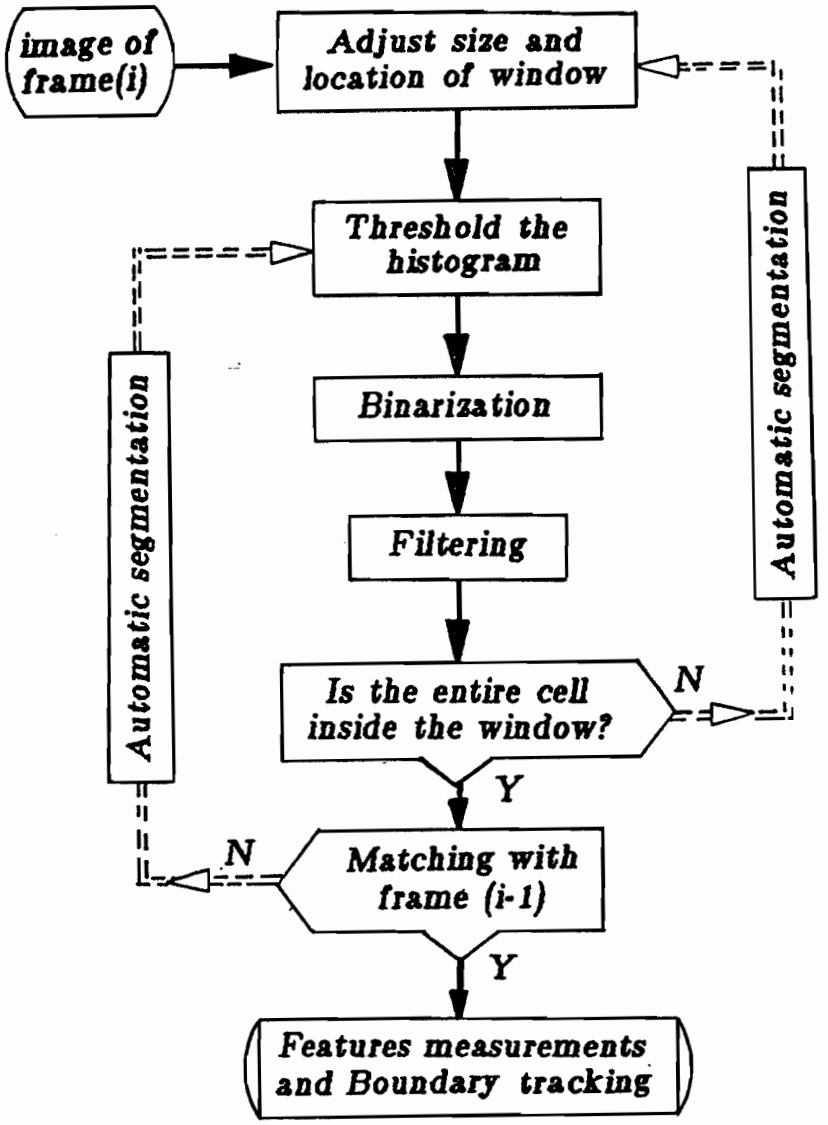
Besides the above difficulties, in the case of real-time analysis, where the images must be captured at short time intervals, the segmentation should be a fast process. Therefore, most of the sophisticated segmentation techniques are not practical for real-time application, since they require long and expensive computation. Also, in the analysis of the moving cells, the task of the segmentation may not only include the extraction of the cell from the background, but may also include the segmentation

of the different parts of the cell (nucleus and cytoplasm).

An algorithm for the extraction of a moving cell is implemented in our work. It consists of a collection of computational subprocesses cooperating through a common dynamic data memory, the (STM). The different subprocesses and data structures of this segmentation algorithm are shown in Figure (4.2). This includes the following subprocesses: cell windowing, thresholding, binarizing, filtering, matching, and boundary tracking. The activation, deactivation and scheduling of the different subprocesses is accomplished by control rules stored in the LTM. These subprocesses may be executed in iteration until a satisfactory segmentation is obtained. The execution and the number of iterations is controlled by the LTM rules. The actions of these rules are based on the dynamic data (STM) pertaining to the cell morphology in the previous frame, the resulting data from the different segmentation subprocesses of the current frame, and the LTM constraint knowledge. The objective as well as examples of the input and output of each subprocess is given below.

## 4.2.2 Cell Window

The first step towards cell extraction is to locate and adjust the size of the window containing the cell under analysis. The objective is to minimize the search area to that which only includes the tracked cell with a reasonable background. Although it is difficult in many cases to




*Figure(4.2) Algorithm for manual and/or automatic segmentation of cell images.*

isolate the tracked cell within the window, the candidate cells will at least be restricted to those within the window. Examples of typical input images are shown in Figure (4.3). Each image consists of 128x128 pixels, each pixel having 6 bits of gray level information. The images include neutrophil cell(s), red cells, and a piece of bacteria located in the lower left side of the image.

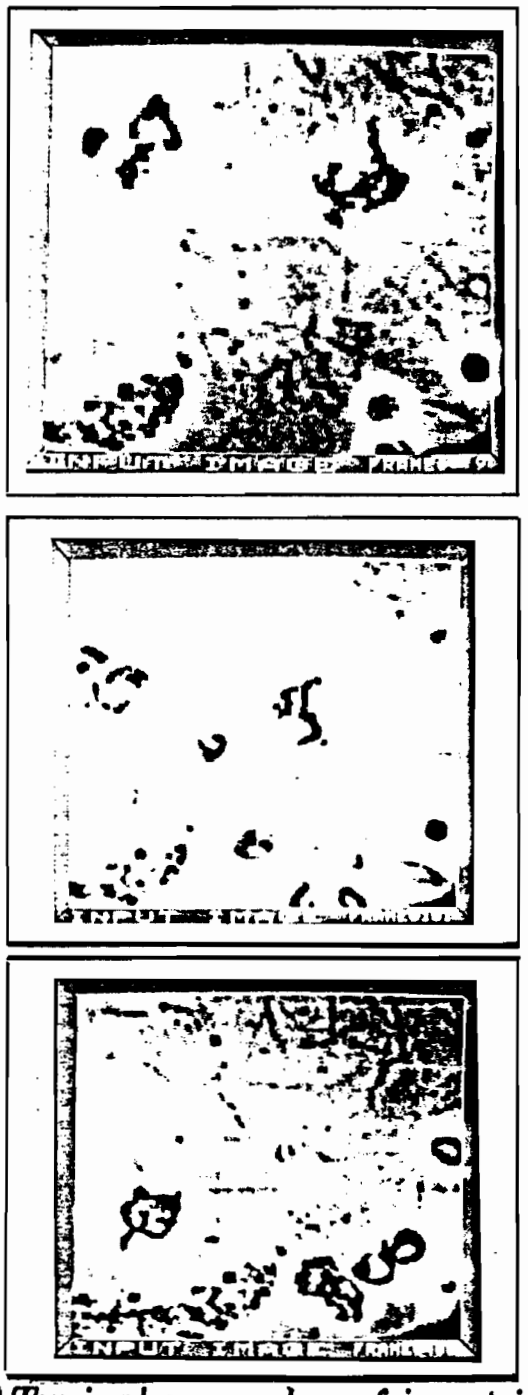
In our system the size and location of the window may be adjusted either interactively or automatically. In the interactive method, the user specifies the location and the size parameter of the window through a keyboard or by using a joystick. In the automatic method, the initial window parameters in a frame are determined by the system, utilizing the location and size of the cell in the previous frame as well as the LTM constraint knowledge (maximum displacement of the cell between two sequential frames). Thus the initial location of the window in frame(i) is determined from the window parameters in frame(i-1) via:

$$\text{centroid} = X(i-1),\ Y(i-1) \qquad (4.1)$$

$$\text{length} = 2S + L(i-1) \qquad (4.2)$$

$$\text{width} = 2S + W(i-1) \qquad (4.3)$$

where X(i-1),Y(i-1) are the X,Y coordinates of the cell centroid at frame(i-1), S is the maximum possible displacement of the cell between two sequential frames, L and W are the maximum extension of the cell in the Y and X directions, respectively in frame(i-1).



*Figure(4.8) Typical examples of input images in a sequence, recording the dynamic movements of a neutrophil cell.*

The above parameters are the best initial expectation. However, the window may be shifted, and the size may be adjusted according to the data extracted from the initial segmentation. Figure (4.4) shows the selected window from an input image.

### 4.2.2.1 Histogram

The histogram is a well-known technique for transforming two-dimensional images into scalar vectors. Thus, the histogram (H) is a vector which represents the frequency distribution of the gray values of an image or window of the image,

$$(H) = (G0, G1, G2, \ldots, Gk, \ldots, G(m-1)) \qquad (4.4)$$

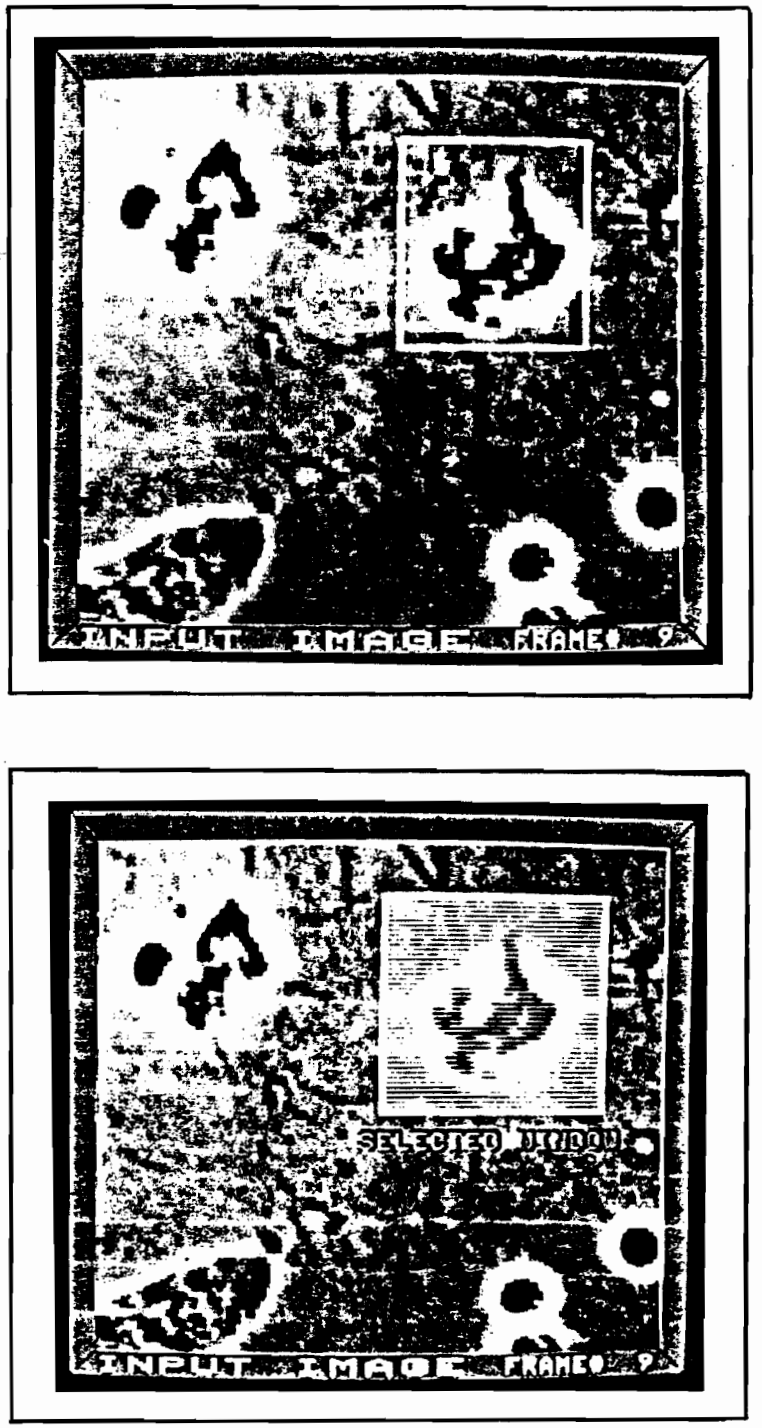
where Gk is the number of pixels which have gray-value equal to k, and m is the number of gray levels. The histogram may be computed as follows:

(1) Set G0, G1, ...,Gm = 0,

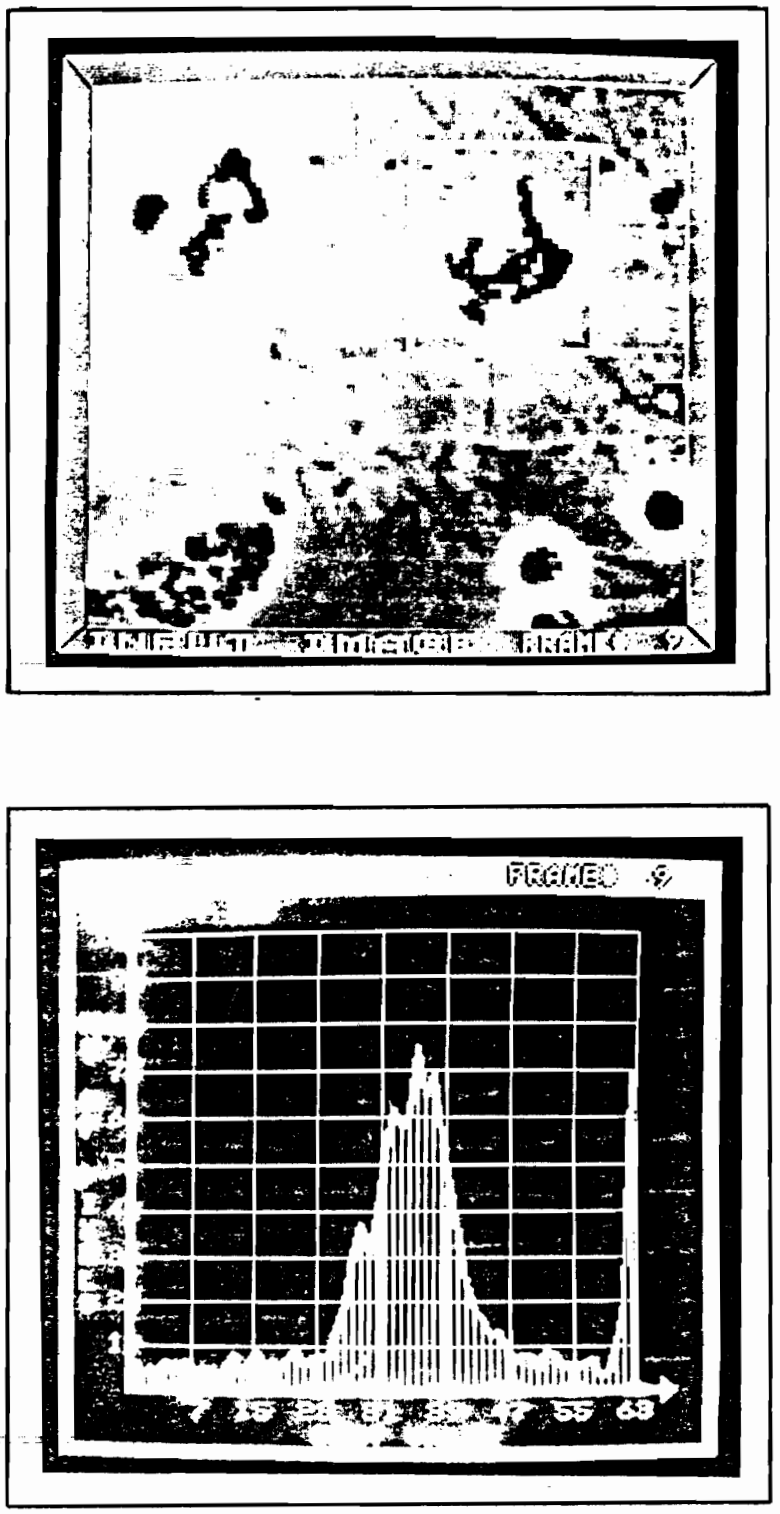
(2) scan the image and for each pixel:

(3) If f(x,y) = k, then Gk = Gk + 1.

The histogram may be plotted as shown in Figure (4.5), where the X axis represents the gray-value, and the Y axis represents the number of pixels. For example, the histogram for a blank image (all the pixels having the same gray-value, say g) is a vertical line located at g. Histogram analysis is an established method for image segmentation where the image contains contrasted objects on a uniform background. The histogram in this case will




*Figure(4.4) Selecting the window containing the cell under analysis.*




*Figure(4.5) The computed histogram of the selected window.*

include two "peaks" representing the objects and the background. The "valley" between the two peaks represents the gray-value which may be used as the threshold to segment the objects from the background. This simple segmentation can be achieved only for simple images. However, in most typical images the resulting histogram is more complex. It either contains a large number of peaks which do not correspond to specific objects, or it does not include clear peaks. For example, in Figure (4.5) in spite of the fact that a human can separate several objects from the background, the histogram contains only one major unconstrained peak, and that peak is not well-defined.

In cases as shown in Figures (4.5), the histogram as it is, may not be useful for direct thresholding. However, a histogram analysis and modification may be helpful for an initial-guess segmentation. In general, most of the histogram information may be extracted from its maxima points. Therefore, the histogram may be represented as a set of extrema {K} (peaks and valleys) as:

$$H = \{K\} = \{K1, K2, \ldots, Kj, \ldots, Kv\} \qquad (4.5)$$

where Kj is a gray-level (i) such that:

Gi number of pixels at level(i), and

G(i-1) < Gi > G(i+1) "peak" or

G(i-1) > Gi < G(i+1) "valley".

This information may be utilized to find the appropriate thresholds for the segmentation.

## 4.2.3 Thresholding

The task of the thresholding subprocesses is to analyze the histogram (H) in order to find the threshold(s) (gray-value(s)) which may be used to segment the cell from the background, and/or the cell into cytoplasm and nucleus. From Figure (4.5) we can see that the histogram includes only one unconstrained mode. That means both the cell and the background have regions which exhibit the same gray-level. However, the original image (Figure 4.5a) shows that the cell is darker than the background. Therefore, a good initial guess is to consider the maximum frequency (peak) as threshold value (Gt). This may be computed as follows:

$$Gt = Max\ [G0,\ G1,\ \ldots,\ G63] \qquad (4.6)$$

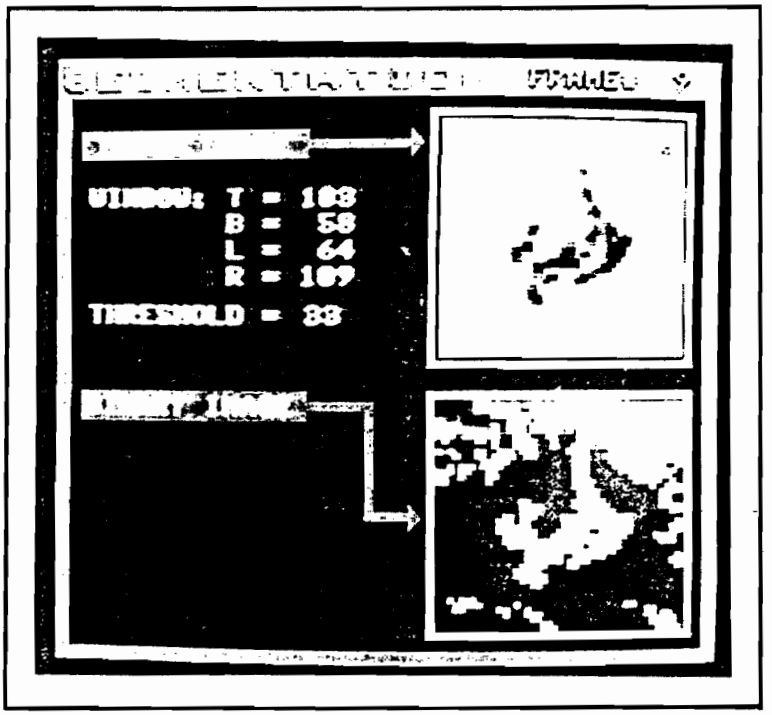
Then the binary image may be obtained as:

IF f(x,y) .GE. Gt, THEN b(x,y) = 0

IF f(x,y) .LT. Gt, THEN b(x,y) = 1

where f(x,y) is the intensity image and b(x,y) is the binary image. Figure (4.6) shows the resulting binary image.

The above method gives a satisfactory segmentation if the cell has a homogeneous gray level. However, as we can see in Figure (4.6) most of the inside region of the cell is segmented as background. This is because the inside regions




*Figure(4.6) Binarization using maximum gray level frequency (highest peak in the histogram) as a threshold value.*

of the cell (nucleus area) have a similar intensity to the background (both are represented by the peak of the histogram). In order to segment the cell as one region from the background, we propose the following modification:

## AVERAGE THRESHOLDING

The proposed modification is based on dividing the histogram into three regions: the low gray-level region (0-Gc) which mainly represents cell pixels, the high gray-level region (Gb-63) mainly represents background pixels, and the medium gray-level region (Gc-Gb) which partially represents cell pixels and partially background area. Figure (4.7) illustrates these three regions. In order to compute the thresholds for these regions, we assume that the maximum frequency Gt (highest peak in the histogram) divides the image into two regions: (a) dark (0-Gt), and (b) bright (Gt-63). The threshold values Gc and Gb are then computed as the average gray level of each of these regions, respectively. This can be computed by scanning the cell window, and assigning each pixel f(x,y) to one of the regions as follows:

(1) set Gtc = 0 and Gtb = 0

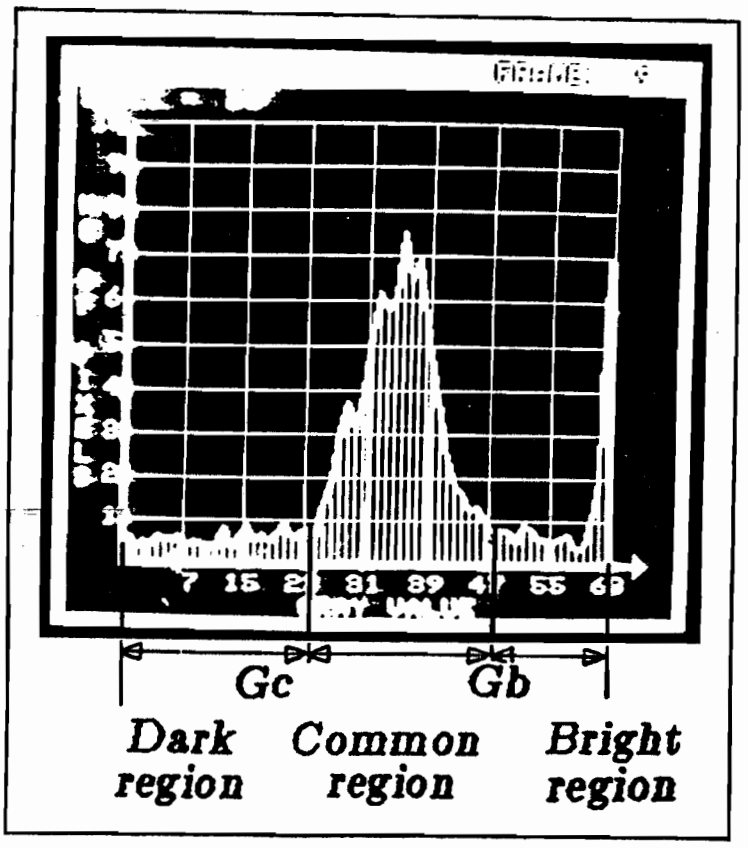
(2) IF f(x,y) .GE. Gt, THEN Gtc = Gtc + f(x,y), AND Nc = Nc + 1

(3) IF f(x,y) .LT. Gt, THEN Gtb = Gtb + f(x,y), AND Nb = Nb + 1

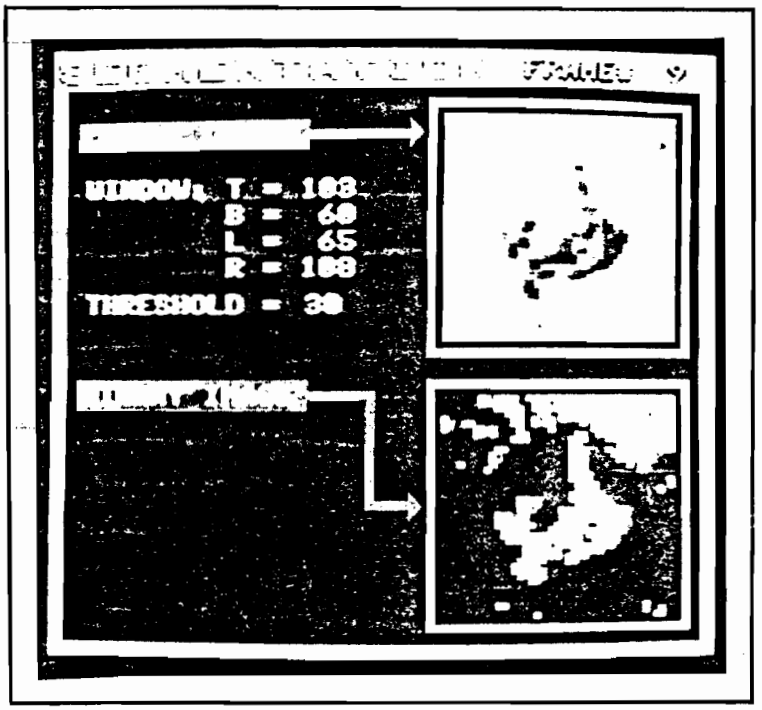
(4) Gc = Gtc / Nc

(5) Gb = Gtb / Nb

where Gtc and Gtb are the sums of the gray levels in the




*Figure(4.7) Dividing the histogram of the cell image into three main regions.*



*Figure(4.8) Binary image obtained by averaging threshold method.*

dark and bright regions, respectively, Nc and Nb are the number of pixels in each region. Using the threshold values Gc and Gb, the binary image can be obtained by scanning the cell window and labeling each pixel as cell (1) or background (0) as follows:

(1) IF f(x,y) .LE. Gc, THEN b(x,y) = 1,

(2) IF f(x,y) .GE. Gb, THEN b(x,y) = 0,

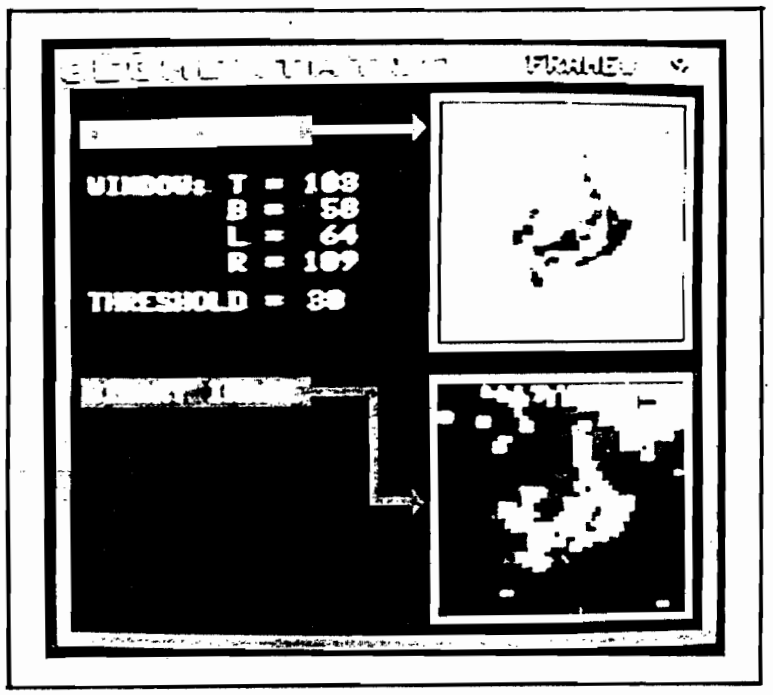
(3) IF (f(x,y) - Gc) .GT. (Gb - f(x,y)), THEN b(x,y) = 1

(4) IF (Gb - f(x,y)) .GE. (f(x,y) - Gc), THEN b(x,y) = 0

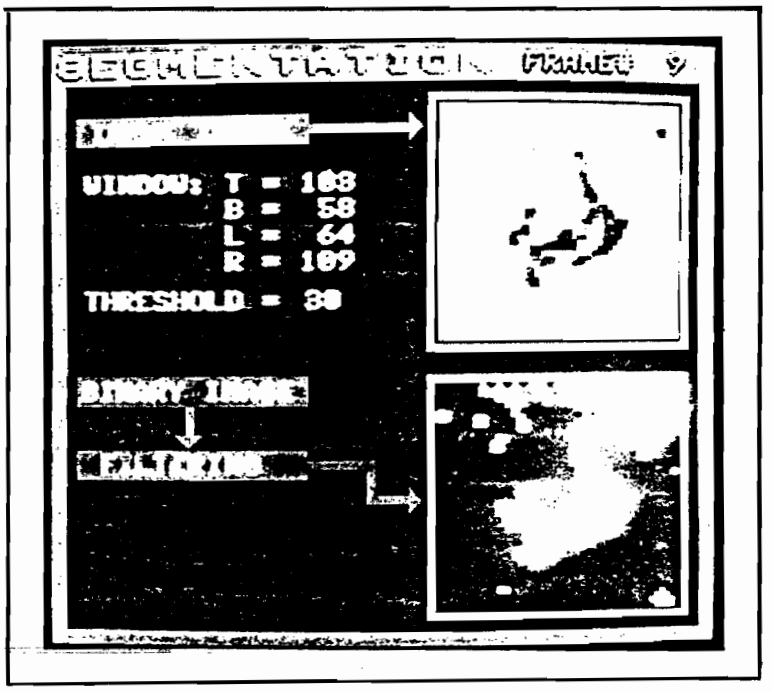
In this way, pixels with gray levels between Gc and Gb are labeled as cell or background according to whether they are closer to Gc or Gb, respectively. Figure (4.8) shows the resulting binary image using the above thresholding method. The result of this thresholding technique is a good initial segmentation, however, there are still some pixels within the cell region which are segmented as background and vice versa (because of the image conditions). This noise may be removed by a filtering operation, which is described in the next section.

## 4.2.4 Filtering

The input of the filtering subprocess is a noisy binary image (Figure 4.9a), and the objective is to reduce the noise by relabelling the noisy pixels (0->1 or 1->0). This objective may be achieved in two steps. In the first step, all single pixels and thin regions (regions of one pixel width) are removed. This is achieved by using the four




(a)



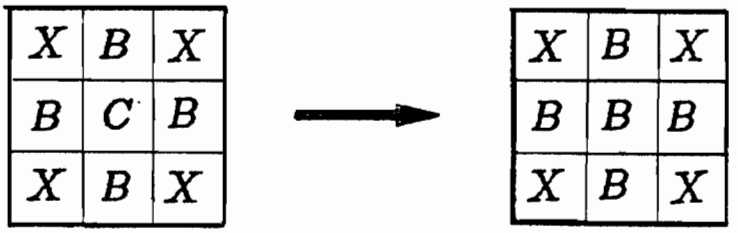

(b)

*Figure(4.9) Removing noise by filttering (a) input (b) output.*

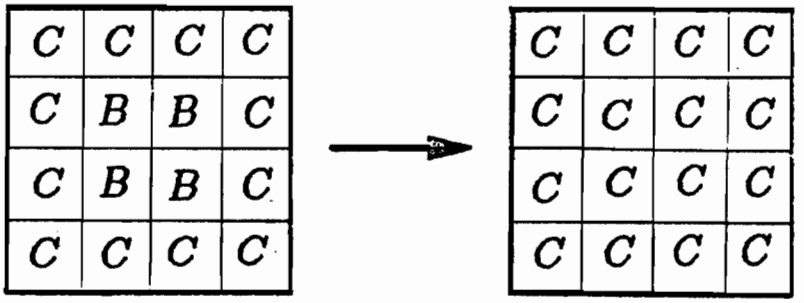
connection criterion shown in Figure (4.10a). For example, if a specific pixel is labeled as cell and its four connected neighbors are labeled as background, then the label of this pixel is changed to background, and vice versa. The second step is based on an inference procedure, which utilizes constraint knowledge pertaining to the cell structure. For example, if a small background region is enclosed by a larger cell region, then this region will be labeled and merged to the cell region. Figure (4.10b) illustrates an example of this case. Figure (4.9b) shows the output of filtering the noise in Figure (4.9a).

## 4.2.5 Cell Selection And Matching

The output of the segmented binary image may contain more than one region. In some cases, there are two regions the cell and the background (the optimum result). However, in most cases, the segmented image may contain more than two regions (where other cell(s) or part of cell may enter the search window), and/or regions of the background are labeled as cell regions. In order to select the region representing the cell under analysis, we use the cell selection and matching process, which has two main functions: (a) to select the cell under analysis, and, (b) to measure the difference between the properties of the selected cell from the current frame and the cell in the previous frame, thereby correcting the segmentation process (see Figure 4.2).




*(a) Removing single pixels and thin regions.*



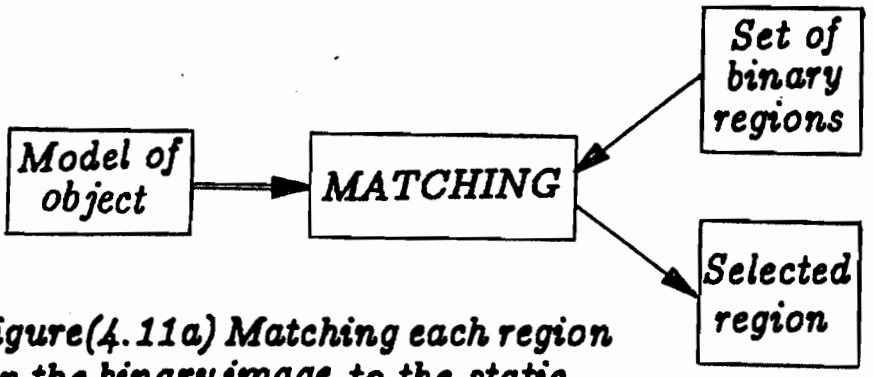

*(b) Using cell shape and structure knowledge to remove noisy regions.*

*C cell pixel B background pixel X any*

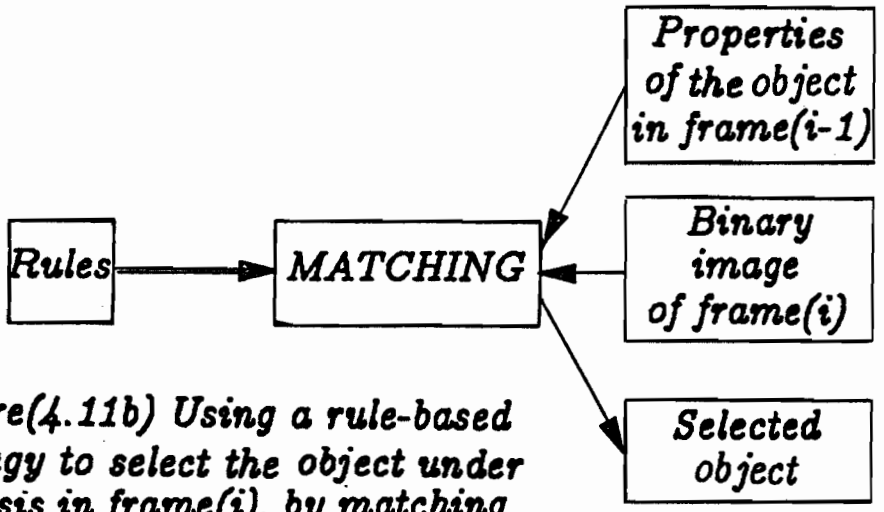
*Figure(4.10) Filtering operations.*

Cell selection and matching may be considered as a pattern recognition process, in that an object which exhibits specific properties and/or matches a specific pattern (model) is to be selected among a set of objects. Figure (4.11a) shows a block diagram of this process for static scene or image sequence analysis of rigid moving objects. In this case, the shape and/or the structure of the moving object does not change within the sequence, therefore a prototype model may be used for selecting the region that corresponds to the moving object among the set of segmented regions. On the other hand, in the case of a non-rigid moving object (such as for example, blood cells), there is no standard model of the moving object, because its shape and/or structure are changed continually within the sequence. Therefore, selecting the cell under analysis in the current frame is based on matching to the cell morphology and location from the previous frame. The properties used for the matching as well as the maximum allowable changes in the values of these properties, between the sequential frames, are specified by the LTM rules. This strategy is illustrated in Figure (4.11b).

The theoretical aspects and quantification of the cell matching will be described in detail in the following chapter (Incremental Change Detection and Quantification). The matching of the cell may be quantified and normalized between zero and one, where the less is the matching, the smaller is the value (0 = no matching, and 1 = identical




*Figure(4.11a) Matching each region in the binary image to the static model of the desired object.*




*Figure(4.11b) Using a rule-based strategy to select the object under analysis in frame(i), by matching the different regions in frame(i) to the object properties in frame(i-1).*

*Figure(4.11) Matching procedures: (a) static image processing and rigid moving object. (b) non-rigid moving object.*

matching). However, because of the continual change, we do not expect an exact match between sequential frames. The acceptable percentage of matching (Em) depends on the maximum possible incremental change in the cell prperties and may be specified by the LTM rules. For example:

RULE(4.1): IF M(Ok, ti, ti+1) .GE. Em

==then==> Oi <--- A CELL CANDIDATE.

where M(Ok, ti, ti+1) is the matching value of object Ok in frame (i+1) to the cell in frame (i).

This rule may result in one of three possibilities:

(a) Only one region categorized as a cell candidate. This is the optimum and desired situation, it occurs in most cases.

(b) More than one region is categorized as cell candidates. In this case, we select the one that exihibits the best matching. This situation occurs if there is more than one cell in the search window.

(c) No regions are interpreted as cell candidates. This case arises due to one or more of the following situations:

(i) Undesired experimental conditions, such as moving the slide containing the cells under the camera.

(ii) Unexpected cell displacement which locates the cell outside the window (completely or partially).

(iii) Error in the initial segmentation due to the three-dimensional motion of the cell.

The actions to be taken by the system in this case, depend on the incoming conditions. Examples of these actions are:

A1: correct threshold values.

A2: shift window.

A3: expand window.

A4: stop processing.

The output of the cell extraction consists of a binary image (0's for background and 1's for cell elements) as shown in Figure (4.9). This image represents the input for the final step of the segmentation, which is concerned with boundary tracking and feature measurements. The objective is to transform the two-dimensional binary image into a scalar vector (boundary coordinates and property values) representing the cell in the current frame.

## 4.2.6 Boundary Tracking

The boundary points are those elements of the object which are adjacent to the background. The boundary point can be defined based as either four or eight connected. The latter can be defined as: if any of the cell pixels has one of its eight neighbors as background, then this pixel is a boundary point. In the case of four connected boundaries, an object pixel is a boundary point if any of its neighbors in the X or Y directions is background. This definition may be modeled as:

RULE(4.2): IF B(Xi,Yi) .EQ. 1 .AND. B(Xi+j,Yi+k) .EQ. 0
==then==> B(Xi,Yi) IS A BOUNDARY POINT.

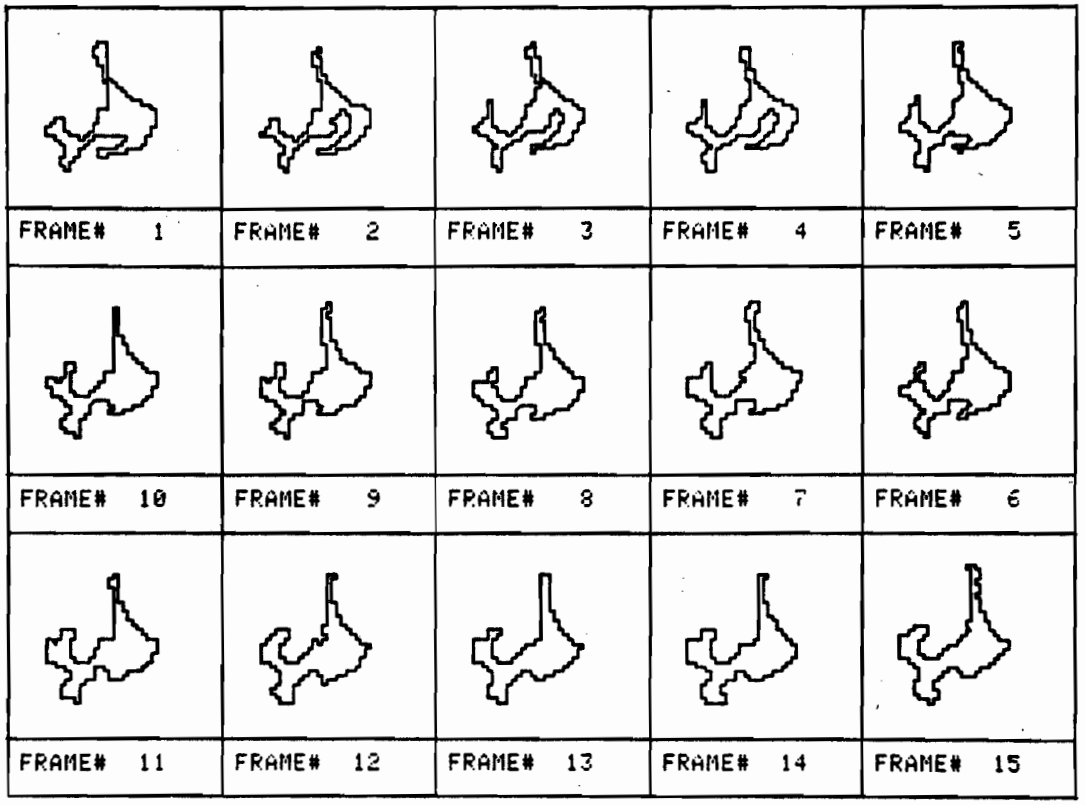
for eight connected boundaries:

$$j,k = (-1,-1),\ (-1,0),\ (-1,1),\ (0,-1),$$
$$(0,1),\quad (1,-1),\ (1,0),\ (1,1)$$

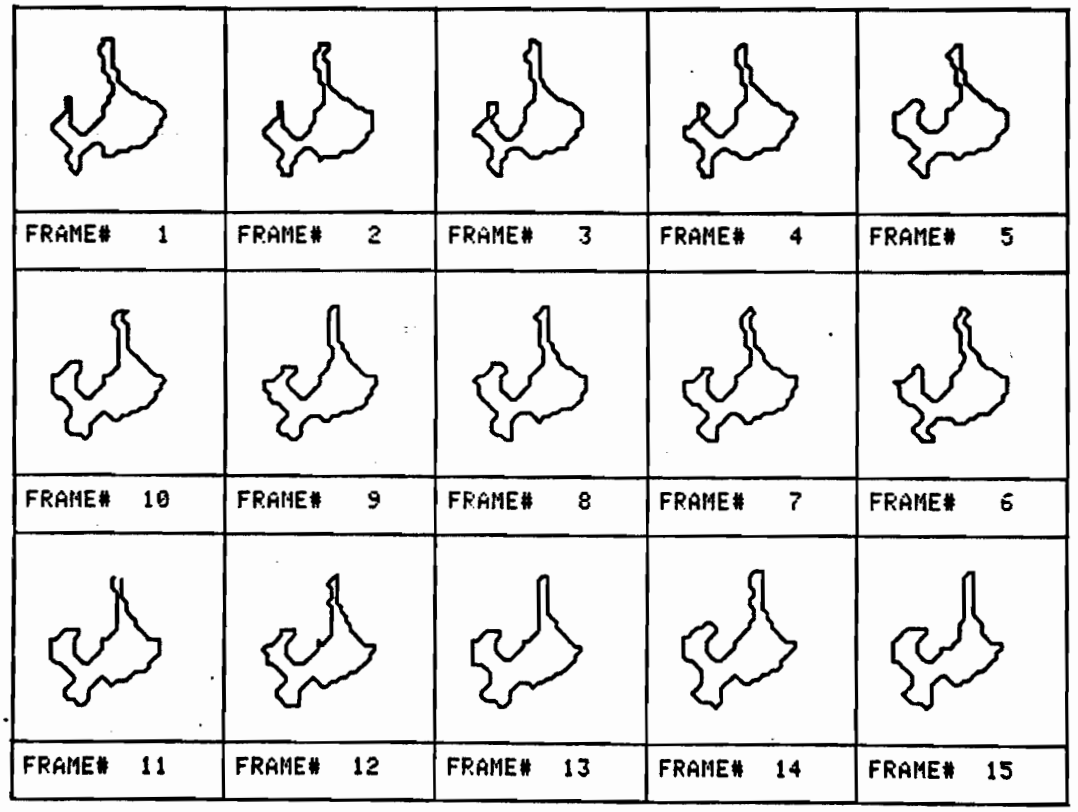
for four connected boundaries:

$$j,k = (-1,0),\ (1,0),\ (0,-1),\ (0,1)$$

The different boundary points {B} may be found by scanning the binary image and extracting the points which satisfy the above rule. However, a faster process is to continue scanning until the first point B1 is found. Then, a tracking algorithm (boundary follower) may be used to find the remaining points {B2, B3, . . . . , B(b+1)}; where B1 = B(b+1), and b is the number of boundary points. The boundary tracking method is not only faster than the scanning, but also represents the boundary as a sequence of points (similar to the chain code method), which is more useful for retrieval purposes. The eight connected method results in contours that are more jagged and have more boundary points than those resulting from the four connected boundaries. Figures (4.12a) and (4.12b) show examples of the boundary points of the cell shapes in a sequence of frames, obtained using the eight and four connected boundaries respectively. From these figures one can see the advantages of using four connected method for this class of objects.




*Figure(4.12a) Eight connected boundaries of a sequence of cells.*




*Figure(4.12b) Four connected boundaries of a sequence of cells.*

PROPERTY MEASUREMENT

The complete description of the selected properties and their theoretical definitions will be presented in Section 4.5. However, at the segmentation level, the following main properties of the cell in the current frame may be computed: area, centroid, average intensity, perimeter, length, width, and orientation.

The preceding sections described three sequential subprocesses for cell segmentation (adjusting size and location of cell window, thresholding, and cell selection and matching). Although these processes may be be executed interactively by the user, they have been designed and implemented so that they may be executed automatically by the model-driven rules. Also, the segmentation may be corrected by iterative execution of these (or some of the) processes according to the specification of the data-driven rules (Figure 4.2). A description and summary of the complete automatic segmentation procedure is given in the next section.

## 4.2.7 Automatic Segmentation

The different subprocesses of the segmentation and cell extraction, which have been described in the previous sections, may be executed and their parameters (window size, location, threshold values,...) adjusted automatically by

model and data-driven control rules. These specify and schedule the different processors (subroutines) that are necessary to complete the analysis of the current process. For example, segmentation is defined as a set of actions by the model-driven rules, to be executed in sequential order:

A(0): START SEGMENTATION
A(1): GET THE CURRENT FRAME
A(2): LOCATE WINDOW
A(3): COMPUTE HISTOGRAM
A(4): SELECT THRESHOLD
A(5): COMPUTE BINARY WINDOW
A(6): FILTER BINARY WINDOW
A(7): RECOGNIZE TRACKED CELL
A(8): TRACK BOUNDARY
A(9): COMPUTE MAIN FEATURES
A(10): RETURN

The model-driven rules which control the execution of the above computational processes may be described as:

RULE(4.3): IF ACT(i) completed ==then==> activate ACT(i+1)

Note that at this low level of analysis, most of the control rules are data-driven, since the next action to be taken is usually dependent on the result of the preceding computation. Consider a typical rule in ACT(5). If the extracted cell touches any of the window borders, the window should be shifted until the complete cell is located inside the window. This situation can be controlled by the following rule:

RULE(4.4):

```
IF XMN .EQ. XLFT  ==then==> XLFT <-- XLFT - SHIFT
IF XMX .EQ. XRIT  ==then==> XRIT <-- XRIT + SHIFT
IF YMN .EQ. YBTM  ==then==> YBTM <-- YBTM - SHIFT
IF YMX .EQ. YTOP  ==then==> YTOP <-- YTOP + SHIFT
```

IF RULE(4.4) IS ACTIVATED ==then==> ACTIVATE (A3 --> A7)

where XMN, XMX and YMN, YMX are the minimum and maximum location of the cell, and XLFT, XRIT, YBTM, and YTOP are the left, right, bottom, and top dimension of the window. SHIFT is a constant value that defines the shifting distance of the window. An example of a rule which control the computation of the threshold value Gt is related to ACT(7). The action is based on the computation of the difference between the area of the segmented cell in the current frame and the previous one. The difference in area dA(i,i+1) can be computed and normalized as follows:

$$dA(i,i+1) = \frac{A(i) - A(i+1)}{A(i)} \quad . \qquad (4.7)$$

This value is used by the following rule, which utilize the constraint knowledge about the maximum change in cell size in the time between two sequential frames, in order to correct the estimated threshold as follows:

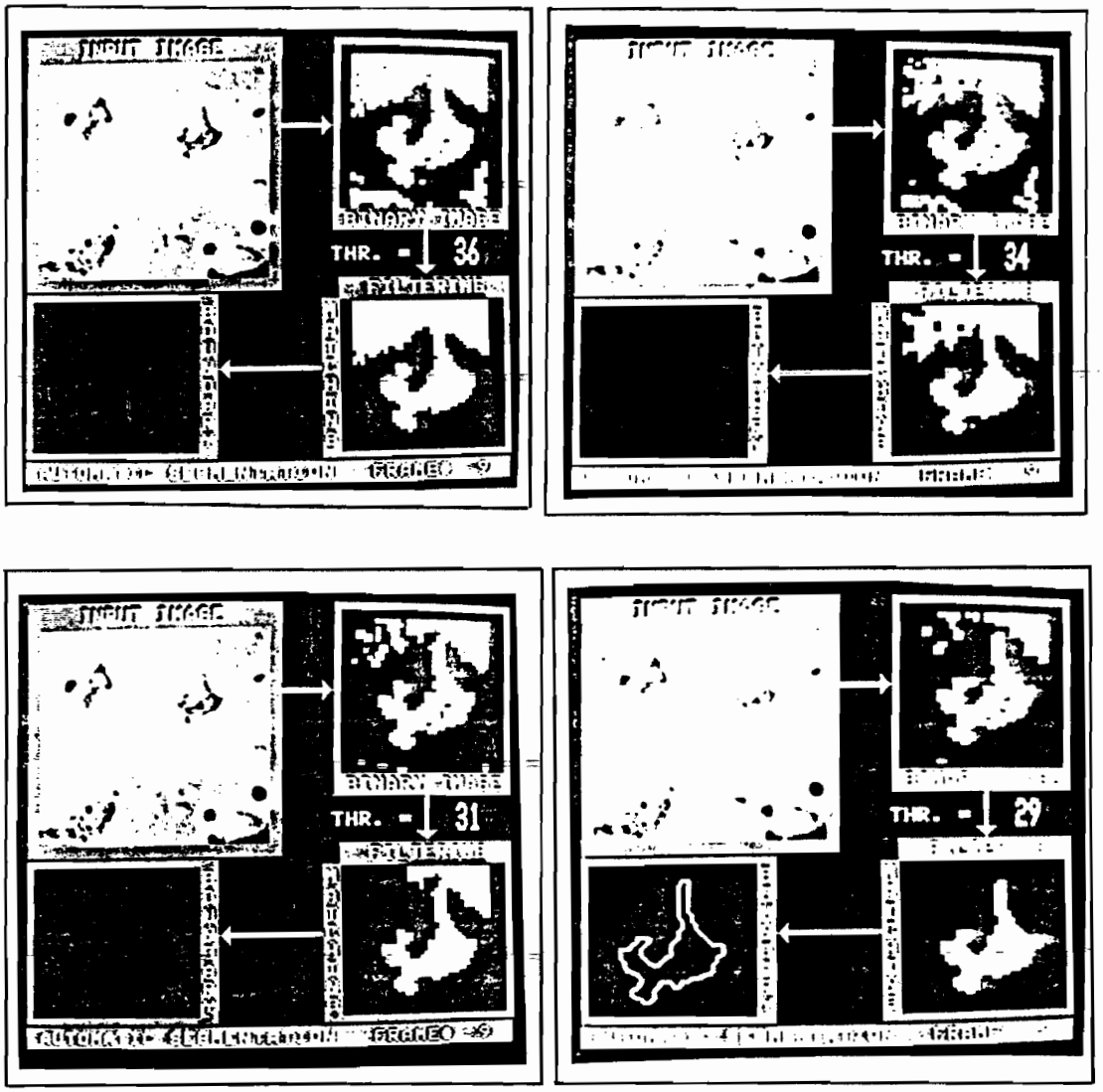
RULE(4.5):

IF A(ti)-A(ti+1) .GT. Et1 ==then==> DECREASE THRESHOLD

IF A(ti)-A(ti+1) .LT. Et2 ==then==> INCREASE THRESHOLD

IF RULE(4.5) IS ACTIVATED ==then==> ACTIVATE (A4 --> A7)

The window parameters and the threshold values may be adjusted iteratively using the above rules until a satisfactory segmentation is achieved. Figure (4.13) shows examples of the output of the different steps of the




*Figure(4.13) The output of the different steps in the automatic segmentation. Several iteration are required to obtain a satisfactory result.*

automatic segmentation. In this figure, the satisfactory segmentation of the cell is achieved after five iterations using threshold values of 36, 34, 33, 31, and 29.

## 4.3 POLYGONAL APPROXIMATION

Polygonal approximation may be categorized as a data compression method. It is a well-known technique in shape analysis for digital image processing and pattern recognition. In this technique, a curve or contour is represented by fitted straight lines. The input to such algorithms can be either the boundary points directly, or their chain code. The output is a list of vertices of the fitted lines. These techniques have the advantage of reducing the noise as well as the amount of data to be manipulated by higher level stages of the system (the number of the vertices will always be less than the boundary points). For example, polygonal approximation often retains the local peaks that are important in the shape analysis of biological objects; and they often retain the shape of the orginal object as it is percieved by the human viewer.

In some applications, the fitted lines or polygons may be used directly for shape recognition or description, while in others they are an intermediate form of data. For example, in our system, the output of the polygonal approximation represents the input for the shape decomposition. The latter is further analyzed by higher level processes of the system (incremental and global

analysis). The mathematical aspects of the polygonal approximation may be found in [Pavlidis, 77]. A review of the significant work in the literature, pertaining to the polygonal approximation, is presented in Section 2.3.4. In this section, first, a summary of the objectives and different methodologies of the polygonal approximation will be presented. Then, an algorithm for polygonal approximation, based on the technique introduced by Ramer [Ramer, 72] will be discussed.

## OBJECTIVES AND METHODOLGIES

The problem of polygonal approximation may be defined as follows: given a set of boundary points representing the shape of a planar object, the objective is to find the minimum set of vertices which pertain to the original shape. Some schemes for constructing polygonal approximations are concerned with selecting the vertices from the boundary points in order to generate the best fitted polygon. Others may allow the vertices to leave the curve itself, if they generate a better fitted polygon. In some algorithms, for example, the minimum distance (not to exceed a specific threshold) between the segment of the boundary and the fitted line is used as a criterion for selecting the best fitted approximating line [Ramer, 72]; whereas in others, the fitted line is picked so as to minimize the difference in area between the approximating line and the original curve, e.g. [McClure, 77]. The definition of the best

fitted polygon (optimum) differs according to the following criteria: minimum number of vertices, minimum perimeter, and minimum error (the difference between the polygon and the object area). Another common objective for any polygonal approximation technique is to minimize the computation time.

The different methodologies that have been used for the polygonal approximation may be classified into three basic techniques: (1) splitting, (2) merging, and (3) merging and splitting. The splitting algorithm starts by dividing the boundary points into two sections and proceeds further by dividing each section in two as long as a uniformity predicate is false. The merging algorithm proceeds in a linear scan evaluating the uniformity predicate as it goes along; when this is false, a new segment is started. In the merging and splitting, the algorithm initially divides the set of boundary points into a number of segments (n) (break points), the latter either splitted or merged.

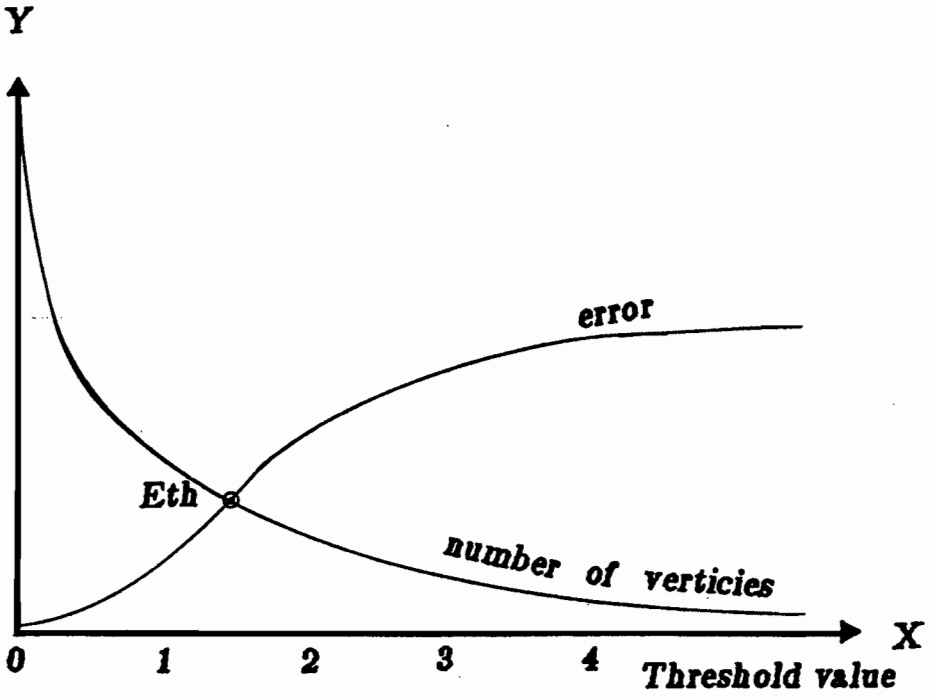
## IMPLEMENTED ALGORITHM

A splitting algorithm for polygonal approximation based on a technique presented originally by Ramer [Ramer, 72], is implemented in our current work. This technique is one of the earliest and most efficient techniques; it selects the polygon vertices from the boundary points. The resulting polygons are not necessarily optimum (minimum number of vertices), but the computational algorithm is much more

efficient and simpler than those generating optimum ones.

The procedure of the algorithm can be described through a binary tree; the root corresponds to the whole boundary. The branches of each node correspond to the subintervals. The bottom leafs are the final intervals (the resulting polygon vertices). In order to improve the result of Ramer's algorithm, a suggestion was introduced by Pavlidis [Pavlidis, 76]. In this modification, the final intervals (polygon vertices) can be used as initial break points for a merging-splitting algorithm. This may result in an optimum polygon (minimum vertices and error). On the other hand, in order to decide whether to merge or split, the computation is quite time consuming. However, a similar result may be obtained without using the merging-splitting algorithm. This can be achieved by the proper selection of the threshold value of Ramer's algorithm.

The approximation threshold value plays an essential factor in the procedure, and the result of the polygonal approximation algorithm. Based on this value, a decision may be taken whether a given interval should be divided into two segments or not. Thus, if the maximum normal distance between the approximating line and the boundary points exceeds the approximation threshold value, then the interval is divided at the boundary point which exhibits the maximum normal distance. Figure (4.14) illustrates the use of different threshold values for a given shape. In this figure, the X axis represents the threshold values, and the

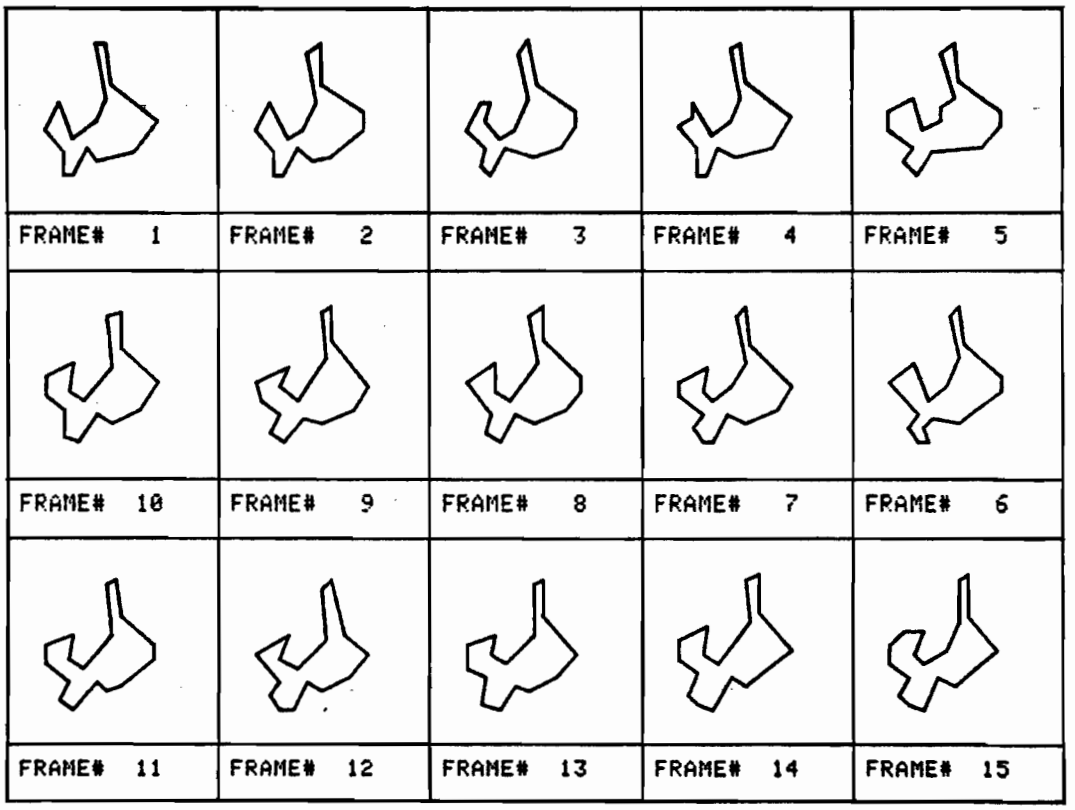



*Figure(4.14) An approximate estimation for the threshold value (Eth) necessary for polygonal approximation, which minimize both number of verticies and the error (the difference between the resulting polygon and the original shape).*

Y axis represents two parameters: the number of vertices and the error. From this figure we can see that, the lower the threshold value, the higher the number of vertices and the less the error (the difference between the polygon and original shape). This means that increasing the efficiency towards one of these desired objectives (minimum number of vertices and minimum error) comes at the cost of the other.

A basic issue is to find the approximation threshold value which minimizes these two conflicting factors simultaneously. From Figure (4.14) we can see that the intersection of the two curves representing the number of vertices and the error (point E), is a good estimation as an approximation threshold value. However, this point is specific for each individual shape. Therefore, this method is more useful in the static scene analysis or image sequence analysis of rigid moving objects, where the shape does not change and is known a priori. Conversely, in the image sequence analysis of non-rigid moving objects, the threshold value should be updated with the shape changes.

In the implemented algorithm for the present system, the threshold value is chosen based on experimental work in order to find the value that retains the cell shape (as it is perceived by a human observer) for a minimum number of polygon vertices. Thus, the value of 1.5 pixels (1% of the average cell perimeter) has been found to be the best threshold for polygonal approximation for most cell shapes. Figure (4.15) shows the polygonal approximation of the cell




*Figure(4.15) Polygonal approximation of the cell shapes in a subsequence of sequential frames. The original boundaries are shown in figure (4.12).*

shapes in a sequence of frames, shown in Figure (4.12). The polygon features to be measured at this stage of analysis are given in Table 4.1.

## 4.4 POLYGONAL DECOMPOSITION

An important objective in our research is to characterize and describe the dynamic behaviour of the different pseudopods formed during locomotion. And, to study the relationships between changes in the shape of the subparts, their movement, and the global locomotion of the cell. Therefore, the decomposition of the cell into its primitive subparts represents an essential process for understanding the structural changes of a moving cell.

Decomposition methodologies are prime examples of structural pattern recognition and shape analysis. They are based on the assumption that shape perception is a hierarchical process [Pavlidis, 68; 72]. In these techniques the original figure is expressed as the union of some of its subsets (primitive components). The shape of the latter may be simpler and, therefore, some of the less complex descriptions may be applicable. A review of the different decomposition techniques is presented in Section 2.3.5. These different techniques can be classified as follows:

(a) primary convex subsets,

(b) concave vertices,

(c) clustering,

(d) k-nearest neighbors,

structure of the cell. Thus, the cell shape has a star like structure, where the center node is the cell body, and the branches are pseudopods or subparts of the cell. Since we are interested mainly in the pseudopods and the subparts forming around the membrane, the decomposition at the convex angles seems to be the most efficient method for cell structural analysis.

The internal angles of the polygonal approximation of the cell may represented as a set of numerical values {V} such that:

$$\{V\} = \{V1, V2, \ldots, Vi, \ldots, Vnv\} \qquad (4.8)$$

where Vi is the internal angle of vertex (i), and nv the number of polygon vertices. The angle Vi may be assigned as convex angle (VX) or concave angle (VC) according to the rule:

RULE(4.6)

IF Vi .VT. 180, ==then==> Vi <--- VX,

IF Vi .LT. 180, ==then==> Vi <--- VC,

thus the set {V} may be expressed as:

$$\{V\} = \{\{ \text{CONCAVE ANGLES} \}, \{ \text{CONVEX ANGLES} \}\} \qquad (4.9)$$

$$\{V\} = \{ \{VC\}, \{VX\} \} \qquad (4.10)$$

$$\{VC\} = \{VC1, VC2, \ldots, VCnc\} \qquad (4.11)$$

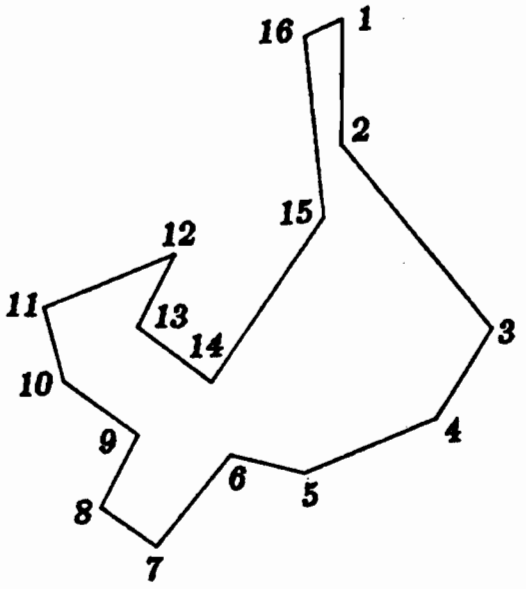
$$\{VX\} = \{VX1, VX2, \ldots, VXnx\} \qquad (4.12)$$

where nc, nx are the number of concave and convex angles respectively, and nc + nx = nv.

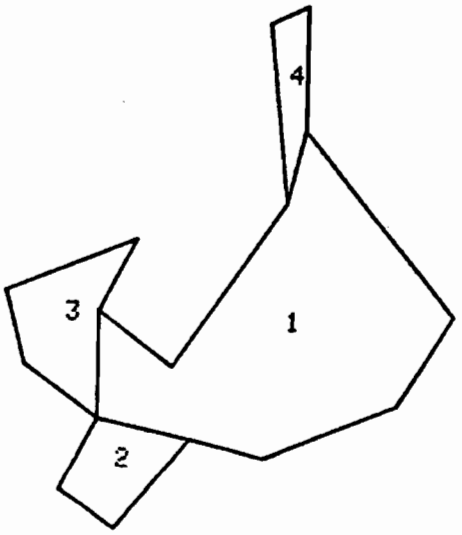
For example the angles of the cell polygon in Figure#(4.16a) are:

{V} = {V1 , V2, . . . . , V16}

= {VC1, VX1, VC2, VC3, VC4, VX2, VC5, VC6, VX3, VC7, VC8, VC9,




(a)




(b)

*Figure(4.16) Decomposition of the cell into its primitive subparts. (a) input, (b) output.*

VX4, VX5, VX6, VC10>

<VC> = <VC1, VC2, . . . , VC10>

<VX> = <VX1, VX2, . . . , VX6>

We define "Convex String" (CXS) as a sequence of convex angles (VX's) without interruption of concave angles (VC's). Each string may contain any number of convex angles (one or more). Thus, the set <VX> may be expressed as:

<VX> = <VXS1, VXS2, . . . , VXSns> (4.13)

where ns is the number of convex angles strings.
In the above example: <VX> = <VXS1, VXS2, VXS3, VXS4>.

The polygon will be decomposed if it contains at least two strings of convex angles.

RULE(4.8):

IF NS .GE. 2, ==then==> ACTIVATE DECOMPOSITION

RULE(4.9):

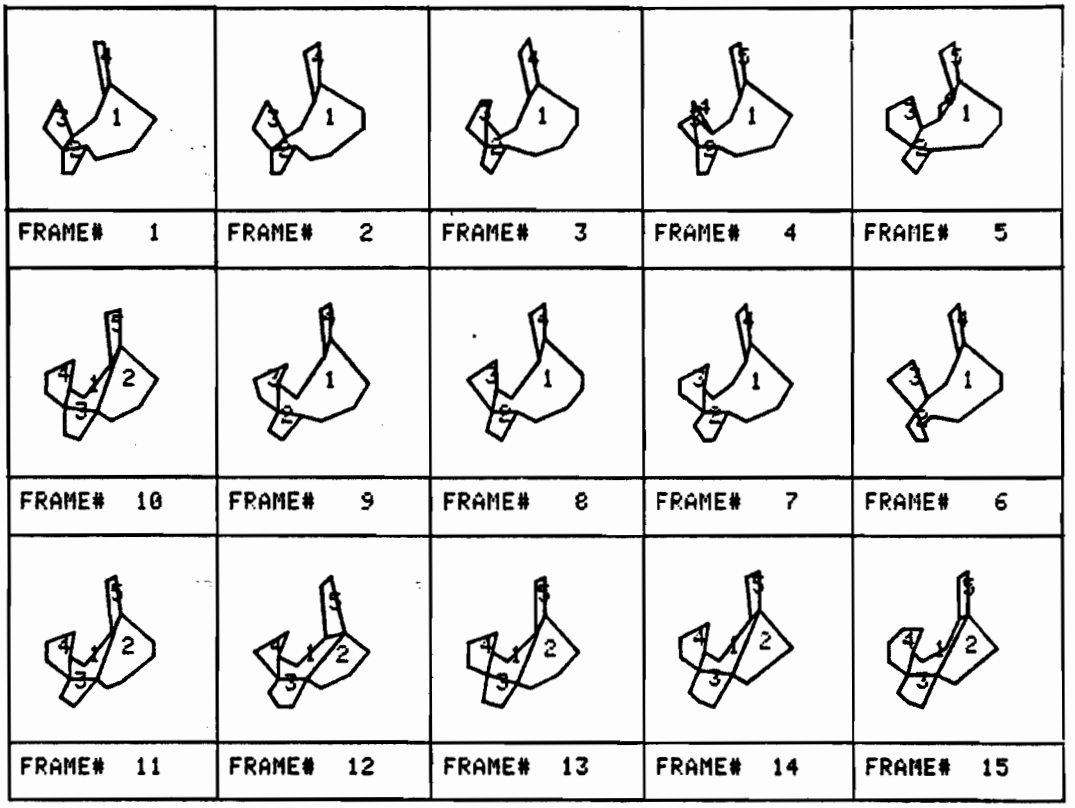
IF NS .LE. 1, ==then==> EXIT

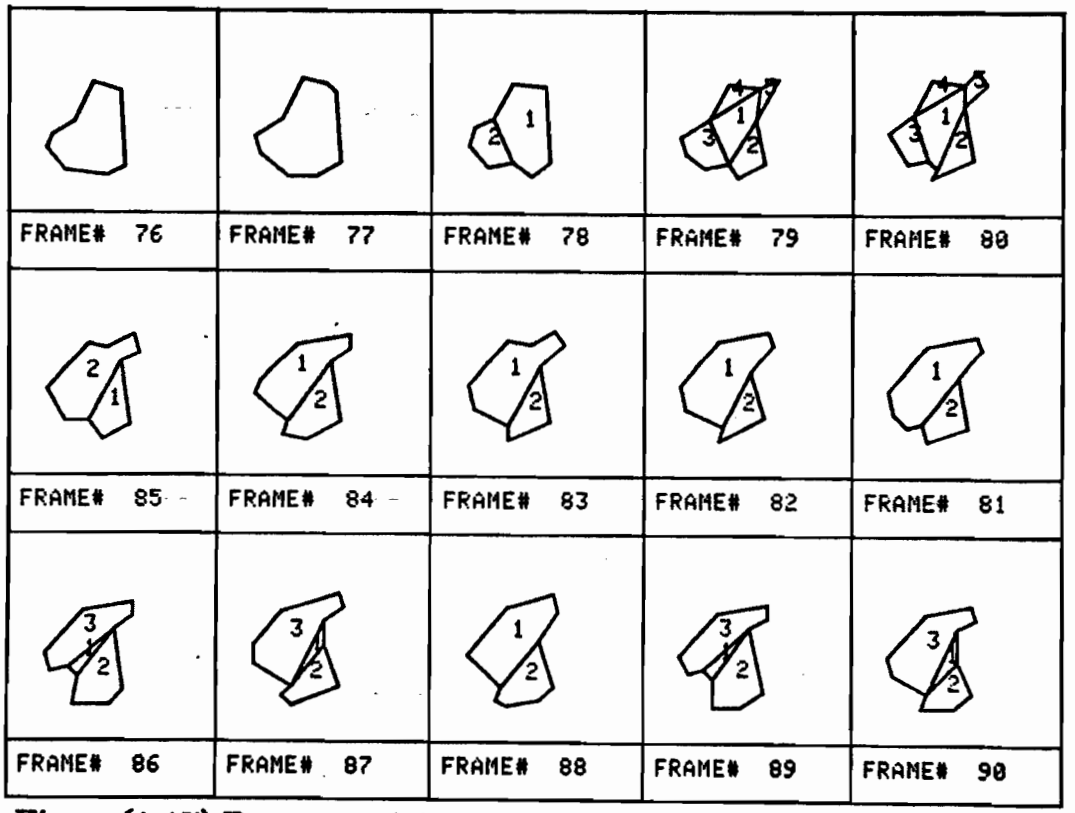
The decomposition process starts by considering the whole polygon as subpart number one. Then, it proceeds by connecting the last vertex in each convex string (VXS(i)) to the first vertex in the next one (VXS(i+1)). The line connecting these two vertices is defined as the "Base Line" (BL). The base line should satisfy the following conditions: (a) it lies entirely inside the polygon, (b) it does not intersect any of the polygon sides or another base line. If the base line satisfies these conditions, the part of the polygon bounded by it is assigned as a new subpart,

and subpart number one is ubdated by subtracting this new subpart. Figure (4.16b) shows the decomposition of the cell in Figure (4.16a), and Figure (4.17) shows the decomposition of the cells in the frames that are shown in Figures (4.12) and (4.15). In Figure (4.17), we can see that the cells in frames 76 and 77 are examples of nondecomposable polygons which may be classified as simple or very simple shapes.

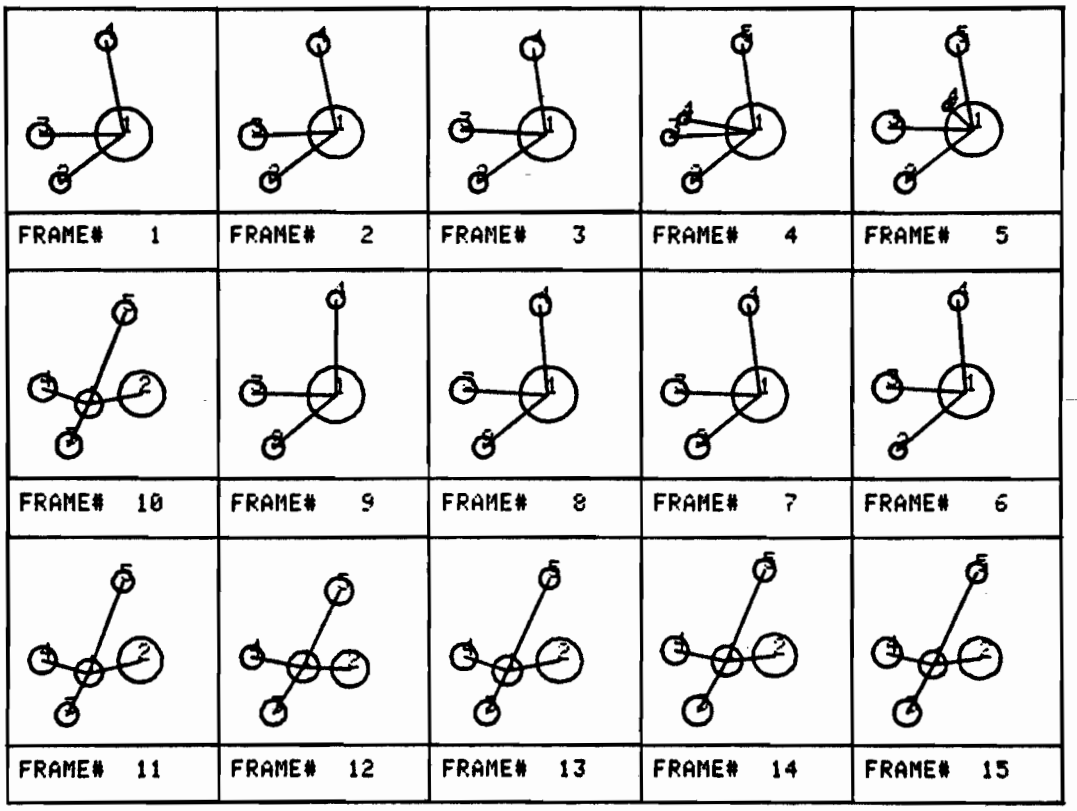
The last procedure in the polygon decomposition is to represent the cell by a labeled graph that has a star configuration. In this graph, the central node corresponds to the main body of the cell, and the different nodes could correspond to the primitive subparts of the cell (mainly pseudopods). The branches connecting the different nodes to the central one, represent the structural relationships between the cell body and the different pseudopods or subparts. Figure (4.18) illustrates the labeled graphs of the polygon decomposition shown in Figure (4.17). The area of a node in the graph is equivalent to the area of the corresponding subparts. The connecting lines between the center node and the different branches are equivalent to the distance between the corresponding subparts and the cell centroid and have the same direction (angle with the X axis).

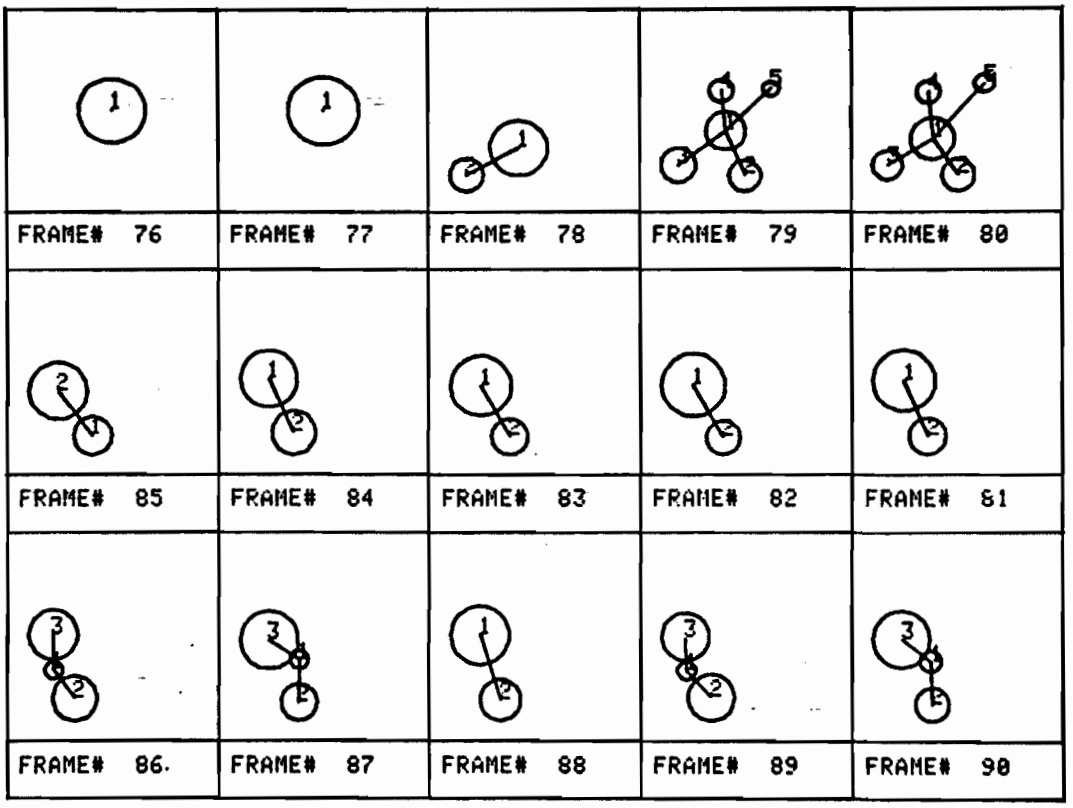
Analysis of the labeled graph will provide information about topological properties of the entire cell structure, its shape, as well as its primitive subparts. In Section 4.6 we demonstrate the analysis of the labeled graph




*Figure(4.17) Decomposition of the cell in a sequence of frames. Note that frames 76 and 77 have simple shapes which cannot be decomposed.*




*Figure(4.18) Graphs representing the geometrical structure of the cell.*

in order to generate the description of the cell in a given frame.

## 4.5 FEATURE EXTRACTION AND SELECTION

Feature extraction and selection is one of the most important and difficult steps in any scene analysis or pattern recognition system. The best definition of this problem is still as it was stated more than twenty six years ago by Selfridge "The extraction of significant features from a background of irrelevant detail" [Selfridge, 55]. This problem can be divided into two main steps: feature extraction and feature selection.

Feature extraction is the process which associates a set of primitive properties {P} with each object (O) or subobject (SO). For example, object (Oi) may be associated with a set of properties as:

$$\{P, Oi\} = \{(P1, Oi), (P2, Oi), \ldots, (Pn, Oi)\} \qquad (4.14)$$

where n is the number of elements in the set {POi}, which represents all the properties which can be measured for object (Oi).

The feature selection is the process of pruning ineffective sets of properties in order to select an effective subset. For example, the selected subset {Ps} for object (Oi) could be:

$$\{Ps, Oi\} = \{(Ps1, Oi), (Ps2, Oi), \ldots, (Psm, Oi)\} \qquad (4.15)$$

where {Ps, Oi} is subset of {P, Oi}, and m is the number of

the selected properties to be measured and used for representing or describing object (Oi).

The selected elements of the subset {Ps, Oi} may be some of the original elements of the set {P, Oi}, or a function of one or more of them. For example, the original set may include the following properties: length, width, area, and perimeter. The selected subset may include the properties: area, circularity, and elongation, where,

circularity = ( 4. $\pi$ . area ) / ( perimeter squared ),

elongation = width / length

The selected subset of properties depends on the type of object under analysis, the objectives of the analysis, and the desired level of description. For example, in geometrical shape analysis, in order to describe or classify a given polygon as: triangle, quadrangle, pentagon, hexagon, ...., only one property is required, that is the number of sides or number of vertices. Whereas, if the desired description requires further classification such as: right-triangle, isosceles-triangle, isosceles-right-triangle, equilateral-triangle, square, rectangle, rhombus, parallelogram, trapezoid, ... etc, the selected subset of properties should include the length of each side and the angle at each vertex.

The aspects of feature extraction and selection are a subject for which there is a substational literature (it is a common problem in almost every system). An early survey

of this subject for general applications may be found in [Levine, 69], and another for radiographic images in [Hall et. al, 71]. More recently, Sklansky introduced an updated summary of this subject [Sklansky, 78]. There are many other lengthy papers, and a survey of them would take us beyond the intended scope of this research. For example, in the analysis of cell images, Lee introduced a method for computing features for classification, storage, and retrieval of leukocytes [Lee, 76]. Fu and Bacus, in order to classify WBC, made a study to select a subset that contains 17 properties out of a complete set including 367 different properties (including the different Fourier descriptors).

In general, most of the previous computer vision systems have been designed to perform tasks or solve problems similar to what humans do. This means that the problem and solution have been defined by humans. Consequently, the properties which have been used by the program to solve a specific problem have been selected to be similar to those used by humans to solve the same problem. For example, the properties which have been used for blood cell classification are those which have been utilized by biologists to recognize the different types of cells.

However, in our current research, we are designing a system to quantify and describe the dynamic behaviour of a moving cell, a problem which has not been yet solved by biologists. This is because the data analysis techniques

necessary to achieve these objectives are not available to biologists. Consequently, the properties and descriptors which may be used have not been defined by them. Therefore, the design and implementation of our system needed interactive sessions with a cooperating biologist, in order to find the most useful and effective properties which may be used to accomplish the desired objectives.

The properties which are used in our system for the different stages of analysis are given in Table 4.1. They have been selected based on three factors:

(a) effectiveness,

(b) number of properties (minimum),

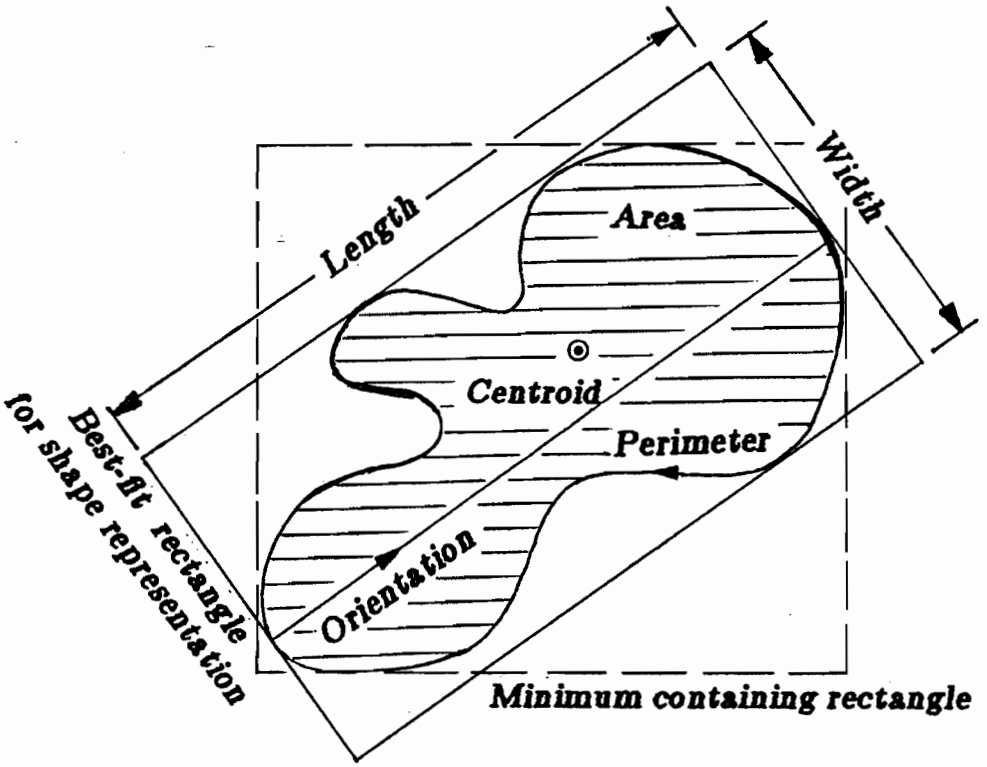
(c) speed of computation.

In the preceding sections of this chapter, we described three levels of cell abstraction: segmentation, polygonal approximation, and polygon decomposition. At each level of this abstraction, the cell is represented by a set of properties as presented in Table 4.1. Figure (4.19) illustrates the basic properties which can be measured for the cell shape. In the remainder of this section, a description and theoretical definition of each property will be given.

AREA:
=====

Is the number of non-zero pixels in the binary image of the object,

$$A = \sum_{y=MNY}^{y=MXY} \sum_{x=MNX}^{x=MXX} B(x,y) \qquad (4.16)$$




*Figure(4.19) Main features of a cell.*

$$\text{where } B(x,y) = \begin{cases} 1 & \text{object element} \\ 0 & \text{else where (background)} \end{cases}$$

PERIMETER:
==========

Is the length of the exterior outline of the object. It can be calculated from the summation of the distance between the adjacent vertices of the object polygon.

$$P = \sum_{i=1}^{m} \left[ \left( X(i+1) - X(i) \right)^2 + \left( Y(i+1) - Y(i) \right)^2 \right]^{1/2} \qquad (4.17)$$

where m is the number of vertices in the polygon. Due to the cyclic nature X(m+1) = X1, and Y(m+1) = Y1. In the case of computing the polygon from the boundary points:

$$P = NB1 + NB2 \cdot (2)^{1/2} \qquad (4.18)$$

where NB1 the number of points where X(i) = X(i+1) or Y(i) = Y(i+1), and NB2 else where, NB1+NB2 = the total number of boundary points.

MINIMUM CONTAINING RECTANGLE:
==========

Defines the size of the rectangle which encloses the entire object. The orientation of the rectangle is parallel to the X and Y axis. This rectangle can be used as a rough estimate of the size and location of the object. Its size and location are defined by the minimum and maximum location of the object in the X and Y directions (MNX, MXX, MNY, and MXY; see Figure 4.19).

CENTROID:
=========

Is the center of gravity of the object. The X and Y coordinates of the centroid CNX, CNY can be computed from the binary image as:

$$CNX = \left[ \sum_{y=YMN}^{y=YMX} \sum_{x=XMN}^{x=XMX} X\, B(x,y) \right] / AREA \qquad (4.19)$$

$$CNY = \left[ \sum_{y=YMN}^{y=YMX} \sum_{x=XMN}^{x=XMX} Y\, B(x,y) \right] / AREA \qquad (4.20)$$

AVERAGE INTENSITY:
==================

The average gray value (brightness) of the object.

$$AVI = \left[ \sum_{y=YMN}^{y=YMX} \sum_{x=XMN}^{x=XMX} f(x,y) \right] / AREA \qquad (4.21)$$

where f(x,y) is the gray-value of the pixel (X,Y) in the intensity image.

BEST FIT RECTANGLE:
===================

The rectangle whose main axes are the length and width of the object.

CIRCULARITY:
============

Is a relative measure of the ratio between the area and the square of the perimeter. Its value varies between 0 and 1, the latter value being obtained for a circle. Circularity has often been used as a measure of "smoothness", or the "complexity" of a boundary, since the

more complex the boundary, the longer the perimeter. This ratio has also been used as a measure of compactness of figures. However, Rosenfeld disputes the reliablity of this measure for digital picture analysis [Rosenfeld, 73]. In a later section of this thesis, we will demonstrate this unreliability through experimental examples. The circularity may be computed as:

$$CIRC = 4 \pi A / P^2 \qquad (4.22)$$

LENGTH:
=======

The distance between the two farthest points on the boundary.

$$LENGTH = \max \{ [ (X_i - X_j)^2 + (Y_i - Y_j)^2 ]^{1/2} \} \qquad (4.23)$$

$i = 1, 2, 3, \ldots, n-1$ and $j = i+1, i+2, \ldots, n$

n is the number of the boundary points.

WIDTH:
======

The maximum extension of the object on both sides of the length and normal to it.

ELONGATION:
===========

The complement of the ratio of the width to the length.

$$ELONGATION = 1 - ( WIDTH / LENGTH ) \qquad (4.24)$$

AVERAGE BENDING ENERGY(ABE):
============================

The rate of change of the Tangent along the boundary.

$$ABE = [ \sum_{i=1}^{i=n} ( dCi / Li )^2 ] / n \qquad (4.25)$$

where dCi is the change in the Tangent at vertex i, and Li is half of two sides at vertex i.

## REGULARITY:

Regularity is a well-known property that humans have always used to describe the shapes of different objects. However, it has not yet attracted much attention as a shape descriptor in computer vision systems. For example, a circle, square, rectangle, pentagon, and hexagon are defined as perfectly regular shapes. These are used by humans as references to approximately describe the shapes of other unfamiliar objects. A measure can be defined to determine the irregularity of an object, by comparing it to a perfectly regular shape. In our research, we found that regularity is a reliable property for global shape description and classification. The power of this property in characterizing various arbitrary shapes will be demonstrated in a later section.

The regularity of a specific shape is based on two criteria, angles and sides. Definitions and mathematical formulae for computing each are given below.

## ANGLE REGULARITY

A measurement of the sum of the differences between the angles of a given polygon and a regular one (equal angles) having the same number of sides. Thus, for a polygon with n verticies a1, a2, a3, . . . , ai, . . . , an, the sum of internal angles

(An) is:

$$An = \sum_{i=1}^{i=n} ai = (n-2) \cdot 180. \qquad (4.26)$$

In the case of a regular polygon, a1=a2=a3=...=ai=...an = ar, and ar = (n-2).180 / n. The angle regularity (AR) can be computed as:

$$AR = \left[ \sum_{i=1}^{i=n} |ai-ar| \right] / K \qquad (4.27)$$

where K is a normalization factor, which is determined in order that AR = 0 for the most regular shapes, and equal 1 for the most irregular ones. The value of K can be computed as:

$$K = \begin{cases} ar(n+2) & \text{for n even} \\ ar(n+1) & \text{for n odd} \end{cases} \qquad (4.28)$$

## SIDE REGULARITY (SR)

The same as above, except the measurement involves the sides instead of the angles. It can be computed as:

$$SR = \left[ \sum_{i=1}^{i=n} | li - L | \right] / \left[ 2L (n-2) \right] \qquad (4.29)$$

$$L = PERIMETER / n \qquad (4.30)$$

where n the number of sides, li is the length of a given side, and L is the length of the side of regular polygon that has the same perimeter and number of sides.

Readers are referred to Appendix A for a short proof of equations (4.28) and (4.29).

## 4.6 STATIC SHAPE DESCRIPTION

Shape perception is a common problem in any computer vision, scene analysis, or pattern recognition system. In this problem, we imitate a very complex process, the human perceptual process. The solution to this problem may be achieved through two stages of processing: shape analysis, and shape description. Figure (2.1) is a schematic diagram that shows the basic steps for shape analysis and description, as well as the input and output data at each step. In the preceding sections of this chapter, we described the different processes associated with the first stage of this task, that is the shape analysis. In these processes, the digitized image of the cell is transformed into two forms: a scalar vector whose elements are measurements of the main shape properties, and a graph. The properties of this graph express the shape and structural properties of the cell. In this section, we will describe the processes which are associated with the second step of shape perception, that is the shape description. The objective is to utilize data which resulted from the shape analysis in order to generate a symbolic summary describing the shape and structure of the cell in the current frame.

Past work in shape description has resulted in developing systems which work remarkably well in simulating human vision. For example, character recognition, waveforms, chromosomes, fingerprints, cells, and machine parts, among others. This work has not resulted in a sound theory of shape description [Hollerbach, 75; Shapiro, 80]. A review of the significant work which has been done in shape analysis and description is presented in Section 2.3.

## 4.6.1 Symbolic Description

Symbolic description is the most natural and powerful method for the human to express himself and his perception of the world. In computer vision, where steps are taken to model human perception, a symbolic description is an important methoh for representing the information in a more natural and informative form. For example, in the automatic processing of microscopic images, it is interesting to describe the output of the analysis in a symbolic terminology which is meaningful to biologists. Bartlett describes this in "Remembering" as:

"Words can indicate the qualitative and relational features of a situation in their general aspect just as directly as, and perhaps even more satisfactory than, they can describe its particular individuality. This is, in fact, what gives to language its intimate relation to thought processes." [Bartlett, 1976].

Symbolic descriptions to represent the data in computer vision was suggested by Minsky [Minsky, 74]. In his Framework for Representing Knowledge, he pointed out the

lack of usefulness of continuous-range numerical data. In using a measurement, what is ultimately used, very often, is not the exact value, but some qualitative judgement based on this value. Also, he showed the power of symbolic description for vision over all the other methods in the following statement:

> "This essay contains quite a few different arguments against quantitative models. Perhaps I should explain the general principle upon which they are based, since I see that separately they are not very compelling. Thesis: the output of a quantitative mechanism, be it numerical, statistical, analogue, or physical (non-symbolic), is too structureless and uninformative to permit further analysis. Number-like magnitudes can form the basis of decision for immediate action, for muscular superposition, for filtering and summing of stimulus features, and so forth. But each is a "dead end" so far as further understanding and planning is concerned, for each is an evaluation -- and not a summary. A Number cannot reflect the consideration that formed it. Thus, although quantitative results are useful for immediate purposes, they impose a large cost on further and deeper development." [Minsky, 74].

One of the few applications which used symbolic description in a manner close to our approach is by [Hollerback, 75]. In his approach to shape description, he generated symbolic descriptions of the different parts of a vase such as body, neck, lip, foot, and handles. He used these descriptions to categorize different vases.

In order for the human to choose the proper symbolic descriptor for a specific property of an object, he compares the perceived data to his a priori knowledge about the same class of object or similar ones. For example, to describe a person as "SHORT or TALL", the human compares the length of the person to the length of an average adult. This process

involves human production rules such as:

"IF PERSON IS TALLER THAN THE AVERAGE, THEN HE IS TALL", or

"IF PERSON IS SHORTER THAN THE AVERAGE, THEN HE IS SHORT".

The above represents an example of a human production rule in its simplest form. The basic elements of data which construct this rule are: (a) data extracted from the perceived scene (the person to be described in the above example), (b) a priori knowledge to be used as reference in order to describe the perceived data (the length of the average person), (c) a comparative operator to be used in the matching or comparison procedure (taller than, shorter than), (d) an adjective descriptor (tall, short). It is clear that (a), (b), and (c) are the basic elements of the left hand side of the rule (conditions), whereas (d) represents the right hand side (action).

This philosophy of human production rules is the foundation of the structure of our representational rules for assigning symbolic descriptors to the different properties of the moving object under analysis. Thus, the Symbolic Descriptor Representational Rules (SDRR) are a group of condition --> action production rules; their main function is to choose, among a set of symbolic descriptors, the appropriate one, for a given numerical value which has been extracted from the sequence of images.

In the above example, if a more precise description is required, a human may expand his description to five classes to include "VERY SHORT, SHORT, AVERAGE, TALL, VERY TALL". In this case, the property of an object is described in multiple levels of description. Obviously the process here will be more difficult and ambiguous. Thus, in describing the length of a person, one may say:

IF THE PERSON IS SHORTER THAN 4.5 FEET,
THEN HE IS VERY SHORT

IF THE PERSON IS TALLER THAN 4.5 FEET,
AND SHORTER THAN 5 FEET,
THEN HE IS SHORT

IF THE PERSON IS TALLER THAN 5 FEET,
AND SHORTER THAN 5.5 FEET,
THEN HE IS AVERAGE

IF THE PERSON IS TALLER THAN 5.5 FEET,
AND SHORTER THAN 6 FEET,
THEN HE IS TALL

IF THE PERSON IS TALLER THAN 6 FEET,
THEN HE IS VERY TALL"

In the above description, approximate values (4.5, 5, 5.5, and 6) have been used as classification thresholds to classify and describe a person as a member of one of five classes "VERY SHORT, SHORT, AVERAGE, TALL, VERY TALL". However, these classification thresholds will be different

amongst the Watusi and Pygmy tribes of Africa. This discussion indicates two basic subjects for analysis: the number of classes, and the classification thresholds. These aspects will be discussed in the following sections.

## 4.6.2 Number Of Classes (Levels) Of A Symbolic Descriptor:

The number of classes of a symbolic descriptor is controlled by the accepted level of approximation which is related to the desirable description. Specifically, the greater the number of classes (levels), the more precise the description. On the other hand, the fewer classes that are defined, the simpler is the programming, the fewer required rules, the fewer symbolic descriptors to be defined, the smaller the processing time, and the more data compression is achieved in general. These factors force the designer to study the optimum number of classes that combines the best of the above conflicting factors.

The simplest method for dividing a set of data into classes is to classify them into two groups similar to transforming a gray-level image into a binary one (black and white). Thus, using a single threshold, each numerical value can be assigned to one of two subdescriptors such as "SHORT, LONG", "FAST, SLOW", ....etc.

In this respect it is generally accepted that human ability to classify a set of data into classes is limited in number. Therefore, we mantain the consistency of the number

of subclasses to five levels in the following order: VERY LOW, LOW, MEDIUM, HIGH, VERY HIGH. Thus, in our present system, all the measured numerical values of the different properties are described symbolically in terms of five levels of description. However, in some special cases, the nature of the property to be described provokes other numbers of levels. For example, in describing the direction of the motion of a moving object (symbolically instead of angles) eight levels are defined (EAST, EAST-NORTH, NORTH, WEST-NORTH, WEST, WEST-SOUTH, SOUTH, EAST-SOUTH). Table 3.2 gives examples of the subdescriptors of the different levels of some of the main properties.

### 4.6.3 Classification Thresholds

The second problem involved in describing a set of numerical values symbolically is how to define the thresholds that can be used to divide this set of data into a given number of classes. This problem seems easier than it is. The solution involves clustering and grouping theories, human psychology, and the problem domain knowledge representation. This problem was the subject of a thorough study by Denofsky at MIT [Denofsky, 76]. The title of his study "HOW NEAR IS NEAR?" is, in fact, a good definition of the problem. Although, he did not develop a mathematical theory for determining the classification thresholds, neither does he specify in detail, how to make the appropriate choice for these parameters in a specific

situation. However, he showed through many examples that commonsense choices lead to good thresholds.

In the example demonstrated in the previous sections, thresholds of 4.5, 5, 5.5, and 6 have been used in order to classify and describe a person as a member of one of five classes "VERY SHORT, SHORT, AVERAGE, TALL, VERY TALL". Also, it was mentioned that these thresholds will be different depending on the class of people. Another example from the motion description, which possesses the same idea is to describe a displacement distance as "VERY SHORT, SHORT, AVERAGE, LONG, VERY LONG". The thresholds here will depend mainly on the type of moving object. For example, twenty microns is a very long distance for a moving blood cell, whereas a thousand miles is a very short distance for the Space Shuttle Columbia.

In order to design a general knowledge representation strategy, which may be used for different classes of objects and motion, a normalization technique of the dynamic data is employed. Hence, a general rule may be used for computing the symbolic descriptors for the different properties. A description of such normalization techniques is given below.

## 4.6.4 Normalization

For any set of numerical data

$$\langle V \rangle = \langle V1, V2, \ldots, Vn \rangle \qquad (4.32)$$

(the data can be the measurements of any of the object or

motion properties), the different elements may have values between (Vmin) and (Vmax), where Vmin and Vmax are the minimum and maximum values of (V1, V2, . . . , Vn), respectively. The different elements of this set may be normalized to range between 0 and 1 as follows:

$$vi = ( Vi - Vmin ) / ( Vmax - Vmin ) \qquad (4.33)$$

where vi is the normalized value of Vi.

Two types of normalization are defined: local normalization (scene-dependent) and global normalization (scene-independent). In the local normalization, the parameters Vmin and Vmax are the minimum and maximum values of the dynamic data measured from the scenes under analysis. In global normalization, these parameters are defined by the constraint knowledge. They are the typical values as retrieved from the general knowledge pertaining to the class of objects under consideration. For example, in the study of the shape changes of a moving cell, in order to describe the change in the size (area) of the neutrophil cell: (i) local: Amin and Amax are the minimum and maximum area of the cell, measured from the sequence under analysis, (ii) global: Amin and Amax are the minimum and maximum area of any neutrophil cell under any conditions. Obviously, in the first case the parameters are dynamic data which may change for the different analyses (STM data), whereas in the second case, they pertain to constraint knowledge related to neutrophil cell (LTM data).

The second stage of the thresholding problem is to find the thresholds which divide the range of the normalized data (0-1) into classes representing the different levels of description. Let us consider the range to be divided into (n) classes, where E1,E2,...,E(n-1) are the threshold values which define the boundaries of each level. Then each normalized value is assigned to the corresponding symbolic qualifier using the following rule:

RULE(4.9) : IF V(Pk) .GE. Ei, AND .LT. Ej,

==then==> Q(Pk) <--- Dl

where V(Pk) is the normalized value of the property Pk, Dl is the corresponding symbolic qualifier (descriptor), Ei,Ej are classification threshold values, which define the boundary of the descriptor Dl, and Q(Pk) is the symbolic qualifier of the property Pk.

Example:

RULE(4.10a):

IF V(area) .LT. E1, =then=> Q(area) <--- V.SMALL

IF V(area) .GE. E1, AND .LT. E2, =then=> Q(area) <--- SMALL

IF V(area) .GE. E2, AND .LT. E3, =then=> Q(area) <--- MEDIUM

IF V(area) .GE. E3, AND .LT. E4, =then=> Q(area) <--- LARGE

IF V(area) .GE. E4, =then=> Q(area) <--- V.LARGE

Estimating the threshold values E1,E2,...,E(n-1) is an interesting subject for study, however, it has not yet attracted much attention. It is known that people seem to respond according to a power law for many tasks in their lives. Based on this fact, Denofsky proposed a thresholding

method based on an exponential scale, in which each threshold is half the previous, and is the geometric mean (GM) of the thresholds on each side of it [Denofsky, ??]. Through copious examples drawn from everyday life, Denofsky claims that this method gives reasonable results, within (+/-)25% of where humans would place the threshold.

In the examples given by Denofsky, he divided the range of data into a number of classes, that are expanded in one direction from a fixed point. Thus, the descriptors of these examples describe the data of a specific property only from one side. For example, how near, far, short, long, small, and large. A more informative description can be generated if we divide the range of data into classes that are expanded in two directions (left and right) from a middle fixed point, where the classification is symmetrical with respect to this point. The classes of the LHS and RHS are described by very low, low, almost low, and almost high, high, very high respectively, and the middle class describes the average portion of the data. This classification method is more useful, especially for data which change between negative and positive values. For example, in the description of the acceleration of a moving object, the LHS and RHS classes would describe negative and positive acceleration respectively. The middle class would describe data which indicates an almost constant velocity. Another example relates to the rotation of an object in clockwise and anticlockwise directions. Such an approach is described

below.

Let us consider the classification of the range 0-1 into (n) classes, where n is an odd number greater than one (3,5,....). The middle class number [(n+1)/2] represents the average level class; on each side of this class, there are [(n-1)/2] classes higher and lower than the average. If we assume that the classification is symetrical with respect to the middle class then:

$$E1 = 1-E(n-1),$$
$$E2 = 1-E(n-2),$$
$$\ldots\ldots\ldots\ldots,$$
$$E((n-1)/2) = E((n+3)/2). \qquad (4.34)$$

We divide the range into (m) equal units. The width of each is (T), T = 1/m. This represents the width of the smallest class (the highest and lowest class). Using the power law, each class is half the previous one (for classes above the middle), and is twice the previous class (for the classes below the middle). Thus, if Wi is the width of class (i) then:

$$W1 = Wn = T,$$
$$W2 = W(n-1) = 2T,$$
$$\ldots\ldots\ldots\ldots\ldots,$$
$$Wi = 2^{i-1} T, \qquad (4.35)$$

and $W((n+1)/2) = 2W((n-1)/2) = 2W((n+3)/2)$,

where W(n+1)/2 is the width of the middle class. This width can be determined in terms of the unit T as follows:

$$W(n+1)/2 = 2^{(n-1)/2} \cdot T \qquad (4.36)$$

For example, if n=5, then the width of the middle class is 4T. The total number of units m can be determined as:

$$m = \sum_{i=1}^{i=n} Wi \qquad (4.37)$$

$$= W1 + W2 + \ldots + Wn$$

$$= T + 2T + 4T + \ldots + 4T + 2T + T$$

$$= W(n+1)/2 + 2\,[W(n+1)/2 - 1]$$

$$= 3\,[W(n+1)/2] - 2 \qquad (4.38)$$

Therefore,

$$m = 3 \cdot 2^{(n-1)/2} - 2 \qquad (4.39)$$

and

$$T = 1 / m. \qquad (4.40)$$

Thus the width of the different classes can be computed, (W1=T, W2=2T, W3=4T, ...., Wn=T). The different threshold values may be estimated as:

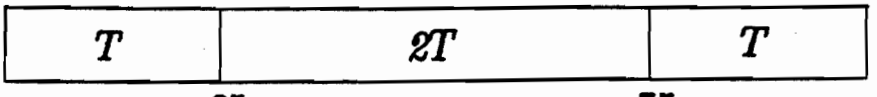
$$E1 = W1 / m$$

$$E2 = (W1 + W2) / m$$

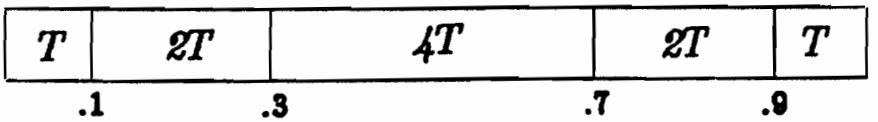
$$Ei = \left[\sum_{j=1}^{j=i} Wj\right] / m \qquad (4.41)$$

Figure(4.20) shows the classification thresholds for different numbers of classes (n=3,5,7,9). In the described system, we divided the normalized range of data into five classes, using threshold values of .1, .3 , .7 and .9. For example, the qualification of the cell area given in the above example may be computed as:

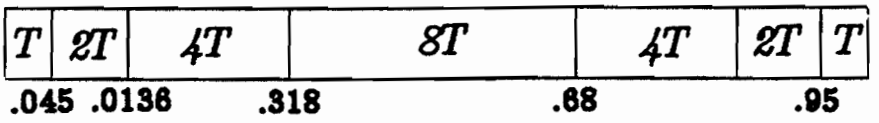
RULE(4.10b):



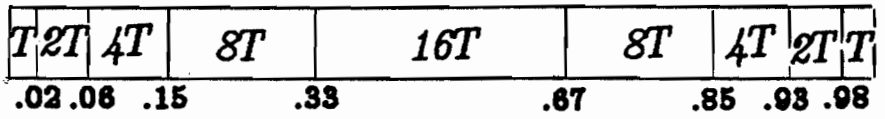

$n = 3, m = 4$ $T = 1/4 = .25$




$n = 5, m = 10$ $T = 1/10 = .1$




$n = 7, m = 22$ $T = 1/22 = .045$




$n = 9, m = 46$ $T = 1/46 = .02$

*n is the number of classes,*
*m is the number of units,*
*and T is the unit width*

$m = 3 \cdot 2^{(n-1)/2} - 2$
$T = 1/m$

*Figure(4.20) Classification a range of normalized data (0-1) into different numbers of classes (3,5,7,9).*

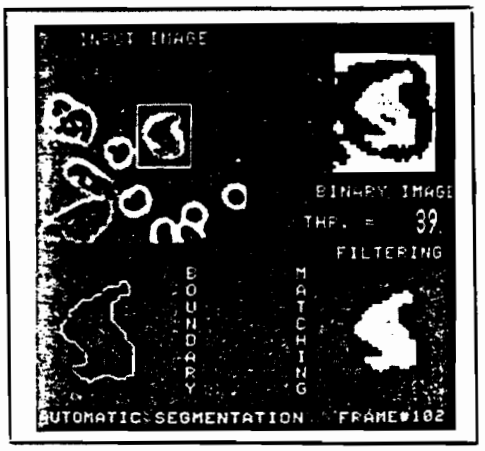
```
IF  V(area)                    .LT. .1, =then=> Q(area) <--- V. SMALL
IF  V(area) .GE. .1, AND .LT. .3, =then=> Q(area) <--- SMALL
IF  V(area) .GE. .3, AND .LT. .7, =then=> Q(area) <--- MEDIUM
IF  V(area) .GE. .7, AND .LT. .9, =then=> Q(area) <--- LARGE
IF  V(area) .GE. .9,              =then=> Q(area) <--- V. LARGE.
```

When selecting the symbolic qualifiers (SMALL, LARGE,....), one should consider the following factors: (a) pertinence to the property to be described. For example, for the size properties we may use SMALL, LARGE,..., and for distance properties we may use SHORT, LONG,..., (b) describe the numerical values to the best approximation. For example, VERY SMALL, SMALL, MEDIUM, LARGE, and VERY LARGE, (c) meaningful to the user. For example, in the description of the cell images, the symbolic qualifiers should be drawn from terminology which are used by biologists.

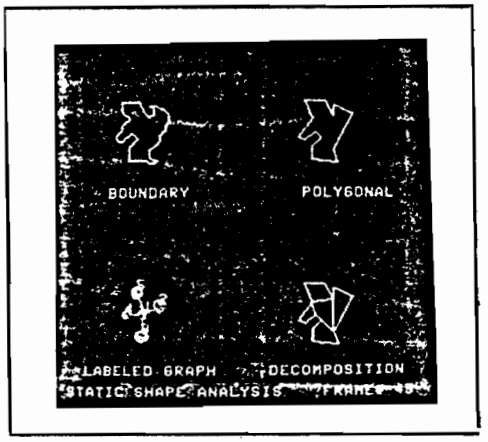
The previous sections define how to generate the symbolic qualifiers which describe the different properties of the object in a specific frame. A final step in the static description is to integrate the symbolic qualifiers into a coherent summary describing the object under consideration. This summary should be informative, understandable, and grammatically correct. Description (4.1) gives an example of the description generated by the system.

## 4.7 SUMMARY

Three stages of analysis are defined to achieve the objectives of understanding and describing the dynamic behaviour of a moving cell. These are: (1) static scene analysis, (2) incremental change detection, and (3) global analysis and description. In this chapter, we have discussed the processes associated with the first stage, the static scene analysis. The objective is to generate a summary description of the cell location, shape, and structure in the current frame. This objective is accomplished in three levels of processing. Low level processes which are responsible for locating and segmenting the cell under analysis; the output of this level is shown in figure(4.21a). The intermediate level is concerned with the shape and structure analysis of the cell. In this analysis, first, the polygonal approximation of the cell is computed in order to reduce the noise around the boundary points, as well as the amount of data to be manipulated by the higher levels. Then, in order to study the cell structural changes, the cell polygon is decomposed into its primitives. The latter represent the different subparts of the cell. Finally, the different subparts of the cell are given by a labeled graph. The properties of this graph represent the geometrical structure of the cell. Figure (4.21b) shows the output of these three steps of analysis. The function of the high level processing is to integrate the output of the analysis processes into a




(a)




(b)

*Figure(4.21) The output of the different steps of the static scene analysis.*

*(a) The segmentation process.*
*(b) Shape and structure analysis.*

coherent description such as the one given in Decription (4.1). This description is presented in a symbolic terminology that is meaningful to the biologist.

## TABLE(4.1) DIFFERENT PROPERTIES FOR CELL REPRESENTATION

| SEGMENTATION | POLYGONAL | DECOMPOSITION |
|---|---|---|
| AREA | AREA | NUMBER OF SUBPOLYGONS |
| PERIMETER | PERIMETER | CONNECTIVE LINE |
| LENGTH | ANGLE REGULARITY | BASE LINE |
| WIDTH | SIDE REGULARITY | SUBOBJECT AREA |
| ELONGATION | NO. OF VERTICIES | RELATIVE AREA |
| CIRCULARITY | LENGTH OF POLYGON SIDES | SUBOBJECT PERIMETER |
| AVERAGE BENDING ENERGY | ANGLE BETWEEN SIDES | SUBOBJECT CENTROID |
| INTENSITY (COLOR) | NO. OF CONCAVE ANGLES | ORIENTATION |
| BOUNDARY | POLYGON CENTROID | CENTROID |
| CENTROID (X,Y) | VERTICIES COORDINATES | |
| ORIENTATION | | |
| CONTAINING RECTANGLE | | |
| FITTED RECTANGLE | | |

## DESCRIPTION (4.1)

# STATIC SCENE ANALYSIS

## DESCRIPTION OF THE CELL IN FRAME : 9

The cell has a COMPLEX shape, which is JAGGY and SLIGHTLY ELONGATED. It is oriented towards the NORTH. The cell has a MEDIUM size and VERY LONG perimeter, with an average DARK gray level.

## DETAILS:

The cell can be decomposed into the following simple (convex) blobs:

The FIRST (The main body of the cell) has a size of THREE-QUARTERS of the total size of the cell.

The SECOND is adjacent to the main body with a SHORT baseline; its size is approximatly ONE-TENTH of the cell. Its centroid is QUITE NEAR TO the cell's centroid in the WEST-SOUTH direction . The length of its perimeter is ONE-FIFTH of the total cell perimeter.

The THIRD is adjacent to the main body with a SHORT baseline; its size is approximatly ONE-FIFTH of the cell. Its centroid is QUITE NEAR TO the cell's centroid in the WEST direction. The length of its perimeter is THREE-TENTHS of the total cell perimeter.

The FOURTH is adjacent to the main body with a SHORT baseline; its size is approximatly ONE-TENTH of the cell. Its centroid is QUITE NEAR TO the cell's centroid in the NORTH direction . The length of its perimeter is A QUARTER of the total cell perimeter.

# CHAPTER 5

# INCREMENTAL CHANGE DETECTION

## 5.1 INTRODUCTION

Incremental change detection or inter-frame comparison is an established methodology in image sequence analysis. The different techniques and the previous work based on this methodology are reviewed in Section 2.2.2.2. In the preceding chapter we described algorithms for static scene analysis, in which the input is a single frame and the output is a description (numeric and symbolic) of the cell under analysis, as well as its different subparts and their structural relationships. The incremental change detection between two sequential frames, their qualifications, and descriptions will comprise the material of this chapter.

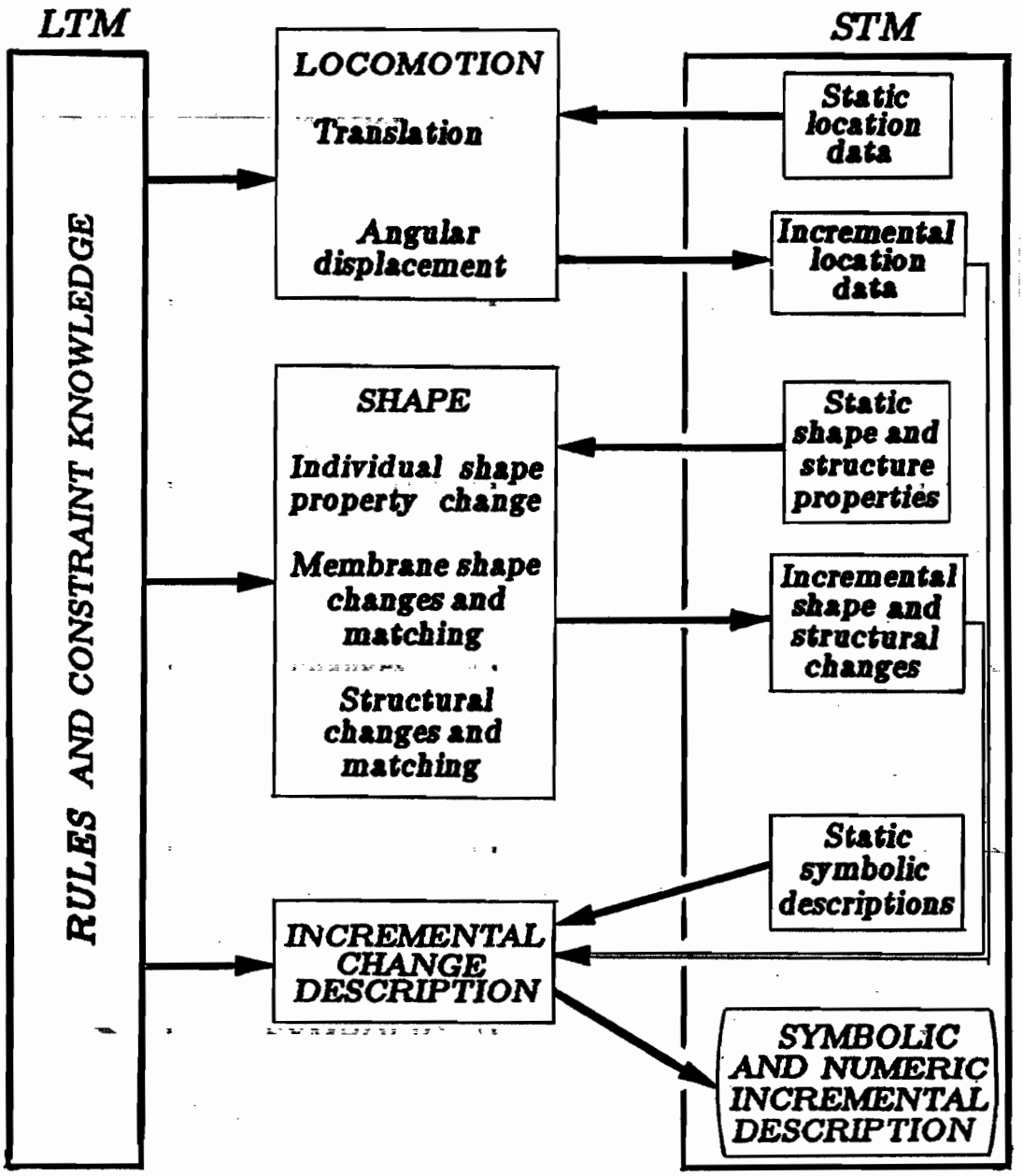
The problem of incremental change detection can be defined simply as: detect the differences between two pictures of the same scene taken over a period of time or from two different perspectives. The task of the processes solving this problem may be as simple as the detection of the object(s) which appear in only one of the two images [Ulstad, 75]. Or it may be as difficult as assigning labels to the corresponding objects and parts of objects in both

images, and then estimating the change in the location and shape for each object [Youssef and Levine, 80].

Given the location and geometric features of a specific object in two different frames (the results of the static scene analysis), the main objective of this stage of analysis is to detect and describe the changes in the location, shape, and structure of the cell and its subparts between the two frames (see Figure 1.5b). Figure (5.1) illustrates the different processes and data structure of the incremental change detection, and Description (5.1) gives a sample of the description generated by the system. The processes which generated this description will be described in the remainder of this chapter. The discussion is given in three main Sections 5.2, 5.3, and 5.4. They describe the changes in the location, shape, and structure respectively. In each of these sections, the different aspects associated with the incremental change detection, qualification, and description is discussed. Finally, Section 5.5 gives some concluding remarks.

## 5.2 LOCATION CHANGE DETECTION

Detection of the change in the location of a moving object from a sequence of images is a basic and common technique for tracking. The objective is to produce the path of the moving object, so that the motion pattern of the object may be understood. An example is the tracking system developed by Levine and Youssef. They quantified the path




*Figure(5.1) Processes and data structure of the incremental changes in location, shape, and structure of a non-rigid moving object.*

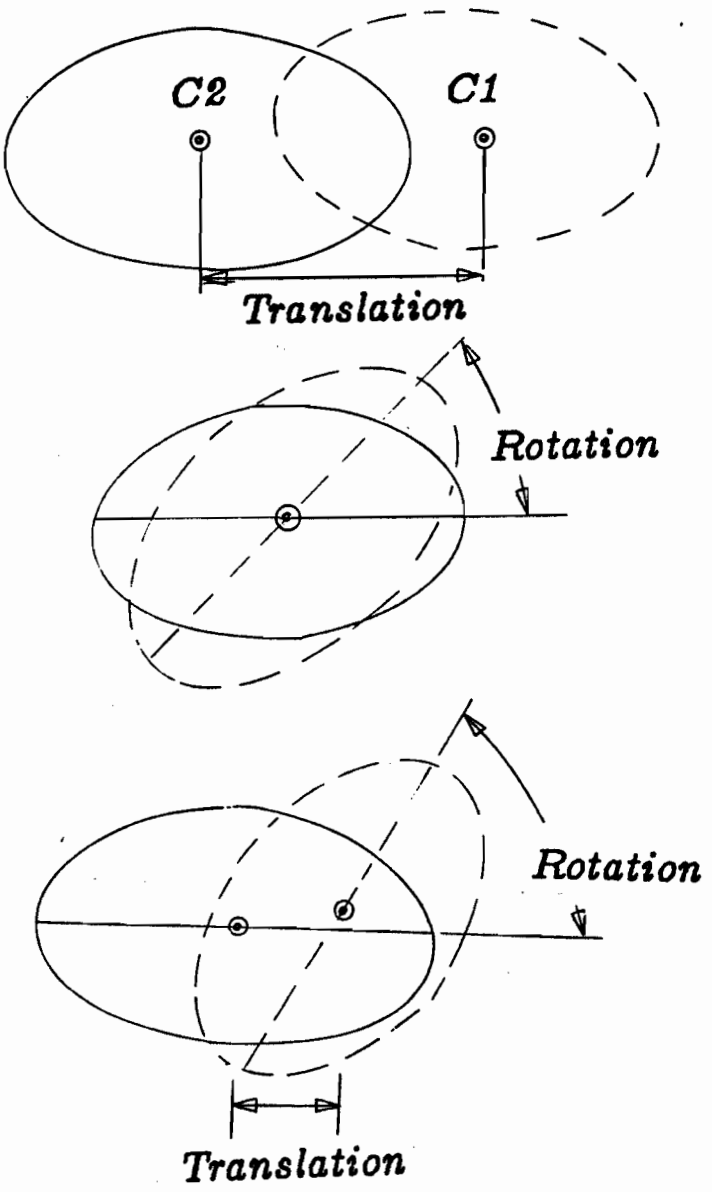
data of a group of live blood cells, thereby computing the steady-state probabilities that the cells will ultimatly move in a specific direction [Levine and Youssef, 78; Levine et al., 81].

The change of the location of an object is a function of the morphology of its elements. This process may result in the translation and/or rotation of the object. The two types of motion are illustrated in Figure (5.2). Any moving object can be classified into one of two classes:

(a) Rigid Object: An object which retains constant size and shape in three-dimensions, such as for example, a moving vehicle.

(b) Non-Rigid Object: An object which changes its size , shape, and/or structure with time, such as for example, a blood cell.

In the motion of a rigid object, the geometric relationships of its elements are constant, whereas the non-rigid object has two types of motion which occur simultaneously. One is the relative motion of the different components, and the other, the global motion of the object. Therefore, the term "translation" may be used to identify the change in the location of a rigid object, whereas the term "locomotion" is used to describe the motion of a non-rigid object. Therefore, we refer to blood cell movement as locomotion.




*Figure(5.2) Different types of motion.*

## 5.2.1 Computation Of The Change In Location

In the case of a rigid object, the change in the location may be computed as follows:

(i) Displacement in the X direction:

$$dX = XC(i) - XC(i+1) \quad (5.1)$$

(ii) Displacement in the Y direction:

$$dY = YC(i) - YC(i+1) \quad (5.2)$$

Therefore, the total incremental displacement is given by:

(iii) Translation:

$$TR = ( dX^2 + dY^2 )^{1/2} \quad (5.3)$$

Similarly, the direction of motion can be computed as:

(iv) Direction:

$$DR = Tan^{-1} ( dY / dX ) \quad (5.4)$$

where XC(i),YC(i) and XC(i),YC(i+1) are the X and Y coordinates of the cell in frames (i) and (i+1) respectively.

ROTATION

The rotation of an object can be defined by its angular displacement. This can be computed as the change in the orientation (angle between the major axis of the object and the X axis). Thus:

Rotation:

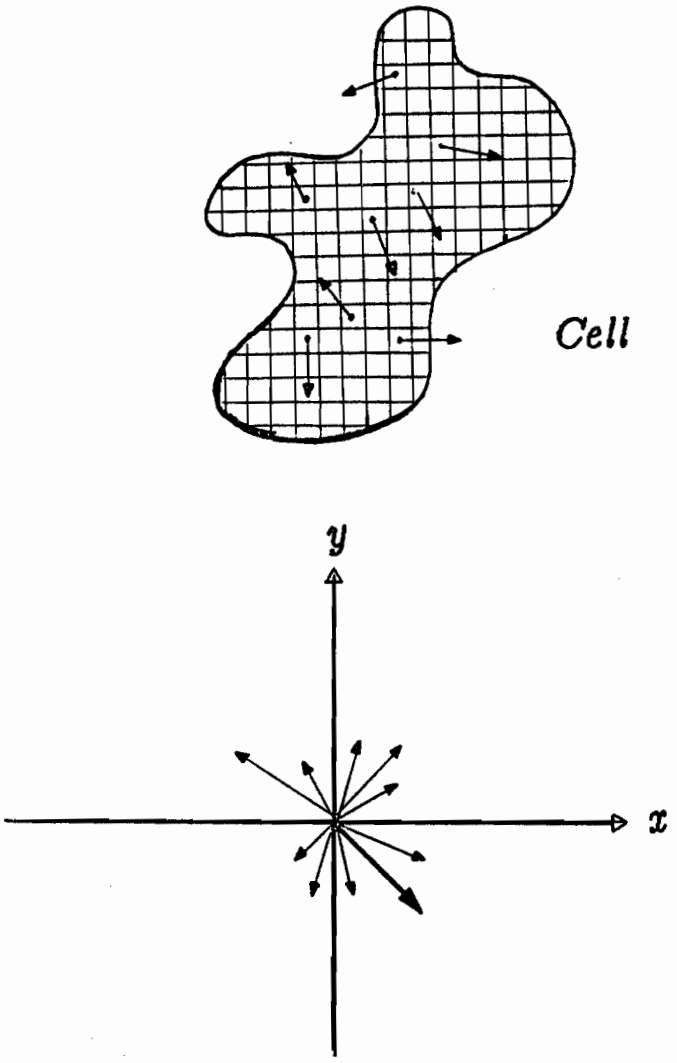
$$RO = OR(i) - OR(i+1) \quad (5.5)$$

where OR(i) and OR(i+1) are the cell orientation in frames

(i) and (i+1) respectively (see Figure 5.2).

Computation of the change in location of a non-rigid object is problematical. This is because the relative motion of the different elements of the object may change the centroid coordinates, even when the entire object is in a stationary position, and vice versa. In order to compute its change in location, we may conceptually consider each of its elements as a rigid moving object. Then, the change in location of each element (k) may be computed individually and represented as a vector v(k). The resultant of all the vectors can be estimated and represented by the vector (V). The latter yields the change in location of the entire object. Figure (5.3) illustrates this method.

The above method for estimating the change in the location of non-rigid objects is theoretically possible if the different elements can be distinguished in both locations (the two sequential frames). In most cases, however, it is impractical, since we cannot distinguish these different elements. An example is the moving cell under study. However, using a similar technique, we can detect the change in location by approximation. This is accomplished by estimating the change in the location of certain critical points computed from a knowledge of the boundary of the object. Examples are points of maximum curvature, points of the maximum extension of the object in the plane, and the centroid. Since these points are shape dependent, the approximation here depends on which is




*Figure(5.8) Computing the displacement of a non-rigid moving object by considering each element in the object as an independent rigid moving object.*

faster, the change in shape or location. However, the main assumption in image sequence analysis is the smooth change between sequential frames. Therefore, these critical points could be considered, to a large extent, shape independent between two sequential frames.

The previous analysis of the change in the location of non-rigid moving objects is necessary if a detailed and exact description of the instantaneous changes is the main objective. However, if incremental change detection is an intermediate step in the global analysis of the dynamic motion, we do not really need to go through this detailed computation. For example, in the analysis of the global locomotion of a moving cell, a change is only encountered if the cell exhibits displacement greater than a specific threshold (which is always a function of the cell diameter). Therefore, we only need an approximation of the displacement between the sequential frames. The latter may be wrong, but will be corrected in the global analysis. Because of this, the change in the cell location between two sequential frames will be considered as the displacement of the centroid, and the rotation is estimated as the change in the orientation of the cell.

Description (5.1) gives the numerical description of the incremental change in the cell location between two sequential frames. It includes the exact values of the displacement of the critical points which specify the cell location, as well as the direction of the motion

(considering the position in the first frame as the origin).

## 5.2.2 Symbolic Qualification Of Incremental Location Change

The incremental change in the cell location may be qualified and described symbolically by using the qualification rules described in Section 3.2.2. For example, the displacement of the cell (DL) between two sequential frames may be qualified as VERY SHORT, SHORT, MEDIUM, LONG, or VERY LONG. Two other qualifiers: STATIONARY and ARTIFACT are used to describe the stationary (no change in location) and artifactual movement of the cell. The latter could be due to undesired experimental conditions (these conditions are discussed in Section 4.2.5). These qualification rules are:

RULE(5.1):

```
IF d1 .LT. E0                ==then==> Q(d1) <--- STATIONARY
IF d1 .GE. E0  AND .LT. E1   ==then==> Q(d1) <--- VERY SHORT
IF d1 .GE. E1  AND .LT. E2   ==then==> Q(d1) <--- SHORT
IF d1 .GE. E2  AND .LT. E3   ==then==> Q(d1) <--- MEDIUM
IF d1 .GE. E3  AND .LT. E4   ==then==> Q(d1) <--- LONG
IF d1 .GE. E4  AND .LT. E5   ==then==> Q(d1) <--- VERY LONG
IF d1 .GE. E5                ==then==> Q(d1) <--- ARTIFACT
```

where d1 is the normalized value of DL(i,i+1), the displacement of the cell between frames i,i+1. E0, E1,..., E5 are threshold values (0-1) specifying the boundaries of each qualifier (see Section 4.6.3). The normalization of DL

may be obtained as:

$$dl = DL(i, i+1) / K \qquad (5.6)$$

where K is normalization factor, which is a function of the cell diameter and the time interval between sequential frames. Another method of normalizing the value DL is described in Section 4.6.4. Thus, by either using the local or global maximum and minimum values of DL, dl may be obtained as:

$$dl = \frac{DL(i, i+1) - DL(min)}{DL(max) - DL(min)} \qquad (5.7)$$

where DL(min) and DL(max) are the minimum and maximum possible values for the displacement of the cell between two sequential frames.

The direction of motion (DR) between two sequential frames may be qualified and described as EAST, EAST-NORTH, NORTH, WEST-NORTH, WEST, WEST-SOUTH, SOUTH, or EAST-SOUTH according to the following rules:

RULE(5.2):

IF DR .GT. 337 AND .LE. 22 ==then==> DR <--- EAST

IF DR .GT. 22 AND .LE. 67 ==then==> DR <--- EAST-NORTH

IF DR .GT. 67 AND .LE. 122 ==then==> DR <--- NORTH

IF DR .GT. 122 AND .LE. 157 ==then==> DR <--- WEST-NORTH

IF DR .GT. 157 AND .LE. 202 ==then==> DR <--- WEST

IF DR .GT. 202 AND .LE. 247 ==then==> DR <--- WEST-SOUTH

IF DR .GT. 247 AND .LE. 292 ==then==> DR <--- SOUTH

IF DR .GT. 292 AND .LE. 337 ==then==> DR <--- EAST-SOUTH

The incremental change in cell orientation (rotation, RO) may be described symbolically using two qualifiers. The first is used to describe the direction of the rotation as CLOCKWISE or ANTICLOCKWISE according to whether the value of RO less than or greater than zero. The second qualifier is used to describe the amount of rotation between two sequential frames as NO ROTATION, SLIGHTLY, PARTIALLY, CONSIDERABLE, or SIGNIFICANT. This qualification may be obtained by the following rules:

RULE(5.3):

```
IF   RO . LT. 0                ==then==> Q(RO) <--- CLOCKWISE
IF   RO . GT. 0                ==then==> Q(RO) <--- ANTICLOCKWISE

IF   RO . EQ. 0                ==then==> Q(RO) <--- NO ROTATION
IF !RO! . GT. 0  AND . LE. E1  ==then==> Q(RO) <--- SLIGHTLY
IF !RO! . GT. E1 AND . LE. E2  ==then==> Q(RO) <--- PARTIALLY
IF !RO! . GT. E2 AND . LE. E3  ==then==> Q(RO) <--- CONSIDERABLE
IF !RO! . GT. E3               ==then==> Q(RO) <--- SIGNIFICANT
```

Thus, the cell rotation between two frames (i) and (i+1) may be described as:

THE CELL HAS NO ROTATION.

or

THE CELL EXHIBITS SLIGHT ROTATION IN THE CLOCKWISE DIRECTION, THE ORIENTATION HAS CHANGED FROM OR1 TO OR2.

Description (5.1) is a typical example of the incremental change description between two sequential frames.

## 5.3 INCREMENTAL SHAPE CHANGE

The objective of this process is to detect and describe the incremental changes in the cell shape between two sequential frames. The process includes two main objectives:

(a) individual shape property change,

(b) membrane shape change and matching.

Description (5.1) is an example of the generated description of the shape and structural changes of a cell between two sequential frames. A description of the processes and algorithms which produce these results is given in the following sections.

### 5.3.1 Individual Shape Property Change

The objective is to compute the change in each shape property of the cell. These properties are: area, perimeter, length, width, number of convex angles, circularity, regularity, elongation, average bending energy, and intensity. The change may be computed by one of two methods: symbolic comparison (qualification) or numeric comparison followed by symbolic qualification.

The symbolic comparison method is based on comparing the symbolic qualifiers describing the same property in the frames under study. The purpose of the comparison is to detect if the property has the same qualifier in both frames, or if it has been changed. This comparison may be

described simply by the following rule:

RULE(5.4):

IF Q(Pj,i) .EQ. Q(Pj,i+1)

==then==> THE PROPERTY HAS THE SAME DESCRIPTION IN BOTH FRAMES.

IF Q(Pj,i) .NE. Q(Pj,i+1)

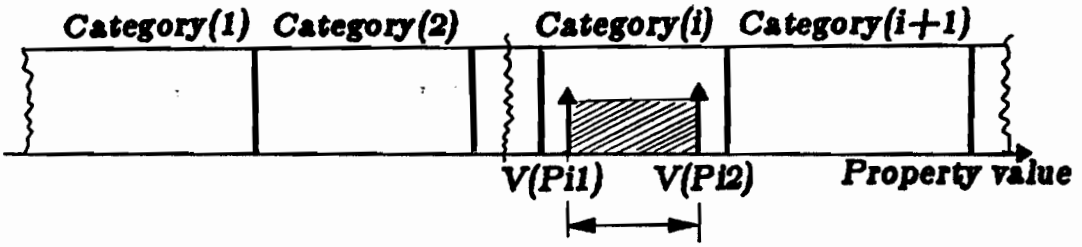
==then==> THE PROPERTY QUALIFICATION HAS CHANGED FROM Q1 AT FRAME(i) TO Q2 AT FRAME(i+1).

where Q1=Q(Pj,i) and Q2=Q(Pj,i+1) are the qualifiers of property (Pj) in frames (i) and (i+1), respectively. This method of incremental change detection and description has the advantage of being simple, fast, and does not include any numerical computation. On the other hand, it does not truly represent the exact change in the property value, as demonstrated by Figure (5.4). In this figure, we can see that symbolic comparison may result in no change, even for certain large magnitude changes in the property values. Sometimes these may be larger than differences which do produce qualification change.

The disadvantage of symbolic comparison can be avoided by first normalizing the exact change in the property (numerically), then using a symbolic qualifier to describe the computed change. This may be accomplished as follows:

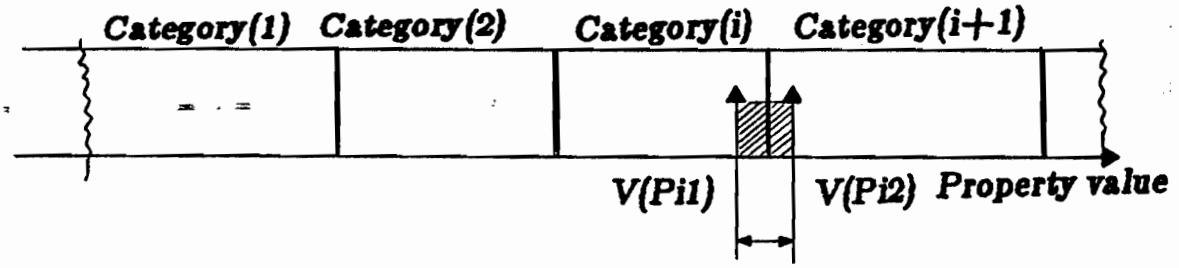
$$dP_j = \frac{| P_j(i) - P_j(i+1) |}{\max [ P_j(i) , P_j(i+1) ]} \qquad (5.8)$$

where dPj is the normalized value of the change in property (Pj) between frames (i) and (i+1). This value may be




**Exact change in the property value**

***(a) A symbolic comparison would not detect a change.***




**Exact change in the property value**

***(b) A symbolic comparison detects change, even though the magnitude is much smaller than shown in (a).***

*Figure(5.4) Disadvantage of a symbolic comparison to detect incremental change in a specific property.*

qualified and described symbolically as follows:

RULE(5.5):

IF dPj .LT. E1 ==then==> Q(dPj) <--- NO CHANGE

IF dPj .GE. E1 AND .LT. E2 ==then==> Q(dPj) <--- SLIGHTLY

IF dPj .GE. E2 AND .LT. E3 ==then==> Q(dPj) <--- PARTIALLY

IF dPj .GE. E3 AND .LT. E4 ==then==> Q(dPj) <--- CONSIDERABLE

IF dPj .GE. E4 ==then==> Q(dPj) <--- SIGNIFICANT

where E1, E2, E3, and E4 are the qualification thresholds which may be estimated in a similar way to that described in previous sections.

The above method has the advantage of describing symbolically the exact change in a specific property between two sequential frames. However, the normalization for a large sequence of frames may be costly in computation. Therefore, if the final goal of the analysis is the global behaviour of the property, a compromise may be made between the above two methods. This can be achieved by using the symbolic comparison to describe whether there is change in the qualification of the property, and the sign of the value Pj(i) - Pj(i+1) to describe whether its value has increased or decreased. This can be accomplished by using the following representational rules:

RULE(5.6):

IF Pj(i) - Pj(i+1) .GT. 0 ==then==> DECREASE

IF Pj(i) - Pj(i+1) .LT. 0 ==then==> INCREASE

An example is:

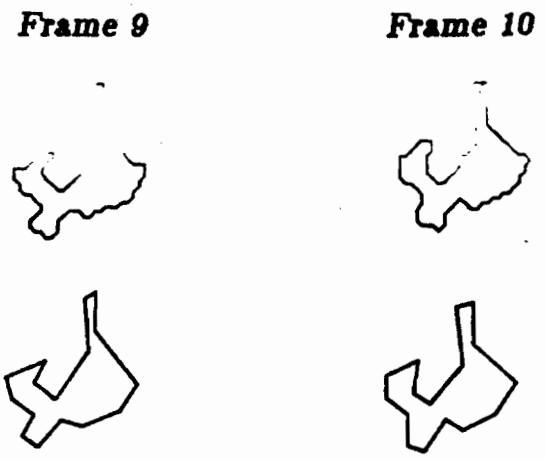
IF LENGTH(i) - LENGTH(i+1) .GT. 0 ==then==> Q(dP) <-- SHORTER

IF LENGTH(i) - LENGTH(i+1) .LT. 0 ==then==> Q(dP) <-- LONGER where SHORTER and LONGER are qualifiers for changes in any distance properties, such as length, width, or perimeter. Other qualifiers may be used according to the nature of the property, such as for example, SMALLER and LARGER for size properties, or LESS and MORE for other properties such as elongation, circularity, or regularity. An example of the generated description, using this method is: THE ELONGATION OF THE CELL DECREASED. Description (5.1) gives the complete description of the cell between two sequential frames.

It is worth mentioning here that the descriptions given in this section and the following ones are based on expert knowledge. They are motivated by discussions with Dr. P.B. Noble, Faculty of Dentistry, McGill University, about which type of information is sought, how much detail is desired, what are the most meaningful symbolic qualifiers.

## 5.3.2 Incremental Membrane Shape Change And Matching

In the preceding section, we demonstrated methods for detecting, quantifying, and describing the incremental changes in each of the individual shape properties. Now, given the shape of the cell membrane in two sequential frames, as shown in Figure (5.5), is there any change in the membrane shape?. The answer to this question will be the subject of discussion in this section.




*Figure(5.5) Two successive frames.*

*Figure(5.6a) Cell decomposition.*

*Figure(5.6b) Graph representation.*

The shape of the contour of an object (silhouette) is a function of all the properties describing it. Therefore, the change in a single property is insufficient to specify the change in shape. However, a function of the differences of some of the shape properties can be used to measure the change in membrane shape between the different frames. In this way, a comparison can be made between different shapes. The selection of these properties (which properties and how many?) is a very important issue, which will be discussed in detail in chapter 7 (Global Shape Analysis). However, it is desirable that these properties be translation, rotation, and size independent. Examples are circularity, regularity, average bending energy, and elongation. The change in each of these properties has a different effect on the membrane shape change. Therefore, weighting factors may be necessary in order to normalize the effect of different property change. Thus, the membrane shape change (MSC) may be computed as:

$$MSC = \left[ \sum_{k=1}^{k=m} \left| \frac{Pk(i) - Pk(i+1)}{Pk(i) + Pk(i+1)} \right| . Wk \right] / m \qquad (5.9)$$

where m is the number of selected properties and Wk is the weighting factor. The larger the value of MSC, the more change in the membrane shape, and vice versa. The percentage of membrane shape matching (MTCH) can be estimated as:

$$MTCH = ( 1 - MSC ) . 100 \ \% \qquad (5.10)$$

Thus, small values of MTCH indicate less matching of the

membrane shape between two sequential frames, and vice versa. For example, MTCH is equal to 100% if we match the membrane shape in a specific frame to itself.

In similar fashion, the numerical value of the percentage matching MTCH may qualified and described symbolically as follows:

RULE(5.7):

```
IF MTCH .LT. E1                ==then==> Q(MTCH) <-- QUITE DIFFERENT
IF MTCH .GE. E1 AND .LT. E2    ==then==> Q(MTCH) <-- DIFFERENT
IF MTCH .GE. E2 AND .LT. E3    ==then==> Q(MTCH) <-- ALMOST SIMILAR
IF MTCH .GE. E3 AND .LT. E4    ==then==> Q(MTCH) <-- SIMILAR
IF MTCH .GE. E4                ==then==> Q(MTCH) <-- VERY SIMILAR
```

## 5.4 INCREMENTAL STRUCTURAL CHANGE

In the previous section, we discussed the detection and quantification of incremental change in the membrane shape, as well as their percentage matching in two sequential frames. Incremental structural changes of the cell between two sequential frames will be discussed in this section. The process includes two main steps:

(a) structural matching of the subparts of the cell, in order to find the corresponding subparts between two sequential frames,

(b) determination of incremental changes in shape and geometrical properties of the corresponding subparts.

In static scene analysis, the cell is decomposed into its primitive subparts (Figure 5.6a), and then represented by a labeled graph as shown in Figure (5.6b). Given the topological properties of each subpart and their interrelationships in two sequential frames, we seek the following:

(a) To find which subpart in the second frame corresponds to which one in the first.

(b) For two corresponding subparts, detect the incremental changes in shape and relative position.

(c) Quantify and describe the results of (a) and (b) numerically and symbolically. Description#(5.1) and Table#(5.1) give typical examples of this result.

## 5.4.1 Subpart Matching

The problem of matching the different subparts of the cell in two sequential frames may be defined as follows. Given two sets of objects, it is required to recognize the corresponding elements (objects) from the two sets. In other words, if we have two sets {A} and {B} such that:

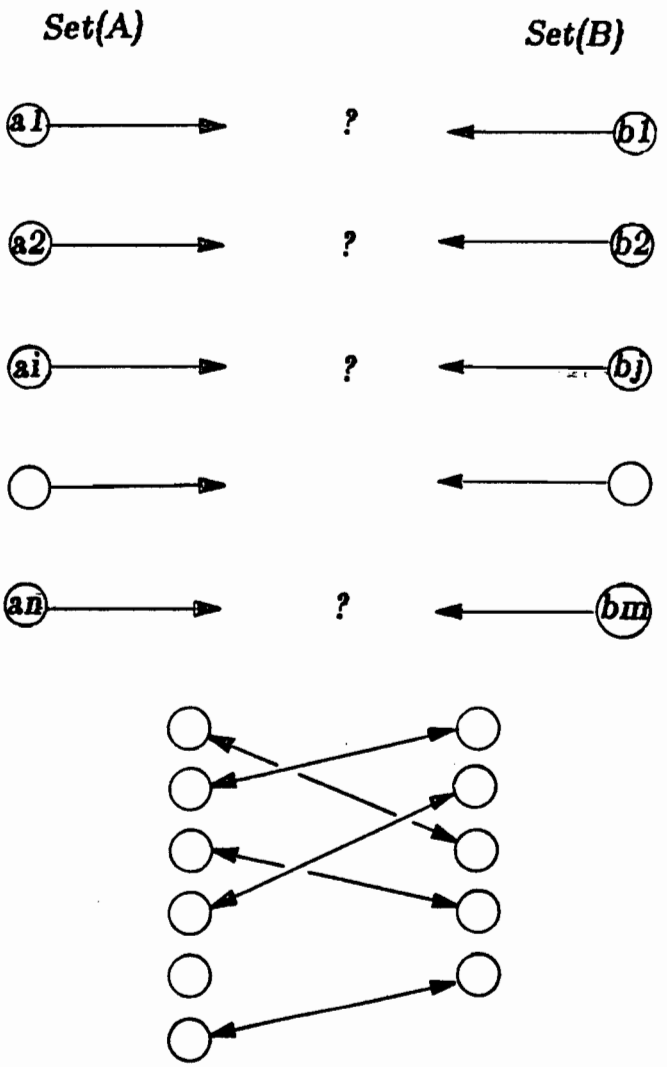
$$\{A\} = \{a1, a2, \ldots, ai, \ldots, am\}$$

and $\{B\} = \{b1, b2, \ldots, bj, \ldots, bn\}$,

the objective is to find the different elements of the set {A,B}

$$\{A,B\} = \{(a1, b1), (a2, b2), \ldots, (ak, bk), \ldots\},$$

where ak is an element of {A} and bk is an element of {B}. Figure (5.7) illustrates this problem and illustrates an arbitrary solution.




*Figure(5.7) Definition of the subpart correspondence problem.*

This problem can be considered as a simple pattern recognition procedure if: (a) both sets have the same number of elements, (b) the corresponding elements in both sets have the same property values. However, besides the standard problems of the pattern recognition in a static scene, we also experience other types of difficulties, due to the nature of cells as non-rigid objects. These difficulties are:

(a) Any slight change in the membrane shape can cause different decompositions.

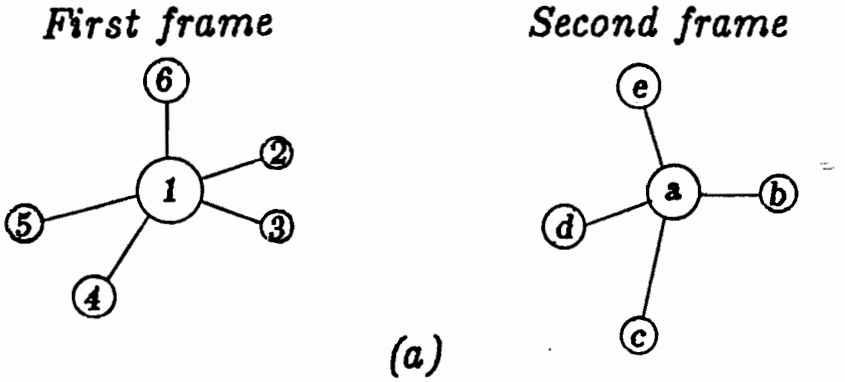
(b) The cell in both frames may not have the same number of subparts because pseudopods are continually growing and contracting.

(c) The same subpart of the cell may not have the same topological properties in subsequent frames.

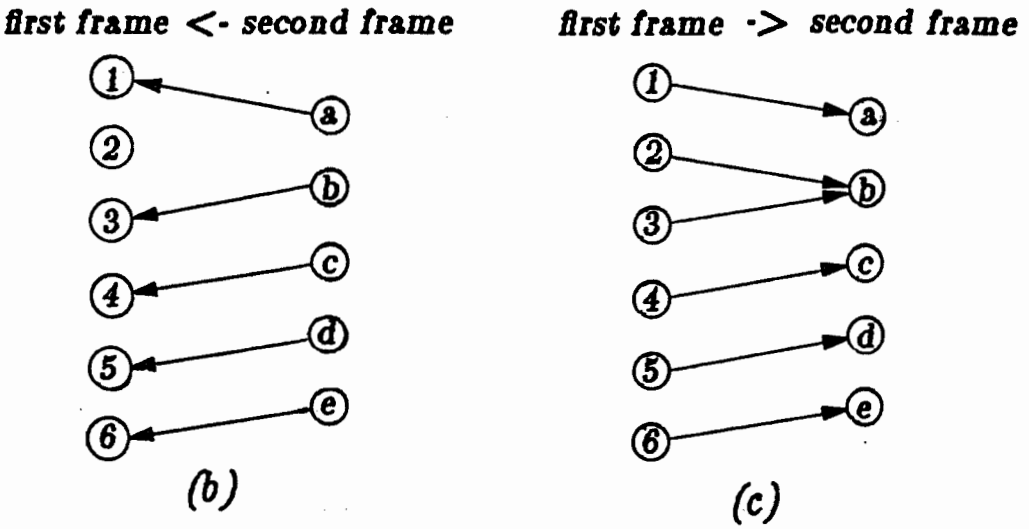
(d) The cell may have more than one subpart with the same simple shape but in different relative positions.

(e) Noise is caused by the registration of the different frames.

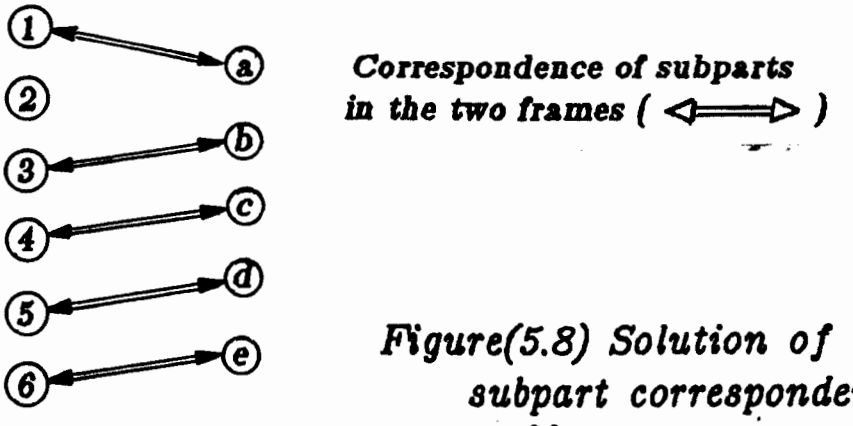
Considering the above difficulties, this section describes an algorithm for recognizing the corresponding subparts of the cell between two sequential frames. Figure (5.8a) shows the labeled graph of the cell in two sequential frames. The subparts of the cell in the first frame are labeled 1, 2, ..., n, and in the second frame a, b, ..., m, where n and m are the number of subparts of the cell in the first and second frames, respectively (in Figure 5.8a, n=6 and m=5). The main steps of the algorithm




(a)




(b) (c)




(d)

**Correspondence of subparts in the two frames ( ⟺ )**

*Figure(5.8) Solution of the subpart correspondence problem.*

are:

(1) Consider the subparts of the first frame as models (prototypes), and for each subpart (i) in the second frame find the subpart (j) in the first frame which gives the best match (Mmax), where:

$$Mmax = \max (Mi1, Mi2, \ldots, Mij, \ldots, Min) \qquad (5.11)$$

Mij is the match between subpart i (second frame) and subpart j (first frame), and is defined by

$$Mij = \left[ \sum_{k=1}^{k=m} \left| \frac{Pk(i) - Pk(j)}{Pk(i) + Pk(j)} \right| . Wk \right] / m, \qquad (5.12)$$

where m is number of subpart properties, and Pk(i),Pk(j) are the values of the property (k) for subparts i and j, respectively. Figure (5.8b) illustrates this match. The subpart properties which are used in this matching process are: area, perimeter, circularity, regularity, average bending energy, centroid coordinates, base-line, and connective-line, and orientation. We may note that the first six properties pertain to shape, whereas the rest specify the relative position of a subpart in the cell structure.

(2) Repeat step number(1), considering the subparts of the second frame as the models of the subparts in the first frame. Figure (5.8c) illustrates this step.

(3) Subparts i and j correspond if Mij = Mji. Thus, two subparts that have been matched in both steps (1) and (2) are said to correspond (see Figure 5.8d).

The result of the above algorithm is a set $\langle A, B, M \rangle$ of the corresponding subparts in the two frames and their percentage match. Thus,

$$\langle A, B, M \rangle = \{(a1, b1, m1), (a2, b2, m2), \ldots, (ai, bi, mi), \ldots, (al, bl, ml)\} \quad (5.13)$$

where ai is the number of the subpart in the first frame which corresponds to the subpart number bi in the second frame, with percentage match equal to mi. This result may be described symbolically as: BLOB NUMBER i IN THE SECOND FRAME CORRESPONDS TO BLOB NUMBER j IN THE FIRST FRAME, or BLOB(S) NUMBER (.....) IN THE SECOND FRAME DO(ES) NOT CORRESPOND TO ANY IN THE FIRST FRAME. Table (5.1) gives an example of the result of computing the correspondence matching of the different subparts between two sequential frames. This result is summarized symbolically in Description (5.1).

## 5.4.2 Incremental Changes In Corresponding Subparts

In the preceding section, we described how to compute the correspondence between different subparts of the cell in two sequential frames. The changes in the topological properties of these subparts and their relationships to the global changes of the entire cell are the basic elements for understanding the dynamic behaviour of the cell. Measurement, qualification, and description of the changes in the corresponding subparts between the sequential frames, are described below.

The computation of the incremental change between the corresponding subparts is very similar to the computation of the incremental change in the location and membrane shape of the entire cell (which are described in Sections 5.2 and 5.3). The differences are: (a) the incremental changes are computed for each two corresponding subparts instead of the entire cell, (b) besides the changes in the shape and location, in the case of the subparts , the changes in the topological properties (structural relationships of the different subparts) are also computed. Thus, the different properties which are considered for the changes in the different subparts are:

| Shape Properties | Structural Properties |
|---|---|
| area | centroid coordinates |
| perimeter | base-line |
| circularity | connective-line |
| regularity | orientation |
| elongation | |
| relative area | |
| average-bending-energy | |

An example of the exact values of the change in each of the above properties between the corresponding subparts of two sequential frames are given in Description (5.1). These values are normalized as follows:

$$DPi = \frac{|dPi|}{\max [Pi(t), Pi(t+1)]} . \qquad (5.14)$$

As before, the changes in the different properties of the corresponding subparts can be qualified and described symbolically as: NO CHANGE, SLIGHTLY, PARTIALLY, CONSIDERABLE, SIGNIFICANT (see Description 5.1).

In systems where incremental change detection is an intermediate form of data to a high level analysis, the above descriptions may give more information than is required at this stage. Usually, this stage is only necessary to describe whether a change in property value has taken place, and if so, whether a decrease or increase has occured. Therefore, to obtain only the required information, make the description simpler, and to save unnecessary computation, our symbolic description of the incremental changes in the different subparts is generated as shown in Table 5.2.

## 5.4.3 Total Structural Matching

The total structural match (SM) of the cell between two sequential frames is computed as a percentage value (0-100%) which depends on the number of corresponding subparts and their percentage match. Thus, SM will equal to 100% if (a) the cell has the same number of subparts in both frames, (b) there is a one to one match between all subparts, and (c) there is no change in any of the subpart properties. The

structural match of the cell may equal 0% if there are no corresponding subparts between the two frames. The value of SM may be computed as:

$$SM = (M1 + M2 + \ldots + Mk) / m , \qquad (5.15)$$

where M1, M2, ..., Mk is the percentage match of the corresponding subparts (see equation (5.12)), k is the number of the corresponding subparts, and m the number of the subparts in the second frame. The latter is used if we are matching the structure of the cell in the second frame to that in the first frame. In the case where there is no order between the two frames, the value m should be replaced by (n+m)/2, where n the number of subparts in the first frame.

The total structral match of the cell may be described symbolically as QUITE DIFFERENT, DIFFERENT, ALMOST SIMILAR, SIMILAR, or VERY SIMILAR. Thus, looking at the cell in the two successive frames given in Figure (5.5), it appears that they are similar. This judgement will change after comparing the labeled graphs that represent the cell in both frames (Figure 5.6). Obviously, there are changes in shape and structure. Using the analysis described in the preceding sections, these changes are detected, qualified, and presented in a summary, such as Description (5.1).

## 5.5 SUMMARY

In this chapter, we presented processes for detecting, qualifying, and describing the incremental changes in the location, shape, and structure of a moving cell. First, the difference between the motion of rigid objects and non-rigid objects was discussed. The change in location was computed in terms of the displacement and rotation of the cell. Second, the changes in each of the shape properties were computed in order to estimate the change in membrane shape. Based on the latter, the matching of the cell shape between two frames was computed as a percentage value. Third, the change in cell structure was computed in terms of the correspondence of different subparts of the cell. Finally, changes in shape and relative location of corresponding subparts were computed in order to quantify and describe the structural matching of the cell in successive frames.

The procedures of most of the processes presented in this chapter are based on comparisons of the symbolic qualifiers of the different properties in the two frames under consideration. In some cases, however, in order to produce a precise description of the changes, a comparison of the numerical property values in both frames is first estimated, from which a symbolic qualifier is determined. The symbolic qualifiers which describe the changes in the different properties are chosen by representational rules, as described in the previous chapters.

Structural matching of subparts is very important in understanding the dynamic behaviour of the moving cell. This is because, using this data, we can estimate the time when a specific subpart started to grow (appear) or contract (disappear). Also, the study of changes in the topological properties of the different subparts is important in order to recognize if a specific subpart is a candidate for a pseudopod or not, and to quantify the behaviour of the different pseudopods. These issues are the subject of thorough study in Chapter 8.

## TABLE(5.1) MATCHING OF SUBPARTS BETWEEN FRAME 9 and 10

### (A) MATCHING FRAME# 10 TO FRAME# 9:

| | FRAME# 10 | ======> | FRAME# 9 | MATCH |
|---|---|---|---|---|
| SUBPART# | 1 | | 3 | 0.892 |
| SUBPART# | 2 | | 1 | 0.842 |
| SUBPART# | 3 | | 2 | 0.918 |
| SUBPART# | 4 | | 3 | 0.926 |
| SUBPART# | 5 | | 4 | 0.938 |

### (B) MATCHING FRAME# 9 TO FRAME# 10:

| | FRAME# 9 | ======> | FRAME# 10 | MATCH |
|---|---|---|---|---|
| SUBPART# | 1 | | 1 | 0.867 |
| SUBPART# | 2 | | 3 | 0.918 |
| SUBPART# | 3 | | 4 | 0.926 |
| SUBPART# | 4 | | 5 | 0.938 |

### (C) CORRESPONDENCE OF SUBPARTS:

| | FRAME# 10 | ======> | FRAME# 9 | MATCH |
|---|---|---|---|---|
| SUBPART# | 1 | | NONE | 0.0 |
| SUBPART# | 2 | | NONE | 0.0 |
| SUBPART# | 3 | | 2 | 0.918 |
| SUBPART# | 4 | | 3 | 0.926 |
| SUBPART# | 5 | | 4 | 0.938 |

# TABLE(5.2)

| Change in Property | Value of Change | Type of Change | Change Description | Nature of Property |
|---|---|---|---|---|
| | 0 | STATIONARY | NO CHANGE | AREA |
| | | | | PERIMETER |
| | | | SMALLER | CIRCULARITY |
| dP = | -VE | DECREASE | SHORTER | REGULARITY |
| | | | LESS | ELONGATION |
| | | | | AV-BEN-ENG |
| | | | LARGER | BASE-LINE |
| | +VE | INCREASE | LONGER | CON-LINE |
| | | | MORE | REL-AREA |

where dp is the value of the incremental change in a specific property. Description (5.1) is a sample of a typical description of the incremental changes between two successive frames.

## DESCRIPTION <5.1>

# INCREMENTAL CHANGE DESCRIPTION

## OF THE CELL IN FRAMES : 9 AND 10

## LOCATION

### DISPLACEMENT :

The centroid of the cell has moved a VERY SHORT distance (approximately ONE-TENTH of the cell diameter) in a WESTERLY direction.

### ROTATION

There was SLIGHT rotation in the ANTICLOCKWISE direction

### ORIENTATION

The orientation of the cell has changed from NORTH to NORTH-EASTERLY

## SHAPE

The description of the change in the shape is given below in two parts: (1) the global change of the membrane shape, and (2) the structural changes in the primitive parts of the cell and their interrelationships.

### MEMBRANE SHAPE CHANGES

The general matching of the cell's shape between the two frames is VERY GOOD. This is due to the change of the main shape properties which can be described as follows.

There is NO change in the COMPACTNESS, ELONGATION REGULARITY, SIZE, and the PERIMETER of the cell.

There is a SLIGHT change in the COMPLEXITY of the cell; it became LESS COMPLEX.

## STRUCTURAL CHANGES

The structure of the cell in the two frames is ALMOST SIMILAR. This conclusion is based on a comparison of the primitive parts of the cell in the two frames. A detailed description is given below.

## DETAILS

Blob numbers(1,2) in the second frame DO NOT correspond to ANY blobs in the first frame.

Blob number(2) in the first frame CORRESPONDS to blob number(3) in the second frame. The latter has a LARGER SIZE, LONGER PERIMETER, and a LONGER BASE-LINE. The CONNECTIVE-LINE is SHORTER, and is ROTATED in ANTICLOCKWISE direction. The SHAPE is LESS REGULAR.

Blob number(3) in the first frame CORRESPONDS to blob number(4) in the second frame. The latter has a LARGER SIZE, the SAME PERIMETER, and a SHORTER BASE-LINE. The CONNECTIVE-LINE is SHORTER, and it is ROTATED in a CLOCKWISE direction. The SHAPE is MORE REGULAR.

Blob number(4) in the first frame CORRESPONDS to blob number(5) in the second frame. The latter has a LARGER SIZE, LONGER PERIMETER, and a LONGER BASE-LINE. The CONNECTIVE-LINE is THE SAME, and is ROTATED in a CLOCKWISE direction. The SHAPE is MORE REGULAR.

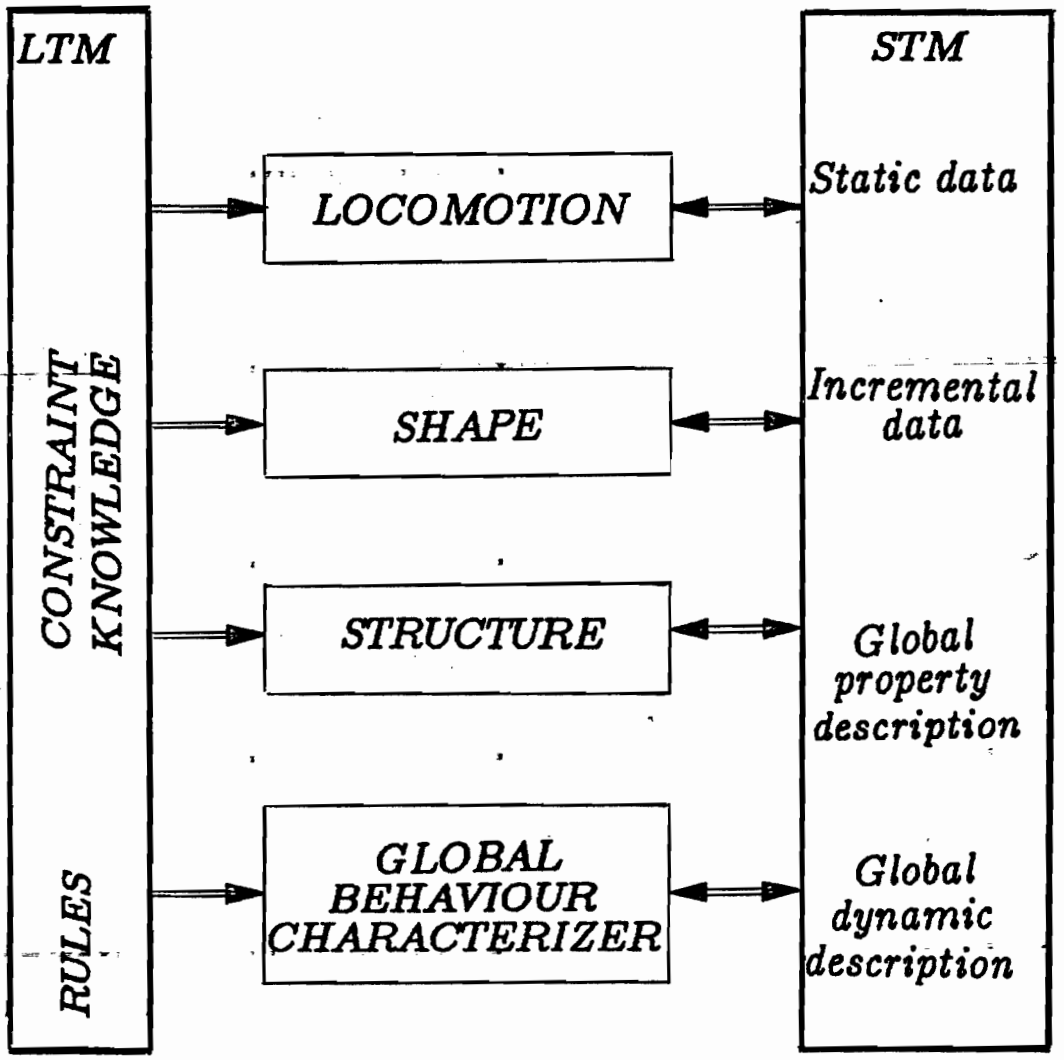
# CHAPTER 6

# GLOBAL LOCOMOTION ANALYSIS

## 6.1 INTRODUCTION

The main objective of the present system is to understand and describe the dynamic behaviour of a non-rigid moving object from a sequence of pictures. To achieve this objective, three stages of analysis have been defined: static scene analysis, incremental change detection, and global analysis. The first two stages are described in chapters 4 and 5 respectively. The global analysis (the third stage) presents the highest level in the hierarchy of the system. The objective is to analyze the multitudinous data which is extracted from the static and incremental analysis in order to detect and describe the global changes from the irrelevant and noisy ones. Figure (6.1) shows the main processes and data structure of the global analysis stage.

The global analysis will be described in three Chapters (6-8). First, the global locomotion of the object will be analyzed and described in this chapter. Second, the global changes in the shape will be discussed in Chapter 7. Finally, Chapter 8 involves two basic issues. The first




*Figure(6.1) Main processes and data structure of the global analysis stage.*

pertains to the global structural changes, and the second is concerned with the integration of the three aspects pertaining to the locomotion, shape, and structure in order to generate a coherent description of the dynamic behaviour of the moving object.

The global locomotion analysis of a moving object involves two basic steps. The first is responsible for motion detection (tracking) by locating the object in each image of the sequence, and detecting the incremental change in location. The second step is concerned with motion analysis. The output of the former is the path which represents the object movements, whereas the motion analysis should provide a description of the motion pattern or behaviour.

Most of the early work in dynamic scene analysis was mainly concerned with motion detection. Constructing knowledge-based systems for motion analysis, understanding, and description is the recent trend in image sequence analysis. A review of the significant work and evaluation of the current status of the gained experience in this field is presented in Section 2.2. The system under discussion is designed as a rule-based system for understanding the dynamic behaviour of a moving cell.

Cell movement is a fundamental process of some importance to host defense mechanisms. Most of the research in understanding the mechanisms which regulate these

processes was concentrated in cell locomotion and chemotaxis analysis. The latter is the response of a motile cell to the directional influence of external factors, such as bacteria, tumour or chemical substances. The goal of these studies is to provide answers to some of the basic questions about the cell locomotion, such as :

(a) How does the cell move from point A to point B ?
Does it move in a straight line ? Curved path ?
Zig-zag trajectory ? Any specific pattern ?
Does the cell exhibit any velocity or acceleration ?

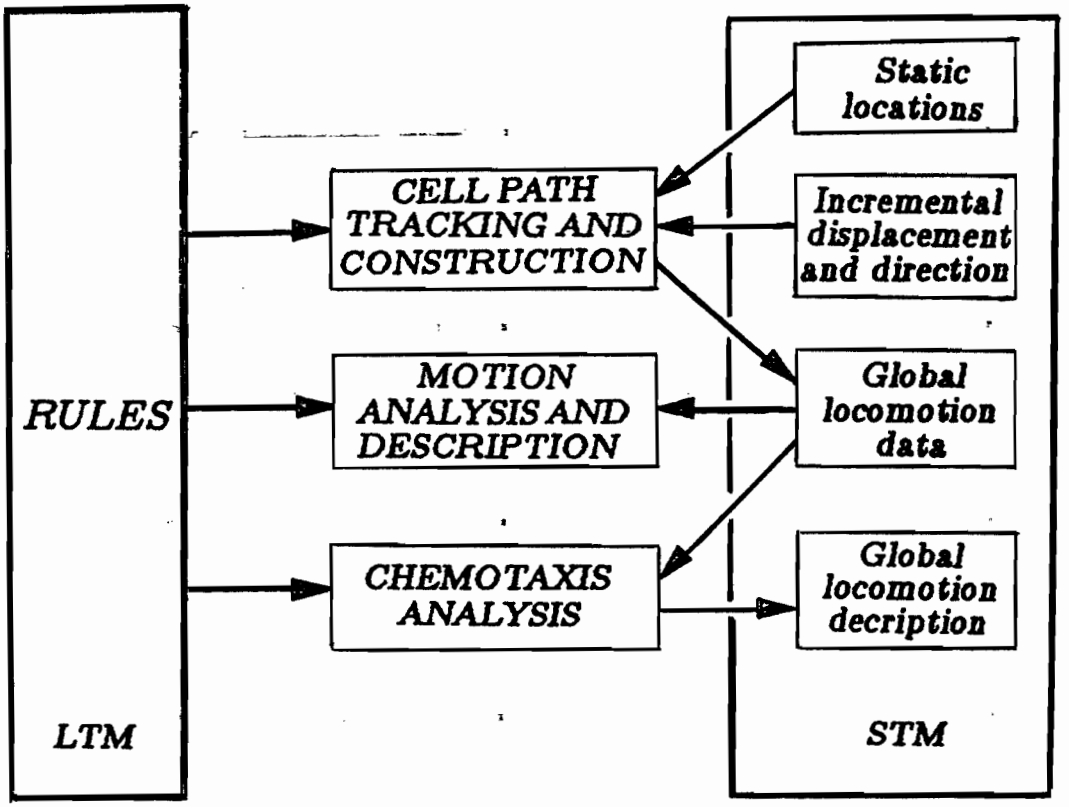
(b) Is the cell movement random or chemotactic ?
If it is chemotactic, is it positive or negative ?
From the analysis of the cell locomotion, can one predict the future behaviour of the cell ?
Can the cell behaviour be modified by changing any of the environmental conditions ?

(c) What is the role of the cell surface in regulating the social behaviour of the cell?
Is there any relationship between changes in the membrane shape and/or structure and the cell locomotion?

Most of the previous work in cell motion analysis was restricted to cell tracking either by manual or automatic production of the cell path and/or quantifying its data in order to provide answers to the questions in group (a) above. The recent work by [Levine et. al, 80] was different in that they computed the steady-state

probabilities of a cell moving in a particular direction. Their analysis provided answers to questions in group (b) above. In our current research, the cell locomotion analysis is designed to provide a description of dynamic cell behaviour. In this way, questions in group (c) may be answered.

Processes for locating the moving cell in each frame of the sequence and computing the incremental displacements (motion detection) are described in Chapters 4 and 5. In the remainder of this chapter we will describe processes for global locomotion analysis and description. Figure (6.2) shows the global locomotion analysis processes and data structure. The input data for this stage of the system are the static location of the cell at each single frame, the incremental displacement, and the direction of motion between two sequential frames. The output is a description of cell locomotion behaviour. A typical example of the generated summary is given in Description (6.1). In the following sections, the global cell tracking and path construction is described. The motion analysis and description is discussed in Section 6.3. Section 6.4 is concerned with the quantification and description of chemotaxis behaviour. Finally, in Section 6.5, a summary of this chapter is presented.




*Figure(6.2) Global locomotion analysis processes and data structure.*

## 6.2 CELL TRACKING AND PATH CONSTRUCTION

A cell path is the trajectory of its locomotion. It can be constructed by connecting the centroid points of the cell at the different locations of its movement. The simplest method of constructing a cell path is by recording the X and Y coordinates of the centroid at a constant time interval, and then connecting these points in the same sequence as recorded. This produces a cell path ⟨P⟩ where

$$P = \langle P1, P2, \ldots, Pi, \ldots, Pm \rangle, \qquad (6.1)$$

and m is the number of points in the path.

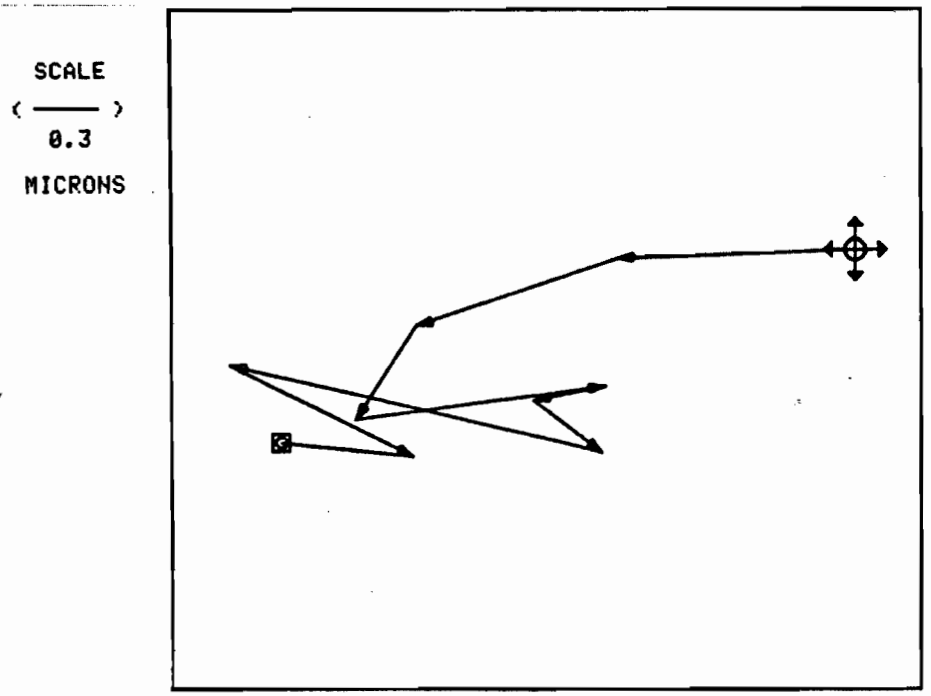
In image sequence analysis the time interval between two successive points in the cell path is the time interval between two sequential frames. Figures (6.3a), (6.3b), and (6.3c) show typical cell paths sampled at the minimum time interval (the filming rate .5 seconds). This method is adequate for analysis of cell movement for a short period of time where the detail of each incremental displacement is desired. But for long time periods, this method is not useful. The following reasons may be cited:

(a) The path includes irrelevant movements which make the detection of the global changes more difficult.

(b) Sensitivity to noise.

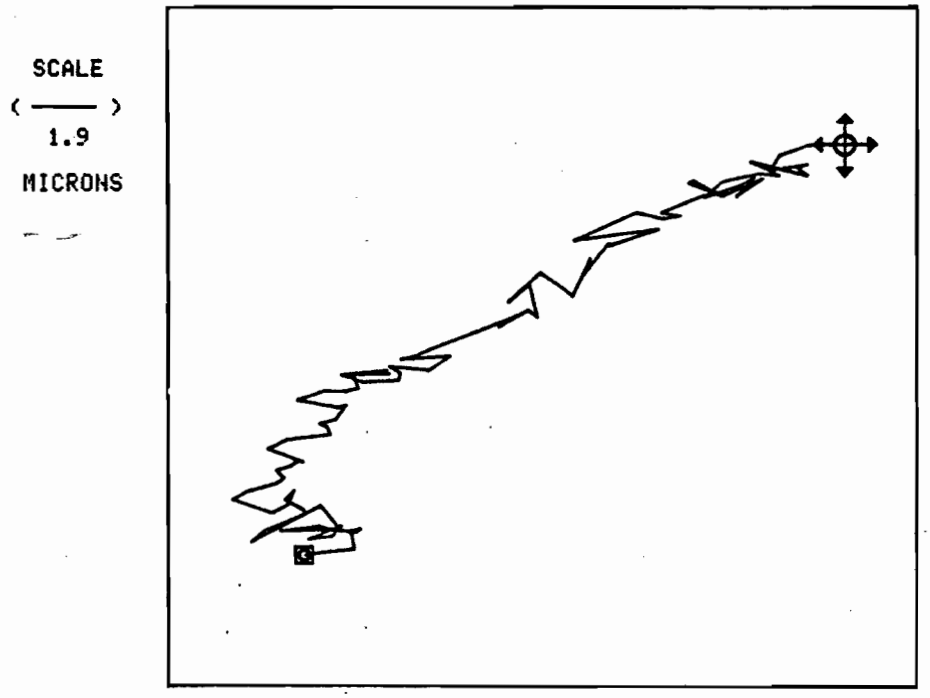
(c) The coordinates of the cell centroid are, to some extent, shape dependent.

**Frame number: First=1, Last=10, Sampling=1 frame (.5 seconds)**



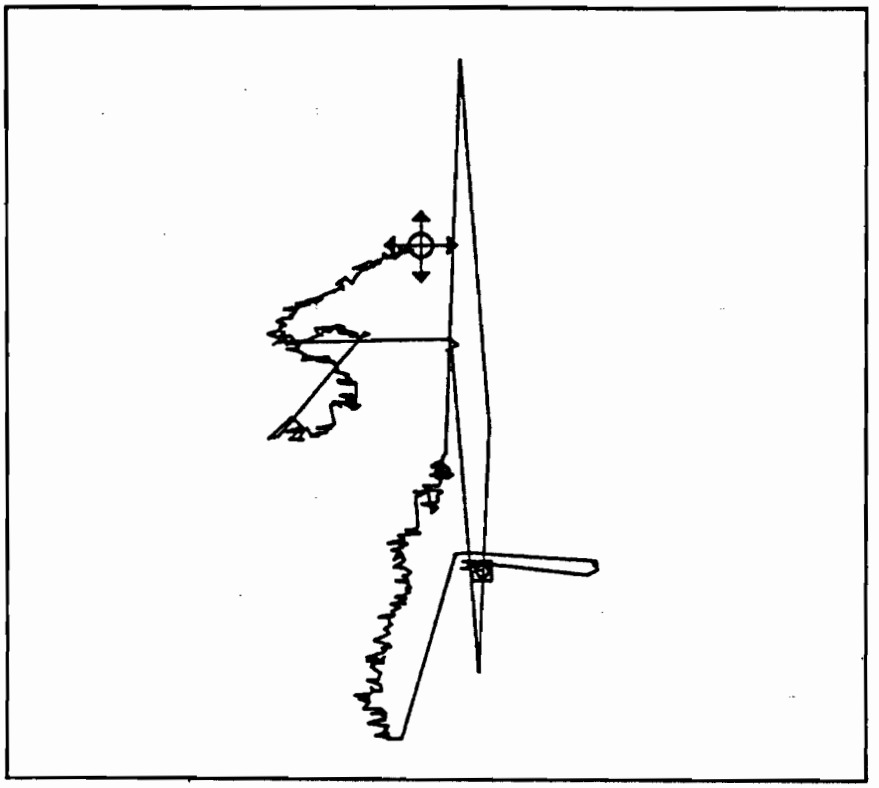

*Figure(6.3a) Time sampling of the cell path.*

**Frame number: First=1, Last=100, Sampling=1 frame (.5 seconds)**




*Figure(6.3b) Time sampling of the cell path.*

***Frame number: First=1, Last=450, Sampling=1 frame (.5 seconds)***




***Figure(6.3c) Time sampling of the cell path.***

For the purposes of global locomotion analysis, we should smooth, simplify, and at the same time retain all the global changes of the cell path. To do that, we may consider two methods, time and distance sampling. A description of each method is given below.

In the construction of the cell path using time sampling, we sample the path using longer time intervals (Tm), where Tm = m. to, and to is the time interval between two sequential frames. Thus, we consider the location of the cell in each of m frames. This will reduce the number of path points by a factor of m, but it will not smooth it. This is especially true when the cell is stationary or in motion with very short displacements. Therefore, we may consider the distance sampling method, which is described below.

## 6.2.1 Cell Path Construction Using Displacement Sampling

This method is based on the assumption that the cell has moved only if it exhibits a displacement which exceeds a specific threshold (Ed). This threshold is a function of the cell diameter. Thus,

$$Ed = K \cdot CD, \qquad (6.2)$$

where CD is the average cell diameter, and K is a constant. To construct the cell path using a constant displacement threshold, we carry out the following steps:

(1) Take the centroid coordinates of the cell in the first frame (Xo, Yo) as the original point in the path P0.

(2) Find the first subsequent frame (i) where the distance (LS) between the centroid of the cells is greater than the threshold Ed, where

$$LS = [\ (Xo-Xi)^2 + (Yo-Yi)^2\ ]^{1/2} \quad . \qquad (6.3)$$

(3) Take the point (Xi, Yi) as p1 in the cell path

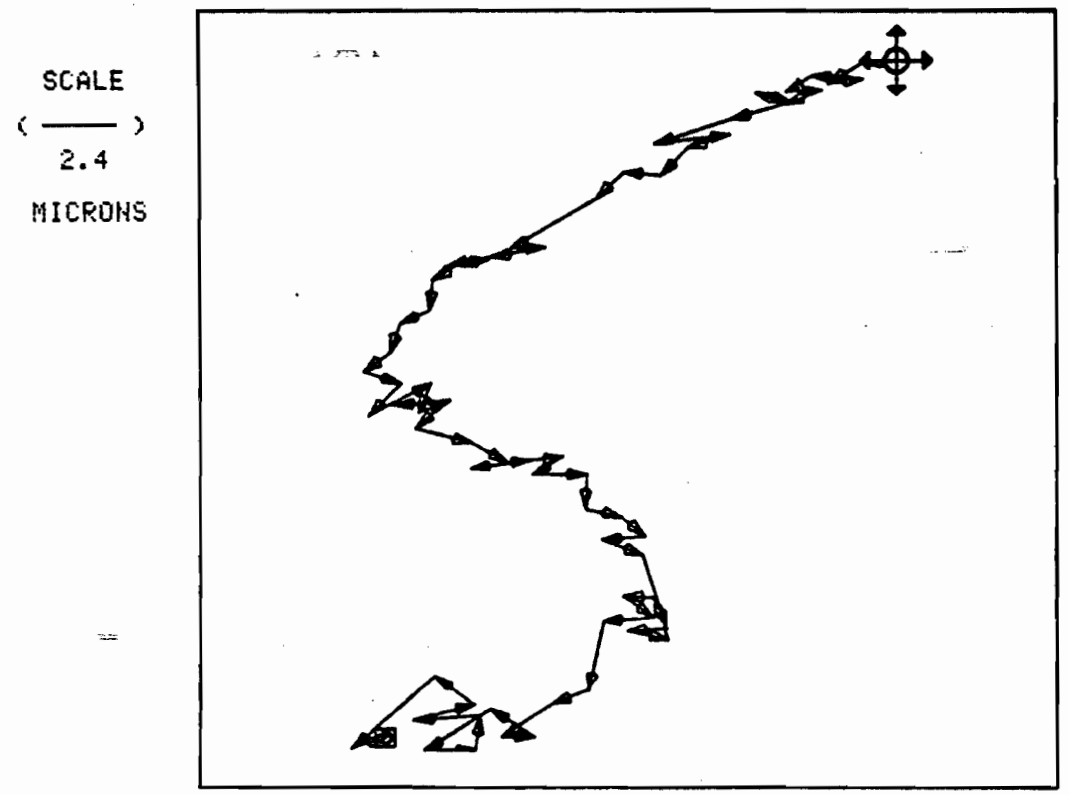
(4) To find points p2, p3, . . . , pn,

Put Xi ---> Xo , and Yi ---> Yo ,

and repeat steps (2) and (3)

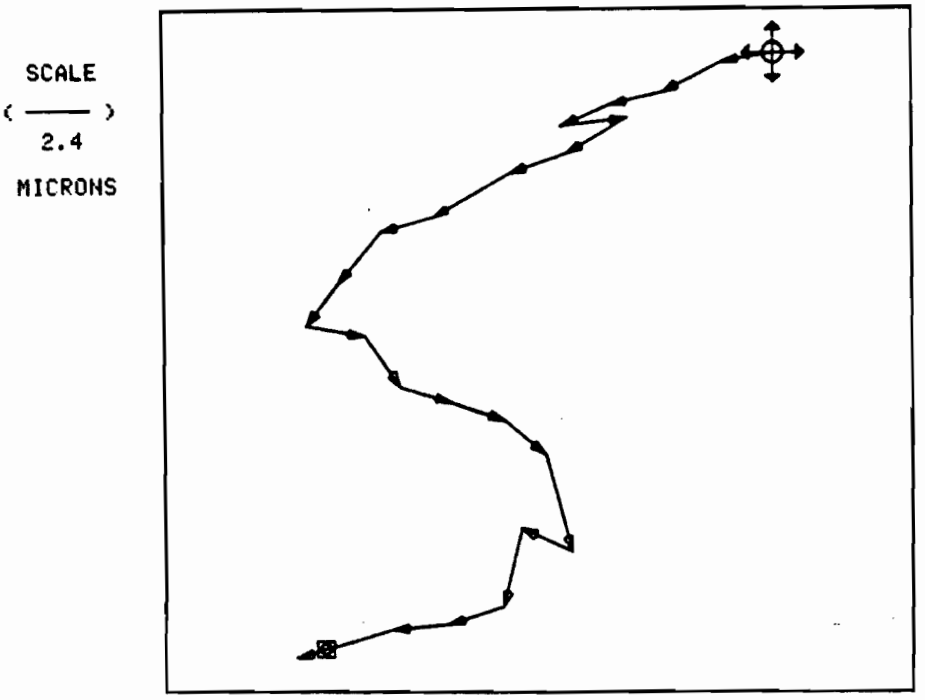
Figures (6.4a), (6.4b), and (6.4c) show the cell paths which were tracked for 200 frames (100 seconds) by using the thresholds E = 1, 2, and 3 microns, respectively. From these figures, one can see that by increasing the threshold (Ed), we increase the simplicity and smoothness of the cell path and at the same time retain the global changes. However, there is a limit to how much this threshold can be increased. At a certain point, we start to lose some of the detail of the cell movements (see Figures 6.4b and 6.4c). Fortunately, because of the slow motion of the cell, this threshold value is not very critical and may vary within a small range without affecting the resulting path. From our previous experience with cell tracking, the threshold has been chosen to be a quarter of the average cell diameter. Thus, E = .25 CD for the neutrophil cell, and this may be considered as constraint knowledge in the LTM.

***Frame number: First=1, Last=200, Sample distance=1 micron (1 pixel)***



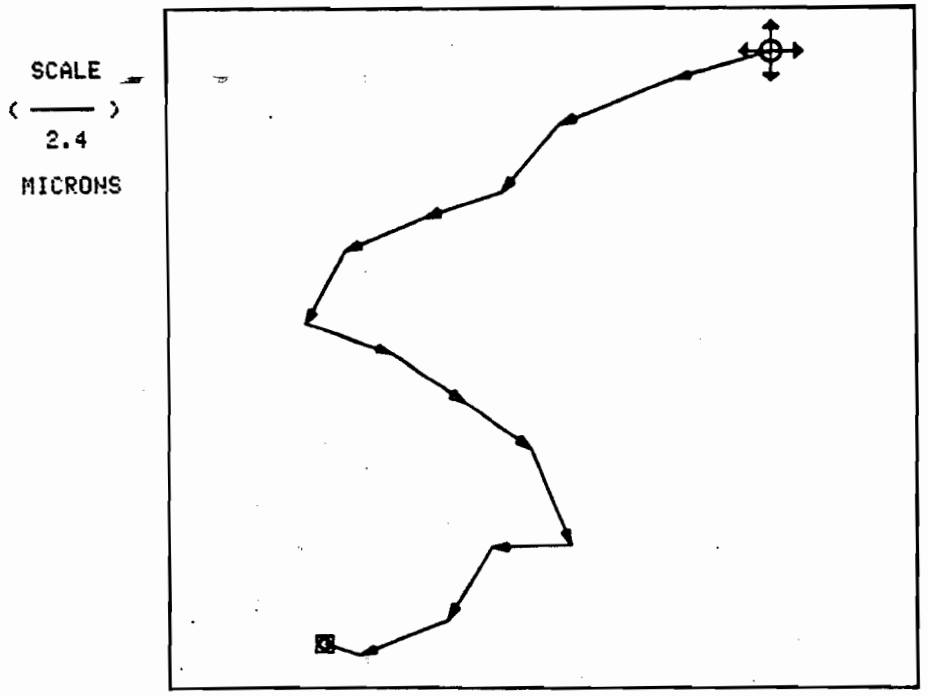

*Figure(6.4a) Distance sampling of the cell path.*

**Frame number: First=1, Last=200, Sample distance=2 microns (2 pixels)**




*Figure(6.4b) Distance sampling of the cell path.*

**Frame number: First=1, Last=200, Sample distance=3 microns (3 pixels)**




*Figure(6.4c) Distance sampling of the cell path.*

In the preceding section, we described how to construct a smooth and simple cell path by eliminating irrelevant or noisy movement, and retaining the global changes. However, in tracking the cell for a long period, we may face another problem in constructing the overall cell path. This problem is mainly due to undesirable experimental conditions, which cause discontinuity in the cell path. A description of this problem and its solution are given below.

## 6.2.2 Connection Of Cell Path Segments

In order to study the change in the cell shape and structure, it is necessary to film the cell at high magnification to get as much detail about its shape and structure as possible. Consequently, the viewing window of filming is reduced. Hence, after some time the cell may move out of this window. In order to keep the cell in view, either the slide containing the cells, or the camera should shifted to relocate the cell accordingly. The shift of the scene causes a sudden jump in the cell location between two sequential frames. This introduces problems for the system at two different levels. First, at the registration stage (automatic segmentation of the cell), and secondly, during global locomotion analysis. Solving this problem at the registration level is reported earlier in Section 4.2. In the remainder of this section we will show how this problem can be solved at a higher level.

The sudden jump of the cell can be detected from the sudden increase in the incremental displacement. In Figure (6.5a), the sudden jump of the cell appears as a long straight line between two segments of the path. In order to obtain a continuous path and to ignore any artifactual movement, we shift the path segments by vector translation of the cell locations in all the subsequent frames after the jump. For example, if there is a jump between frames (i) and (i+1), where (Xi,Yi),(Xi+1,Yi+1) are the locations of the cell before and after the jump, we compute the translation vector (shift) as:

$$X\text{-shift} = XS = Xi+1 - Xi \qquad (6.4)$$

$$Y\text{-shift} = YS = Yi+1 - Yi . \qquad (6.5)$$

Then, for frames i+1, i+2, . . . , n-1, n (n = number of the frames to be processed) the location of the cell will be changed to: (Xi+1+Xs, Yi+1+Ys), (Xi+2+Xs, Yi+2+Ys), . . . , (Xn+Xs, Yn+Ys). This action can be described by the following rule:

RULE(6.1):

IF L(i,i+1) .GE. Edd

==then==>(1) XS <-- Xi+1 - Xi AND YS <-- Yi+1 - Yi

(2) Xi+1 <-- Xi+1 + XS AND Yi+1 <-- Yi+1 + YS

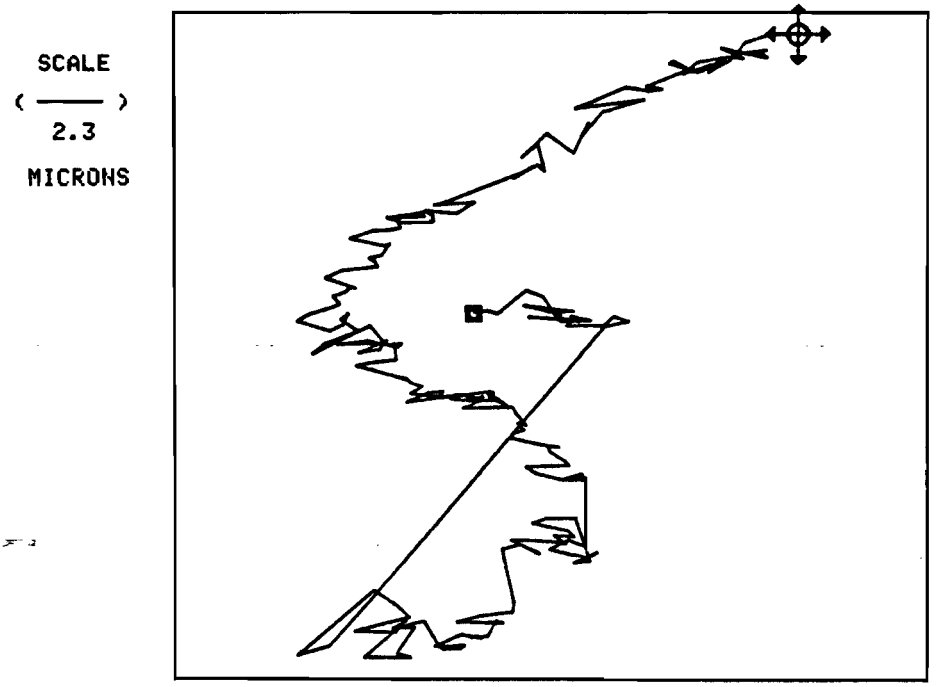
Xi+2 <-- Xi+2 + XS AND Yi+2 <-- Yi+2 + YS

.... .... ... .... .... ...

Xi+n <-- Xi+n + XS AND Yi+n <-- Yi+n + YS

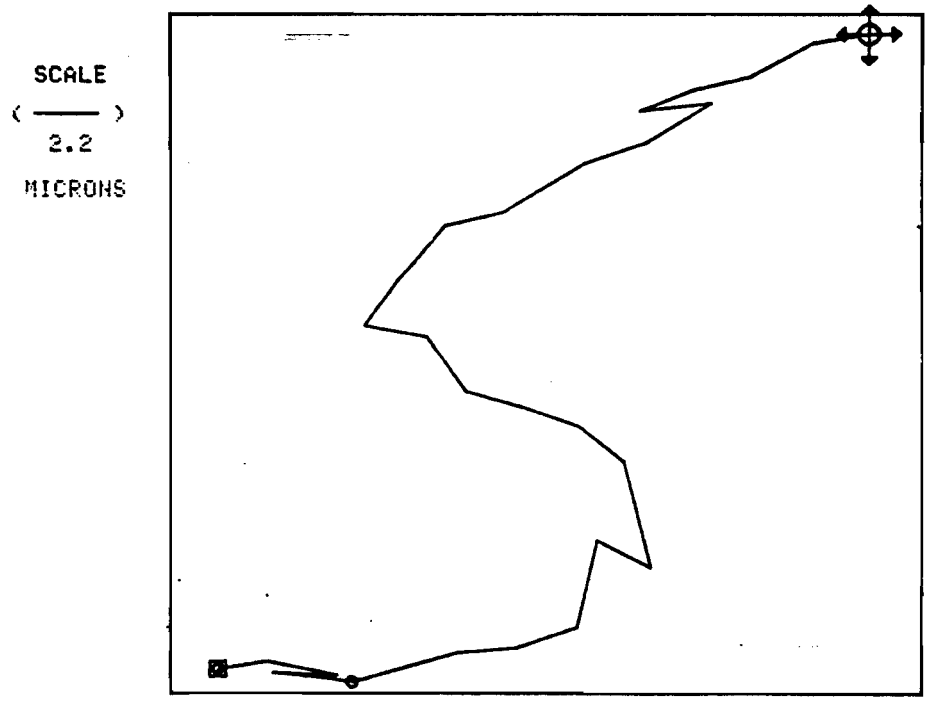
where n is the number of the frames to be processed, and L(i,i+1) is the displacement between two sequential frames. Edd is the threshold value which specifies the maximum

***Frame number: First=1, Last=225, Sampling=1 frame (.5 seconds)***




*Figure(6.5a) Original path (time sampling).*

***Frame number: First=1, Last=225, Sample distance=2 microns (2 pixel)***




***Artifact threshold taken as 6.0 microns***

*Figure(6.5b) Distance sampling with artifact removed.*

acceptable incremental displacement without artifactual movement.

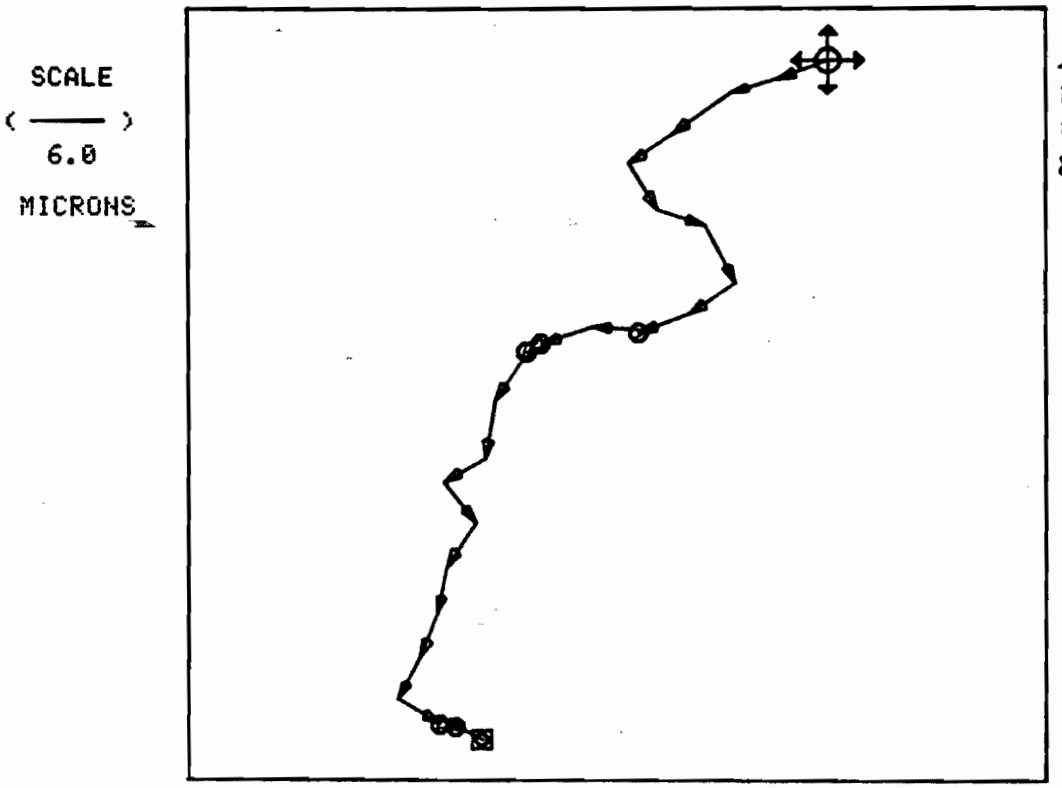
The above is an example of the use of the LTM rules in eliminating some of the undesirable experimental conditions. Figures (6.5a), (6.5b), (6.3c), and (6.5c) show cell paths before and after removing the gaps caused by the cell jump. In Figures (6.5b) and (6.5c) the locations where the cell jumps have been removed are marked with a circle.

In this section, we have described methods for pruning and integrating the large amounts of data which represent the locations and incremental displacements of a cell. This is accomplished by eliminating the irrelevant, noisy, and artifactual movement of the cell. The output of these processes is the path which represents the observable motion of the cell. The latter is further analyzed in order to generate a summary of the global locomotion. The processes which are responsible for generating this description, will be described in the following section.

## 6.3 MOTION ANALYSIS AND DESCRIPTION

In the preceding section we discussed methods for producing the path which represents the global motion of the cell. It consists of a sequence of steps. Each step is associated with a set of numerical values describing the motion properties such as: frame number, time, distance, direction, velocity, and acceleration. Thus, the global

***Frame number: First=1, Last=450, Sample distance=4 microns (4 pixels)***




*Figure(6.5c) Distance sampling with artifact removed.*

motion of a cell can be defined as a sequence of steps {S}, where:

$$\{S\} = \{S1, S2, \ldots, Si, \ldots, Sm\}, \quad (6.6)$$

and each element of this set Si represents a step of the global locomotion, with m the number of motion steps. Each element i is associated with a set of motion properties {P(Si)}, such that:

$$\{P(Si)\} = \{P1i, P2i, \ldots, Pki\} \quad (6.7)$$

In these experiments, the properties were chosen to be time, distance, direction, velocity, and acceleration. The determination of the frame number specifying the start of each motion step was described in the preceding section. Here, we will show how to compute and qualify the other motion properties.

## 6.3.1 Distance

In the locomotion analysis of a moving cell, three types of distances can be computed: total path displacement distance (TDD) (time sampled), total path locomotion distance (TLD) (distance sampled), and total translation distance (TTD) (vector sum of all locomotions). The definition and computation of each is given below.

### TOTAL PATH DISPLACEMENT - TIME SAMPLED (TDD)

The TDD represents the total movement of the cell, including those that are irrelevant, noisy, and random. This distance can be computed by adding all the incremental displacements

of the cell between sequential frames as follows:

$$TDD = \sum_{i=1}^{n-1} L(i, i+1) , \qquad (6.8)$$

where L(i,i+1) is the incremental displacement between two sequential frames, and n is the number of the processed frames. Figure (6.6) shows the total displacement distance of the cell at different times.

## TOTAL PATH LOCOMOTION - DISTANCE SAMPLED(TLD)

The TLD represents the movements which result in moving the cell an observable distance (exceeding a specific threshold). This distance is represented by the global locomotion path (see section 6.2). The TLD can be computed by adding all the lengths of each step:

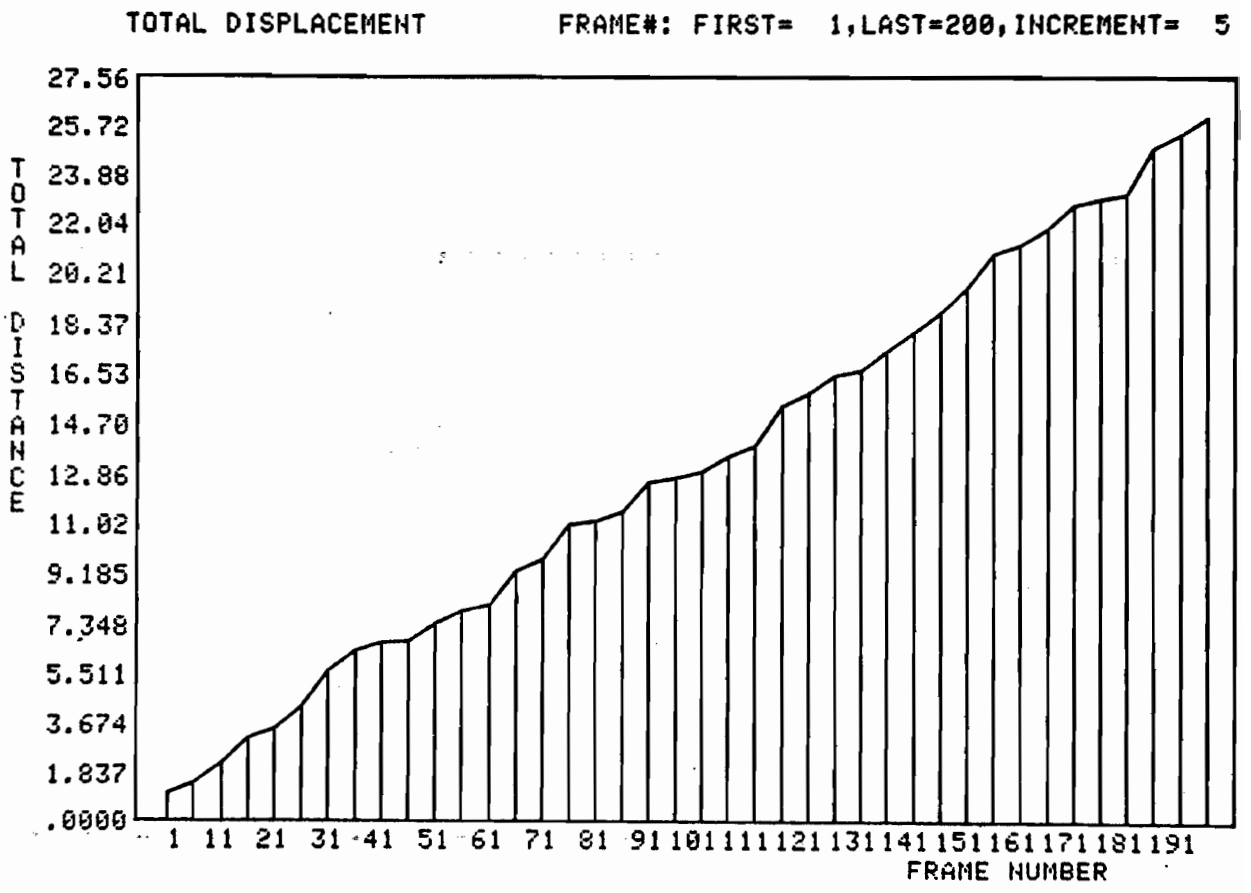
$$TLD = \sum_{i=1}^{m-1} LS(i, i+1), \qquad (6.9)$$

where m is the number of the path points, and LS(i,i+1) is distance between two sequential points:

$$LS = [ (X(Ni) - X(N(i+1))^2 + (Y(Ni) - Y(N(i+1))^2 ]^{1/2} \qquad (6.10)$$

## TOTAL TRANSLATION DISTANCE (TTD) - VECTOR SUM

The TTD is the distance between the locations of the cell in the first and last frames of the sequence under analysis. Thus, if (Xa,Ya),(Xb,Yb) are coordinates of the cell at the locations A,B (the first and last frames) respectively, then TTD is equal to the length AB and can be computed as:




***Figure(6.6) Total displacement of the moving cell.***

$$TTD = [ (Xb - Xa)^2 + (Yb - Ya)^2 ]^{1/2} \quad (6.11)$$

The TTD, in most cases, is less than the TLD, and the latter is less than the TDD; thus, TTD < TLD < TDD. This is because of the nature of the cell movement (the cell does not move in a straight line (see Figures 6.3-6.5). Analysis of the relationships between these distances is important in characterizing the randomness and chemotaxis behaviour of the cell. These aspects will be discussed in Section 6.4.

The distance of each locomation step of the global path can be qualified and described as: VERY SHORT, SHORT, AVERAGE, LONG, or VERY LONG. This qualification is based on a comparison with the average step distance (LSav), which can be computed as:

$$LSav = TLD / NS \quad (6.12)$$

where TLD is the total locomotion distance of the cell path and NS the number of steps. Thus, if a specific step has a distance that is close to LSav, it can be described as AVERAGE. This means that LSav specifies the data in the middle of the qualification range. Therefore, the value of LSav can be used as a normalization factor for the distance as follows:

$$LSni = LSi / 2.LSav \quad (6.13)$$

where LSni is the normalized value of the distance of step (i). Using the representational rules, the distance can be described symbolically as:

RULE(6.2):

IF LSni .LE. E1 ==then==> Q(LSi) <--- VERY SHORT

IF LSni .GT. E1 AND .LE. E2 ==then==> Q(LSi) <--- SHORT

IF LSni .GT. E2 AND .LE. E3 ==then==> Q(LSi) <--- AVERAGE

IF LSni .GT. E3 AND .LE. E4 ==then==> Q(LSi) <--- LONG

IF LSni .GT. E4 ==then==> Q(LSi) <--- VERY LONG

## 6.3.2 Time

The time of each motion step TSi is equal to

$$TSi = ( NF(i+1) - NF(i) ) \; to \qquad (6.14)$$

where (to) is the time interval between two sequential frames, and NF(i), NF(i+1) are the frame numbers where steps i and i+1 are started. The value of TSi can be normalized by comparing it to the average time of a motion step. This can be computed as:

$$TSni = Ti / 2Tav \qquad (6.15)$$

$$TSav = (n-1) . to / NS , \qquad (6.16)$$

where Tni is the normalized time of step i, n is the number of the processed frames, and NS is the number of the motion steps. The value Tni can be described as VERY SHORT, SHORT, AVERAGE, LONG, and VERY LONG using the following rule:

RULE(6.3):

IF Tn .LE. E1 ==then==> Q(Ti) <--- VERY SHORT

IF Tn .GT. E1 AND .LE. E2 ==then==> Q(Ti) <--- SHORT

IF Tn .GT. E2 AND .LE. E3 ==then==> Q(Ti) <--- AVERAGE

IF Tn .GT. E3 AND .LE. E4 ==then==> Q(Ti) <--- LONG

IF Tn .GT. E4 ==then==> Q(Ti) <--- VERY LONG

## 6.3.3 Direction Of Motion

The direction of the cell locomotion between two points in the global path can be computed as:

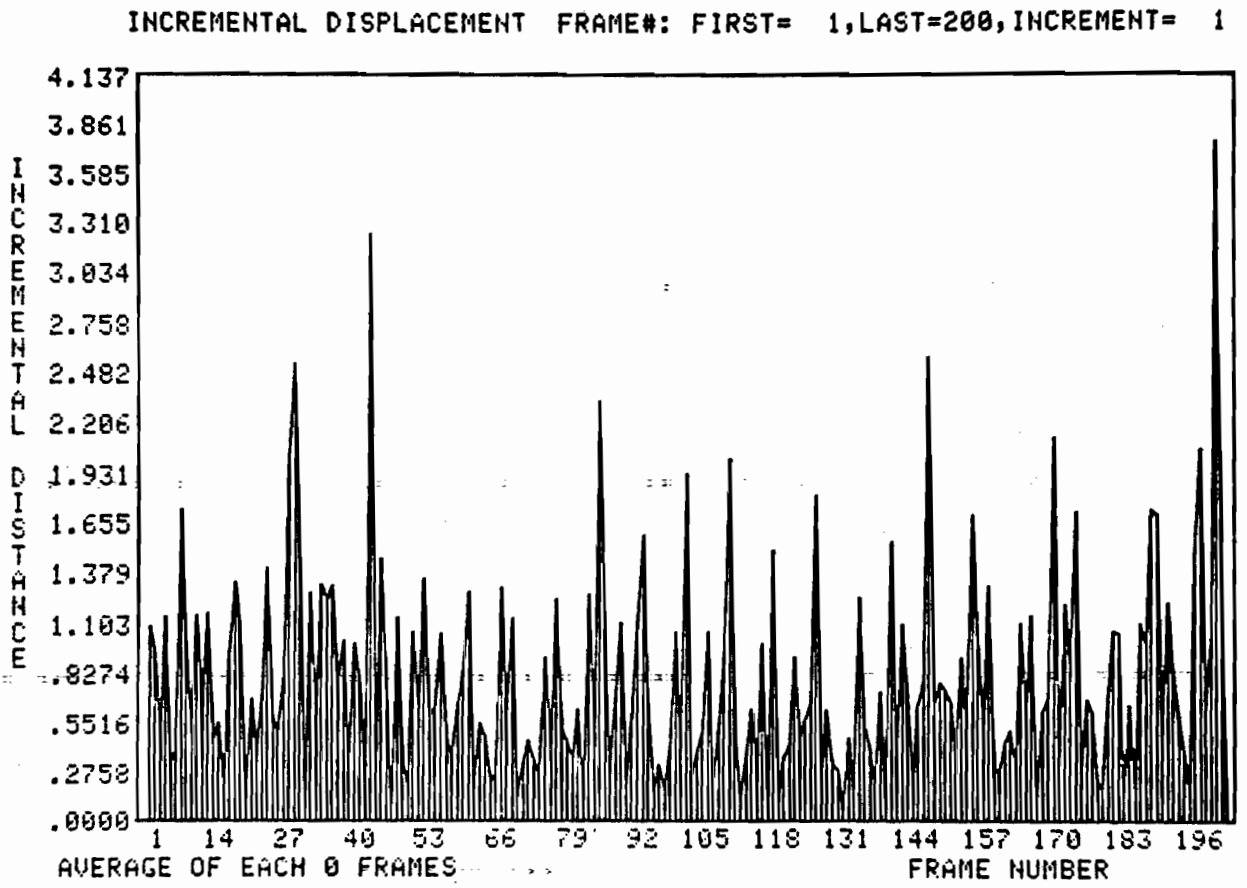
$$DRSi = \tan^{-1}\left[\frac{Y(NF(i+1)) - Y(NFi)}{X(NF(i+1)) - X(NFi)}\right] \qquad (6.17)$$

where X(NF(i+1)),X(NFi),Y(NF(i+1)),Y(NFi) are the X and Y coordinates of two sequential points in the cell path.

The direction DRSi can be described symbolically in terms of the main directions in the plane as: EASTERLY, NORTH-EASTERLY, NORTH, NORTH-WESTERLY, WESTERLY, SOUTH-WESTERLY, SOUTH, or SOUTH-EASTERLY.

## 6.3.4 Velocity

The basic definition of velocity is the rate of change in distance with time (dL/dt). In image sequence analysis, since the time interval between two sequential frames is constant, the incremental displacement can be considered as the velocity of the moving object. Figure (6.7a) shows the velocity (dL/dt) of a moving cell at different times. From this figure one can see that the cell has a random velocity. This randomness is due to two main factors, the nature of the cell movement, and the random change in shape which causes change in the centroid coordinates. In order to detect the global changes in velocity, we may consider two methods for smoothing the relation (dl/dt): averaging and sampling.




*Figure(6.7a) Velocity of the moving cell (dL/dt).*

In the averaging method, the velocity of the cell at specific time Vt, is considered as the average of the velocity from time (t) to the time (t+dt), thus,

$$Vt = [ \sum_{i=t}^{t+dt} Vi ] / dt , \qquad (6.18)$$

where dt is constant and depends on the type of moving object and the desired degree of smoothness of the curve. Figures (6.7b) and (6.7c) show the output of averaging the velocity using the above method on the data of Figure (6.7a) for dt=2.5 and 5 seconds (5 and 10 frames), respectively. After averaging the velocity, we can use a curve analysis technique to detect the points of velocity change.

This method gives adequate results in many cases. On the other hand, it includes two operations, averaging and curve analysis. Also with this approach, we are considering all cell movements, including those due to the change in shape or rotation. This can be avoided by using the sampling method, which is described below.

## VELOCITY CHANGE BY SAMPLING

In this method, the input is the cell path data produced by sampling at constant interval distance (see section 6.2). The cell path is defined as {S}, where:

$$\{S\} = \{S1, S2, \ldots, Sm\},$$

where m the number of points in the path. The velocity of the cell can be defined by {V}, where:

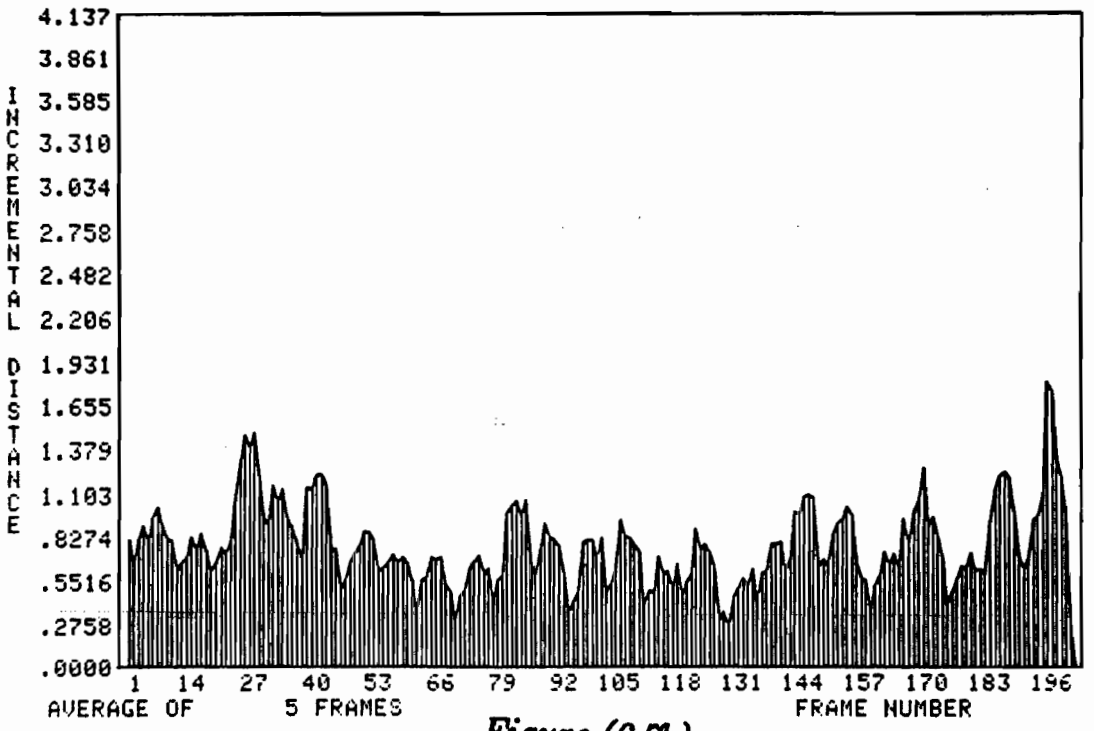



*Figure (6.7b)*

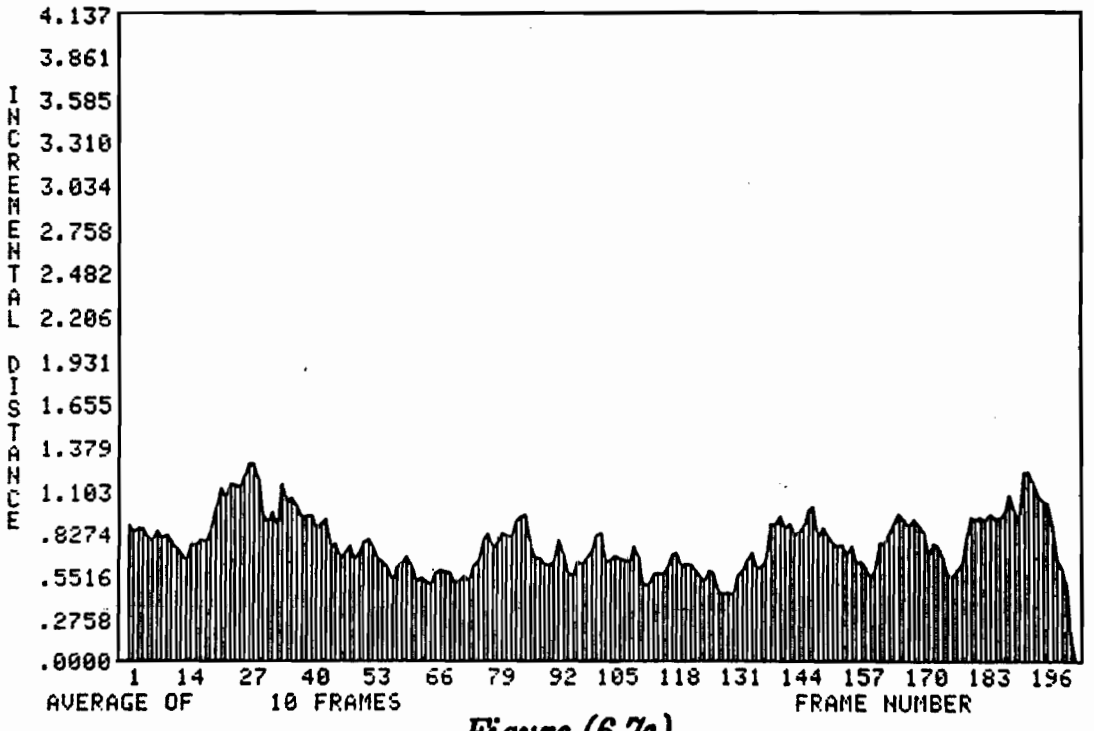



*Figure (6.7c)*

***Figure(6.7b-6.7c) Velocity of the moving cell in Figure (6.7a), where the velocity in each frame is represented by the average of sequence of: (b) 5 frames, (c) 10 frames***

$$\{V\} = \{V1, V2, \ldots, Vi, \ldots, V(m-1)\} \quad (6.19)$$

$$Vi = LSi / TSi \ . \quad (6.20)$$

Table(6.1) gives an example of the velocity of the cell computed for 450 frames in a sequence. This method for computing the velocity of the cell has the advantage that it is only a function of the cell translation (shape and rotation independent), and is less sensitive to noise. Moreover, the velocity may be computed simultaneous with the cell path sampling, thereby reducing the overall cost of computation.

In order to quantify and describe the velocity, we first compute the average velocity of the cell motion. Then, the latter can be used as the normalization factor as follows:

$$Vav = TLD / NS \quad (6.21)$$

$$VSni = VSi / 2.Vav \ , \quad (6.22)$$

where Vav is the average velocity of the cell, and VSni is the normalized value of the velocity at step (i). This can be utilized by the representational rules to describe the cell motion as: STATIONARY, VERY SLOW, SLOW, AVERAGE, FAST, VERY FAST

## 6.3.5 Acceleration

The acceleration (A) of a moving object is the rate of change velocity with time. Thus:

$$A = dV / dt = d2L / dt^2 \ . \quad (6.23)$$

Hence, if the velocity of the cell (dL/dt) is represented by a straight line, then the slope of this line represents its acceleration. In order to compute the acceleration, we can follow the same steps described in the preceding section for the velocity. There is one difference. That is, we compute the second derivative instead of (dL/dt). All the issues which have been discussed for the velocity are applicable to the acceleration. The two methods which were described above for the global velocity computation, can also be used for the acceleration. Therefore, only the basic steps for determining the acceleration will be given below. If the velocity is given as <V> such that:

$$\langle V \rangle = \langle VS1, VS2, \ldots, VSi, \ldots, VS(m-1) \rangle , \qquad (6.24)$$

then the acceleration <A> may be given as:

$$\langle A \rangle = \langle AS1, AS2, \ldots, ASi, \ldots, AS(m-2) \rangle \qquad (6.25)$$

$$ASi = [\ VSi - VS(i-1)\ ] \ / \ TSi \qquad (6.26)$$

and AS1 = VS1, considering the cell started from a stationary position. Table (6.1) gives an example of the results of this computation.

Two qualifiers are used to describe the acceleration at each motion step. The first is to describe it as positive or negative.

RULE(6.4):

IF ASi .LT. 0 ==then==> Q(ASi) <--- NEGATIVE ACC.

IF ASi .GT. 0 ==then==> Q(ASi) <--- POSITIVE ACC.

The second qualifier is used to describe the acceleration as VERY SLOW, SLOW, AVERAGE, FAST, and VERY FAST. Description 6.1 gives an example.

In this section, we discussed methods for the global analysis of the basic cell locomotion properties (displacement, velocity, and acceleration). Table (6.1) summarizes the numerical description of cell motion properties, and their symbolical qualification is given in Description (6.1). In the following section, we will discuss and analyze one of the most important properties of cell locomotion, namely chemotaxis behaviour.

## 6.4 CHEMOTAXIS ANALYSIS

Chemotaxis is the response of a motile cell to the directional influence of an external factor (bacteria, tumour, chemical substances). The importance of chemotaxis lies in the fact that it results in the efficient localization of invading agents. The role which chemotaxis plays in eliminating the tumour cells was a subject of study by [Levine et. al, 81] (see also Section 2.5.5). In their study, the locomotion of cells is characterized as positive chemotaxis, negative chemotaxis, or random motion. Locomotion analysis in the current research is different in the sense that here we attempt to understand and characterize the dynamic behaviour of a single moving cell. This section is concerned with the quantification and description of the relationship between the cell locomotion

and the influence of an external factor (chemotaxis analysis). The objective of this analysis is accomplished in three steps:

(a) Compute the global directional movement of the cell.

(b) Quantify the cell response to the influence of an external factor, thereby quantifying the effectiveness of the influence.

(c) Characterize and describe the global behaviour of the cell locomotion.

A typical example of the quantification of the global cell locomotion is given in Description (6.1). The processes and algorithms which are employed to generate these descriptions are described in the remainder of this section.

## 6.4.1 Computing The Directional Movement Of The Cell

The objective of this analysis is to determine whether the cell motion has a tendency towards a specific direction or if it is random. The input for this analysis consists of the incremental displacement of the cell between sequential frames. The output includes quantification of the movement of the cell in the main directions of the plane.

Let us define the incremental displacement and direction of motion between two sequential frames (i, i+1) as Li and Di, where:

$$Li = [\ dXi^2 + dYi^2\ ]^{1/2} \qquad (6.27)$$

$$Di = Tan^{-1}\ (\ dY\ /\ dX\ ) \qquad (6.28)$$

and $dXi = (X(i+1) - Xi)$ , $dYi = (Y(i+1) - Yi)$

We divide the plane of cell motion into nd equal directions S1, S2, . . . , Sj, . . . , Snd. The extent of each direction is equal to 360/nd. The incremental displacement is considered to be in direction Si if the displacement has an angle Di with the X axis that is between S(j-1) and Sj. The vector that represents the total locomotion in a specific direction (TDR) can be computed as the resultant of all the cell displacements in that direction. Figures (6.8a) and (6.8b) show the total displacement of the cell, where the space is divided into 4 and 8 equal directions respectively. The vector (RR) which represents the global locomotion of the cell is the resultant of all the vectors defining the cell locomotion in the different directions. Thus:

$$RR = \sum_{i=1}^{nd} RDi \; . \qquad (6.29)$$

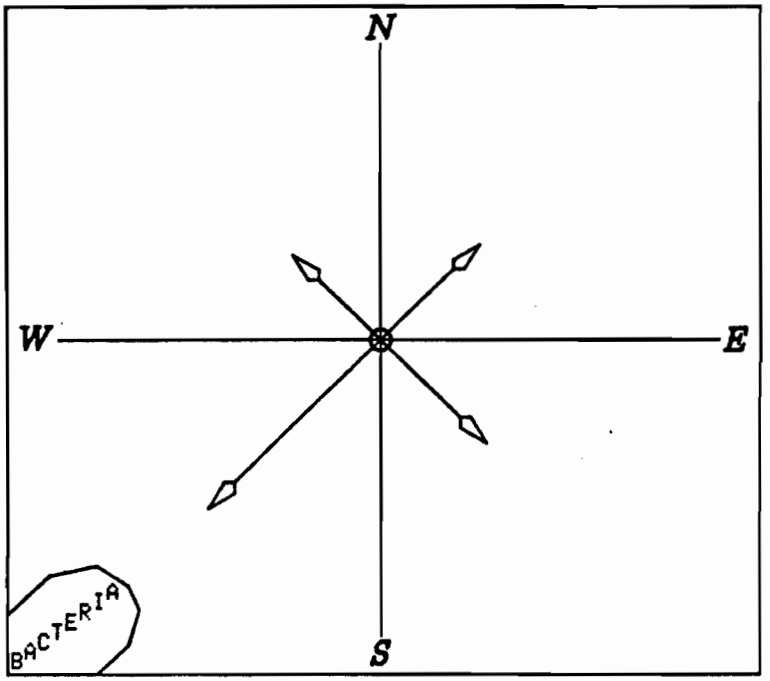
where "sum" in this equation indicates a vector summation.

As an alternative, if the displacements of the cell in the different directions is not required, the vector RR can be computed directly from the X and Y coordinates of the cell location in each frame of the sequence as follows:

$$TDX = \sum_{i=1}^{n-1} [X(i+1)-X(i)], \quad \text{and} \quad TDY = \sum_{i=1}^{n-1} [Y(i+1)-Y(i)]$$

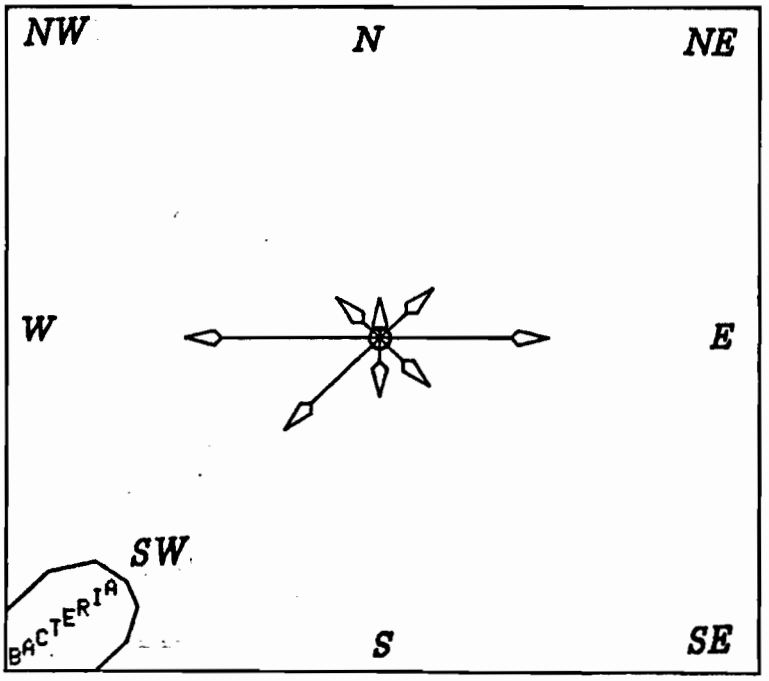
$$LRR = [ ( TDX )^2 + ( TDY )^2 ]^{1/2} \qquad (6.30)$$

**Frame number: First=1, Last=250 4 state analysis**




*Figure(6.8a) Vector sum of locomotion in each of four directions.*

**Frame number: First=1, Last=250 8 state analysis**




*Figure(6.8b) Vector sum of locomotion in each of eight directions.*

$$DRR = Tan^{-1} ( TDY / TDX ) , \quad (6.31)$$

where Xi, Yi are the X and Y coordinates of the cell at frame(i), and n is the total number of frames. Figures (6.9a) and (6.9b) show the locomotion of the cell in the different directions, as well as the global locomotion vector (marked with two arrows).

The directional tendency of the cell is the amount of movement that the cell exhibits in a specific direction. In the case of random locomotion, the tendencies of the cell in the different directions are approximately equal. Consequently the resultant vector RR of the cell locomotion is approximatly zero. In the case of chemotactic locomotion, the cell exhibits a tendency to move in a specific direction.

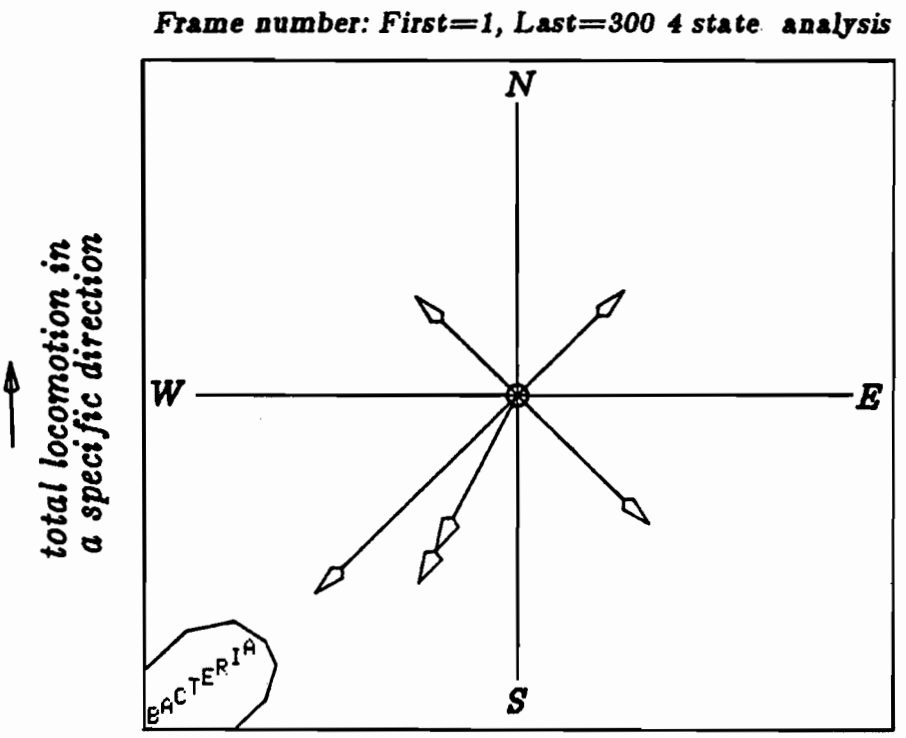
The movement in the different directions, as well as the global locomotion, can be described as NONE, VERY SHORT, SHORT, AVERAGE, LONG, and VERY LONG. This qualification is based on a comparison with typical random motion. In this case, the cell locomotion in each direction is almost equal. This can be computed as:

$$TDRav = TTD / nd \quad . \quad (6.32)$$

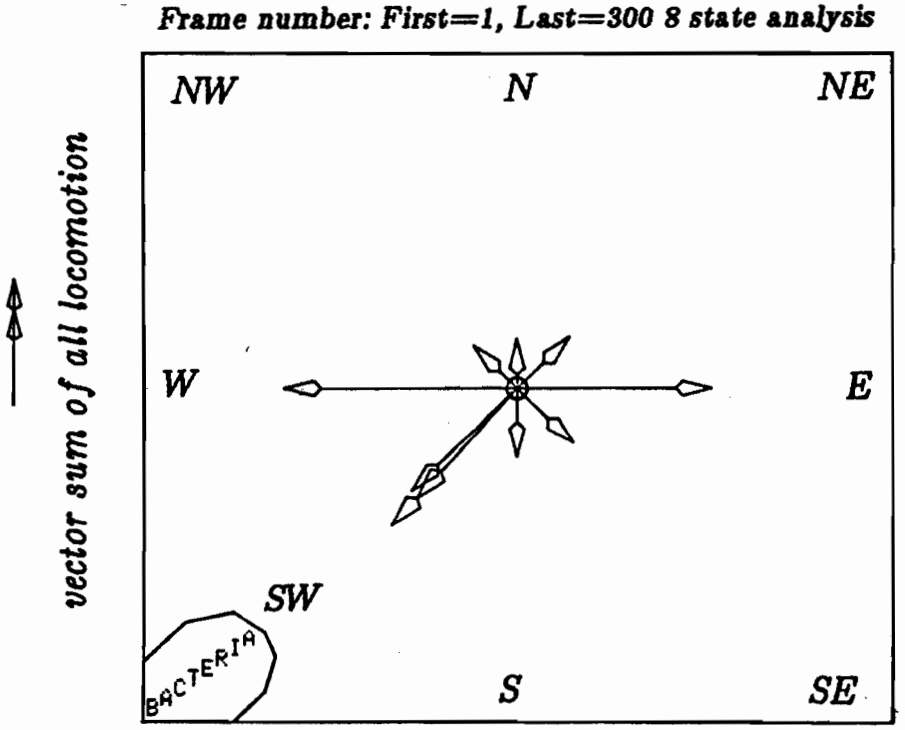
The locomotion in a specific direction can be normalized as:

$$TDRNk = TDRk / TDRav \quad (6.33)$$

Then using the representational rules, the normalized values of the directional movement of the cell can be assigned to the appropriate symbolic qualifiers. For example, we may




*Figure(6.9a) Vector sum of all locomotion.*




*Figure(6.9b) Vector sum of all locomotion.*

*Figure(6.9a-6.9b) Global cell locomotion.*
*(a) 4 states analysis, (b) 8 states analysis.*

describe the cell motion as being AVERAGE if the resultant vector is about equal to TDR.

## 6.4.2 Chemotaxis Quantification

The chemotactic motion of the cell can be measured as the percentage movement of the cell in the direction of influence to the total movements in the space domain. Thus, if the total locomotion of the cell is represented by the vector RR = (LRR, DRR), where LRR and DRR are the length and direction of the vector RR, then the percentage chemotactic motion PCM can be computed as:

$$PCM = LRR / TLD \cdot 100 \ \% \ , \quad (6.34)$$

where TTD is total locomotion distance in the space domain (which is computed above). Thus, using the value of PCM, the cell motion can be characterized as random or chemotactic by the following rule:

RULE(6.5):

```
IF PCM                .LE.  E1 ==then==>  RANDOM
IF PCM .GT.  E1  AND .LE.  E2 ==then==>  ALMOST RANDOM
IF PCM .GT.  E2  AND .LE.  E3 ==then==>  PARTIALLY RANDOM
IF PCM .GT.  E3  AND .LE.  E4 ==then==>  ALMOST CHEMOTACTIC
IF PCM .GT.  E4               ==then==>  CHEMOTACTIC
```

Thus, chemotaxis can be described as positive or negative depending on whether the cell moves towards or against the direction of influence. To compute and describe this behaviour, we consider the cell as having started its motion from point 0 (Xo, Yo), the origin of the two

dimensional plane, with the external factor concentrated at a point E (Xe, Ye). The line OE which connects the original position of the cell (O) to the center of the external factor (E) represents the direction of the influence, as shown in Figure (6.10). In this figure, the line SOS, which passes through the point O and is normal to the line OE, represents the border line which divides the plane into two regions. One is PCR or "positive chemotaxis region" (the region where the external factor is located), and the other is NCR or "negative chemotaxis region". Thus, if the vector that represents the global locomotion is in the region PCR, this indicates positive chemotaxis. If it is in region NCR, negative chemotaxis is implied. This can be quantified as follows:

RULE(6.6):

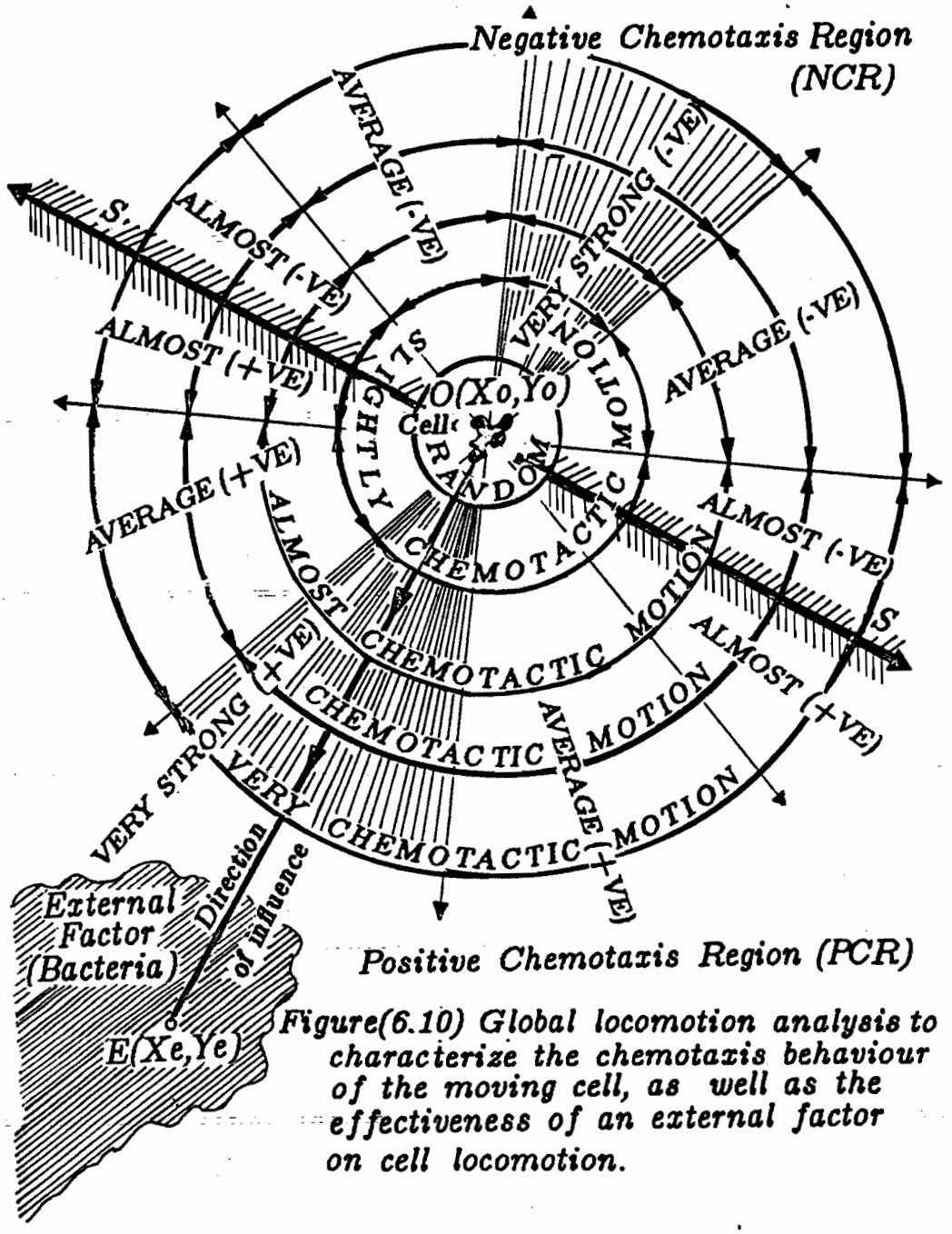
IF DRR .GE. E1 AND .LT. E2 ==then==> NEGATIVE CHEMOTAXIS

IF DRR .GE. E2 AND .LT. E1 ==then==> POSITIVE CHEMOTAXIS

where DRR is the angle of the global locomotion with the X axis (0-360 degrees), E1 and E2 are the angles of the border line between the positive and negative chemotaxis regions.

## 6.4.3 Quantifying The Effectiveness Of Influence

The effectiveness of an external factor (EF) on the locomotion of a moving cell can be defined as its ability to attract the cell in the direction of its influence. This force is a function of two factors: (a) the percentage of the chemotactic movements (PCM), and (b) the angle between




*Figure(6.10) Global locomotion analysis to characterize the chemotaxis behaviour of the moving cell, as well as the effectiveness of an external factor on cell locomotion.*

the direction of motion and influence (DRI). Thus,

$$EF = f(PCM, DRI) . \quad (6.35)$$

The value PCM is described and estimated in the preceding section. The angle DRI can be computed as:

$$DRI = |DR - DI| , \quad (6.36)$$

where DR and DI are the directions of the global locomotion and the influence, respectively. The value DRI can be utilized as an indicator to tell whether or not the cell will ultimately reach the area where the external factor is located. To compute this, the value of DRI can be normalized between (0-1) as:

$$DRIn = ( 90 - DRI ) / 90 . \quad (6.37)$$

Thus, DRIn = -1, if the global locomotion is opposite to the direction of influence (purely negative chemotaxis), and DRIn = +1, if the locomotion is exactly in the direction of influence (purely positive chemotaxis). Using representational rules, the value DRIn can be used to describe the direction of global locomotion compared to the direction of influence as QUITE OPPOSITE, ALMOST OPPOSITE, PERPENDICULAR, ALMOST THE SAME, THE SAME.

Finally, the effectiveness of an influence can be computed as the geometric mean (GM) of PCM and DRIn:

$$EF = ( PCM . DRIn )^{1/2} . 100 \% \quad (6.38)$$

Note that EF lies between plus and minus 100%.

Thus, the value EF is a summary of the chemotactic movement and the effectiveness of the influence. The sign indicates positive or negative chemotaxis, and the amplitude represents the strength. This can be utilized by representatinal rules to describe the global behaviour of the cell locomotion as follows:

RULE(6.7):

IF EF .LT. 0 ==then==> NEGATIVE CHEMOTAXIS

IF EF .EQ. 0 ==then==> RANDOM MOTION

IF EF .GT. 0 ==then==> POSITIVE CHEMOTAXIS

IF |EF| .LE. E1 ==then==> SLIGHT

IF |EF| .GT. E1 AND .LE. E2 ==then==> ALMOST

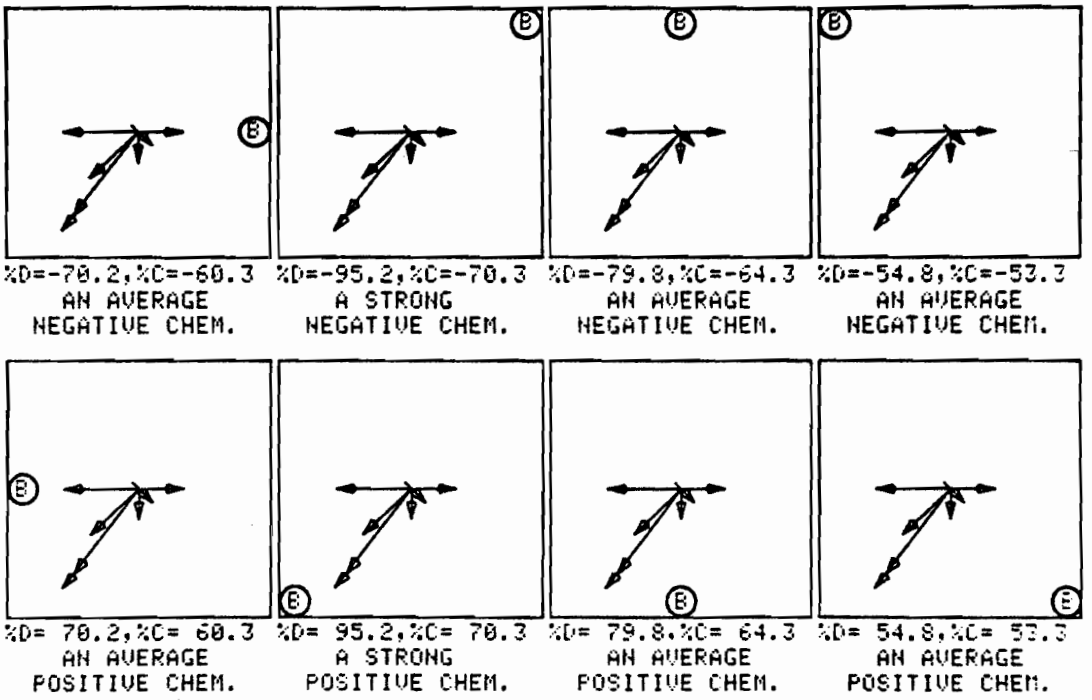
IF |EF| .GT. E2 AND .LE. E3 ==then==> AVERAGE

IF |EF| .GT. E3 AND .LE. E4 ==then==> STRONG

IF |EF| .GT. E4 ==then==> VERY STRONG

Description (6.1) is a typical example of the global locomotion characterization and description generated by the system. Figure (6.11) shows different global locomotion characterizations of the same cell under the influence of bacteria placed in different positions.

The analysis of the chemotaxis property of cell locomotion is presented in this section. This property plays an important role in describing the cell under the influence of an external factor. In this analysis, we first computed the movement of the cell in the different directions, as well as the vector which represents the global locomotion. Secondly, we quantified the response of




*Figure(6.11) Global quantification and characterization of moving cell locomotion.*

*The symbol B represents the location of the external influence.*

***%D refers to the symbol DRI, and %C to PCM (see equation (6.28)).***

the cell to the effect of an external factor. Finally, we estimated the effectiveness of the influence of the external factor on the cell locomotion. In this way, the global behaviour of the cell locomotion is described.

## 6.5 SUMMARY

Motion analysis and description of a non-rigid moving object can be achieved in two steps. The first is concerned with motion detection and tracking of an object in the image sequence. The second step concerns the analysis of data extracted from the first step in order to generate a summary of the locomotion behaviour. In the present system, the processes which are associated with the first step are responsible for locating the cell in each image of the sequence, and detecting the incremental change in its location between sequential frames. These processes are described in chapters 4 and 5. In this chapter, we have described the processes which are associated with the second step. That is, global motion analysis and description. The objectives of these processes are to analyze the multitude of static and incremental data in order to characterize and describe the locomotion behaviour of the cell. To accomplish this, first, the global movement of the cell is detected from the irrelevant and noisy ones. From this data, the path of motion is constructed in a sequence of steps. Each motion step is described by a set of motion properties, such as: time, distance, direction of motion,

velocity, and acceleration.

Chemotaxis is one of the most important descriptors of cell locomotion. It is the response of a moving cell to the influence of an external factor. In order to quantify and describe this behaviour, first, the total displacement of the cell in each of the main directions of the spatial domain is quantified and described in comparison to a similar random motion. The latter implies no external factor, so that the cell should exhibit equal locomotion in the different directions. Then, the vector that represents the global locomotion of the cell is also quantified and described as random or chemotactic. Finally, the chemotaxis is quantified as a function of two parameters: the percentage of the chemotactic motion, and the angle of its direction with the direction of the influence. Thereby, the chemotaxis behaviour of the cell is described as POSITIVE or NEGATIVE. Also, the effectiveness of an influence is described. Description (6.1) presents a summary generated by the system for the global locomotion behaviour of a cell which was tracked for 450 frames (225 seconds).

# DESCRIPTION (6.1)

# GLOBAL LOCOMOTION ANALYSIS AND DESCRIPTION

## INTRODUCTION

The following is a global characterization and discription of the locomotion analysis of a NEUTROPHILE cell. The cell motion was recorded in real time on 16mm cine film at rate of TWO frames per second. The cell was under the influence of BACTERIA which is located in the SOUTH-WESTERLY direction of the original location of the cell. The total observation time was 225 seconds (450 frames). The following is a description of the cell locomotion between frame number 200 and 450 (125.0 seconds)

## CELL PATH and MOTION ANALYSIS

The space domain of the cell motion is divided into EIGHT equal directions(states). First, the cell path was obtained by sampling the displacement between frames in increments of approximatly 2.0 microns. Then sequences with the same incremental direction were merged into one to produce the final cell path which consists of 34 steps. The description of the time, distance, speed, direction, and acceleration of the cell at each step is as follows :

| TIME | DISTANCE | SPEED | DIRECTION | ACCELERATION | |
|---|---|---|---|---|---|
| VERY SHORT | VERY SHORT | VERY SLOW | NORTH-EASTERLY | NONE | |
| VERY SHORT | SHORT | VERY FAST | EASTERLY | VERY FAST | POSITIVE |
| VERY LONG | VERY LONG | AVERAGE | WESTERLY | AVERAGE | NEGATIVE |
| SHORT | VERY SHORT | VERY SLOW | NORTH-EASTERLY | VERY FAST | NEGATIVE |
| SHORT | MEDIUM | VERY FAST | SOUTH-WESTERLY | VERY FAST | POSITIVE |
| SHORT | VERY SHORT | VERY SLOW | NORTH-EASTERLY | VERY FAST | NEGATIVE |
| MEDIUM | SHORT | AVERAGE | SOUTH-WESTERLY | VERY FAST | POSITIVE |
| VERY LONG | MEDIUM | SLOW | SOUTH | NONE | |
| LONG | MEDIUM | AVERAGE | SOUTH-WESTERLY | SLOW | POSITIVE |
| SHORT | MEDIUM | VERY FAST | EASTERLY | VERY FAST | POSITIVE |
| MEDIUM | LONG | FAST | SOUTH-WESTERLY | VERY FAST | NEGATIVE |
| SHORT | MEDIUM | VERY FAST | EASTERLY | VERY FAST | POSITIVE |
| SHORT | MEDIUM | VERY FAST | NORTH-WESTERLY | VERY FAST | NEGATIVE |
| MEDIUM | VERY LONG | VERY FAST | SOUTH | AVERAGE | NEGATIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-WESTERLY | AVERAGE | NEGATIVE |
| LONG | MEDIUM | SLOW | SOUTH | AVERAGE | NEGATIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-EASTERLY | AVERAGE | POSITIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-WESTERLY | SLOW | POSITIVE |
| SHORT | MEDIUM | VERY FAST | SOUTH-EASTERLY | VERY FAST | POSITIVE |
| MEDIUM | MEDIUM | FAST | SOUTH-WESTERLY | VERY FAST | NEGATIVE |

| | | | | | |
|---|---|---|---|---|---|
| LONG | MEDIUM | SLOW | SOUTH | AVERAGE | NEGATIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-WESTERLY | AVERAGE | POSITIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH | NONE | |
| SHORT | MEDIUM | FAST | SOUTH-WESTERLY | VERY FAST | POSITIVE |
| VERY LONG | VERY LONG | AVERAGE | SOUTH | AVERAGE | NEGATIVE |
| MEDIUM | LONG | VERY FAST | WESTERLY | VERY FAST | POSITIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-EASTERLY | FAST | NEGATIVE |
| VERY LONG | MEDIUM | SLOW | SOUTH | AVERAGE | NEGATIVE |
| LONG | MEDIUM | AVERAGE | SOUTH-EASTERLY | SLOW | POSITIVE |
| VERY SHORT | VERY SHORT | VERY SLOW | NORTH-EASTERLY | VERY FAST | NEGATIVE |
| SHORT | MEDIUM | FAST | EASTERLY | VERY FAST | POSITIVE |
| MEDIUM | VERY SHORT | VERY SLOW | NORTH-EASTERLY | VERY FAST | NEGATIVE |
| SHORT | SHORT | AVERAGE | EASTERLY | VERY FAST | POSITIVE |
| MEDIUM | MEDIUM | AVERAGE | SOUTH-EASTERLY | AVERAGE | NEGATIVE |

## CHEMOTAXIS ANALYSIS :

Chemotaxis is the response of a motile cell to the directional influence of a chemical substance or any external factor (BACTERIA in this film). The following is a summary of the directional movements of the cell under analysis when compared to typical random motion of a similar cell:

| DIRECTION | TOTAL DISPLACEMENT |
|---|---|
| EASTERLY | AVERAGE |
| NORTH-EASTERLY | NONE |
| NORTH | NONE |
| NORTH-WESTERLY | SHORT |
| WESTERLY | AVERAGE |
| SOUTH-WESTERLY | VERY LONG |
| SOUTH | VERY LONG |
| SOUTH-EASTERLY | AVERAGE |

## CONCLUSION :

The resultant directional locomotion is in a SOUTH direction, which is ALMOST THE SAME direction in which the BACTERIA is located. This motion represents THREE-FIFTHS of the total displacement of the cell.

From the above analysis we may conclude that :

## THE CELL HAS AN AVERAGE POSITIVE CHEMOTAXIS MOTION

# TABLE <6. 1>

## GLOBAL LOCOMOTION ANALYSIS AND DESCRIPTION

### GLOBAL LOCOMOTION DATA

| | | | |
|---|---|---|---|
| FIRST FRAME NUMBER | = | 1 | |
| LAST FRAME NUMBER | = | 450 | |
| TOTAL TIME | = | 224.5 | Seconds |
| ANGLE TO BACTERIA LOCATION | = | 45.00 | Degrees |
| INCREMENTAL STEP | = | 5.0 | Microns |
| NUMBER OF STEPS | = | 22 | |
| TOTAL DISTANCE | = | 93.50 | Microns |
| AVERAGE DISPLACEMENT /STEP | = | 4.25 | Microns |
| AVERAGE TIME /STEP | = | 10.20 | Seconds |
| AVERAGE VELOCITY | = | 0.42 | Microns/Seconds |
| AVERAGE ACCELERATION | = | 0.00 | Microns/Seconds squared |
| TOTAL DISPLACEMENT | = | 63.80 | |
| AVERAGE ANGLE OF DIRECTION | = | 247.00 | |
| RESULTANT DIRECTION | = | SOUTH-WESTERLY | |

### CELL PATH AND MOTION ANALYSIS :

| STEP No. | INITIAL FRAME No. | FINAL FRAME No. | TIME (sec) | DISTANCE (microns) | DIRECTION 8 STATES | VELOCITY (mic/sec) | ACCELERATION (mic/sec. sec) |
|---|---|---|---|---|---|---|---|
| 1 | 1 | 24 | 11.5 | 5.95 | 5 | 0.52 | 0.04 |
| 2 | 24 | 76 | 26.0 | 15.76 | 6 | 0.61 | 0.00 |
| 3 | 76 | 134 | 29.0 | 10.65 | 8 | 0.37 | -0.01 |
| 4 | 134 | 169 | 17.5 | 6.17 | 7 | 0.35 | 0.00 |
| 5 | 169 | 187 | 9.0 | 5.86 | 5 | 0.65 | 0.03 |
| 6 | 187 | 201 | 7.0 | 0.00 | 2 | 0.00 | -0.09 |
| 7 | 201 | 202 | 0.5 | 0.91 | 1 | 1.82 | 3.64 |
| 8 | 202 | 224 | 11.0 | 5.20 | 5 | 0.47 | -0.12 |
| 9 | 224 | 236 | 6.00 | 0.00 | 2 | 0.00 | -0.08 |
| 10 | 236 | 238 | 1.00 | 1.37 | 6 | 1.37 | 1.37 |
| 11 | 238 | 240 | 1.00 | 0.00 | 2 | 0.00 | -1.37 |
| 12 | 240 | 245 | 2.50 | 0.91 | 6 | 0.36 | 0.14 |
| 13 | 245 | 278 | 16.50 | 5.00 | 7 | 0.30 | 0.00 |
| 14 | 278 | 296 | 9.00 | 5.01 | 6 | 0.56 | 0.03 |
| 15 | 296 | 350 | 27.00 | 10.52 | 7 | 0.39 | -0.01 |
| 16 | 350 | 393 | 21.50 | 11.08 | 6 | 0.52 | 0.01 |
| 17 | 393 | 419 | 13.00 | 5.26 | 8 | 0.40 | -0.01 |
| 18 | 419 | 431 | 6.00 | 0.00 | 2 | 0.00 | -0.07 |
| 19 | 431 | 434 | 1.50 | 1.32 | 1 | 0.88 | 0.59 |
| 20 | 434 | 441 | 3.50 | 0.00 | 2 | 0.00 | -0.25 |
| 21 | 441 | 443 | 1.00 | 0.79 | 1 | 0.79 | 0.79 |
| 22 | 443 | 450 | 3.50 | 1.73 | 8 | 0.50 | -0.09 |

# CHEMOTAXIS ANALYSIS :

| DIRECTION | DISPLACEMENT |
|---|---|
| 1-EASTERLY | 3.03 ==> |
| 2-NORTH-EARTERLY | 0.00 > |
| 3-NORTH | 0.00 > |
| 4-NORHT-WESTERLY | 0.00 > |
| 5-WESTERLY | 17.01 ==========> |
| 6-SOUTH-WESTERLY | 34.13 ========================> |
| 7-SOUTH | 21.69 ==============> |
| 8-SOUTH-EASTERLY | 17.64 ===========> |

## RESULTANT LOCOMOTION :

SOUTH-WESTERLY 63.80 ==========================================>

(a) PERCENTAGE OF TOTAL LOCOMOTION IN DIRECTION OF INFLUENCE ---------- 90.56 %

(b) RATIO OF MAGNITUDE OF CHEMOTACTIC MOTION IN DIRECTION OF INFLUENCE TO TOTAL CELL LOCOMOTION ---------- 68.24 %

(C) EFFECTIVENESS OF THE EXTERNAL FACTOR ---------- 78.61 %

# CHAPTER 7

# GLOBAL SHAPE ANALYSIS

## 7.1 INTRODUCTION

The perception of shape plays a prominent role in both human and computer vision. It is a common problem in any computer vision, scene analysis, or pattern recognition system. The solution to this problem may be achieved through two stages of processing: shape analysis, and shape description. In shape analysis, a digitized image of an object is transformed into a scalar vector whose elements are measurements of some of the shape properties. The second task of shape analysis is to transform the image of an object into a graph. The properties of this graph express the shape and structural properties of the object. Shape description represents the higher level process of shape perception by computer. In this process the scalar vector or graph, the resulting form of the shape analysis, is analyzed using a syntactic analysis methodology in order to generate a summary in a natural language (a symbolic description). It contains all the relevant information pertaining to the shape of the object.

Figure (2.1) shows the basic steps for shape analysis and description, as well as the input and output data at each step. A brief review of current shape analysis and description techniques was presented in Section 2.3, and attention was directed to the most recent surveys in this area [Meagher, 79; Pavlidis, 81]. The shape analysis techniques which have been used or are 00directly related to our current work are also reviewed in the same section. Most of the work which has been done in dynamic scene analysis is restricted to the study of the motion of rigid objects. Some of this work has considered change in shape due to change in the viewing conditions. For example, a car moving in front of a fixed camera [Nagel, 78b], fish swimming in a vat [Yachida et. al, 78], and motion of the left vertricular [Tsotsos, 80]. However, in each of these cases the change is restricted by many constraints and is predictable.

Our study differs from previous work in that we study the motion of non-rigid moving objects. The changes in shape are due to changes in the physical properties, which are non predictable. Analysis of the random changes in the shape of moving objects is very important in studying the characteristic behaviour of biological objects. For example, it has become increasingly evident that the cell membrane plays a pivotal role in the life, development, and regulation of cells. There is no existing method to quantify the observable changes in membrane shape that occur

in locomotion, an important component of cell behaviour.

The general problem of shape description is very difficult. Recently, there has been interest in syntactic pattern recognition techniques which analyze patterns by a hierarchial decomposition parsing process. The advantages of such an approach suggest that it might be appropriate to study hierarchial shape representation in more detail as a vehicle for cell shape description, as well as for the global structural and membrane shape changes which occur during locomotion.

The problem of shape analysis and description of an arbitrary shape in a static scene was discussed in Chapter 4. The techniques used to solve this problem include: (a) segmentation and boundary tracking, (b) curvilinear and polygonal approximation, (c) polygonal decomposition and labeled graph representation, and (d) rule-based syntactic analysis and shape description techniques (to quantify and describe the shape properties symbolically). In addition to the general difficulties of describing an arbitrary shape in a static image, we face the following problems:

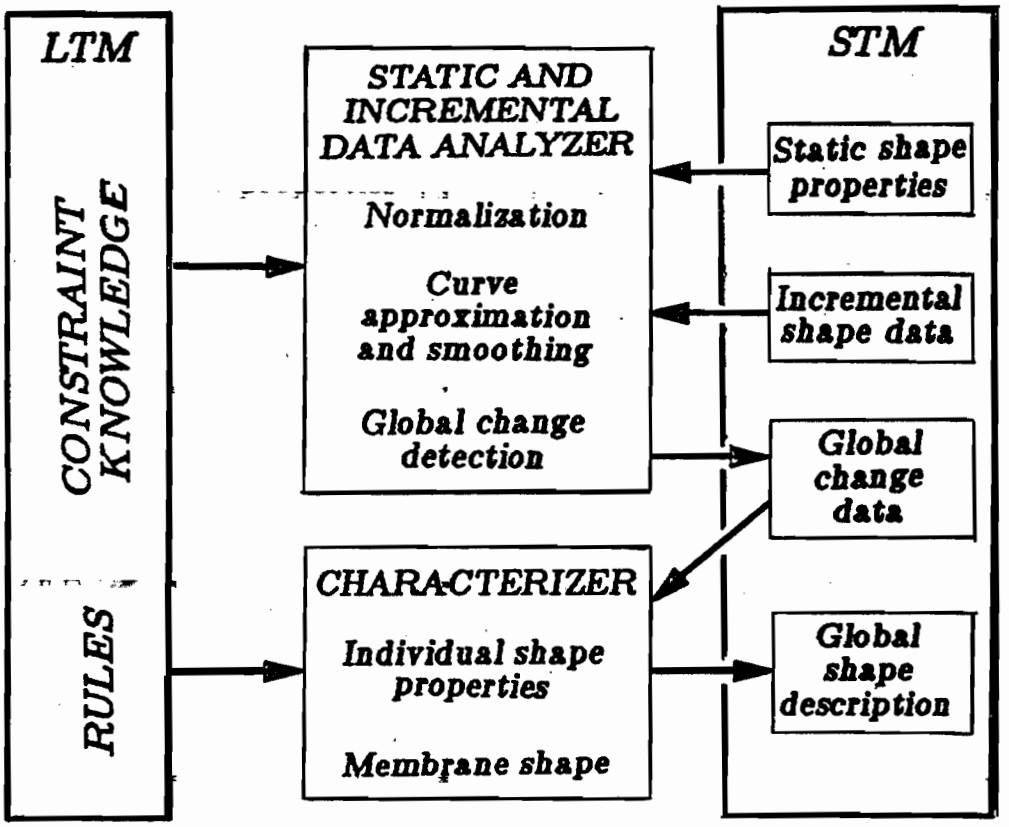
(a) Estimating the incremental change between two different images, of the shape and structure of a non-rigid moving object.

(b) Detecting and characterizing the global shape changes in the morphology of a non-rigid moving object over a period of time from a sequence of pictures.

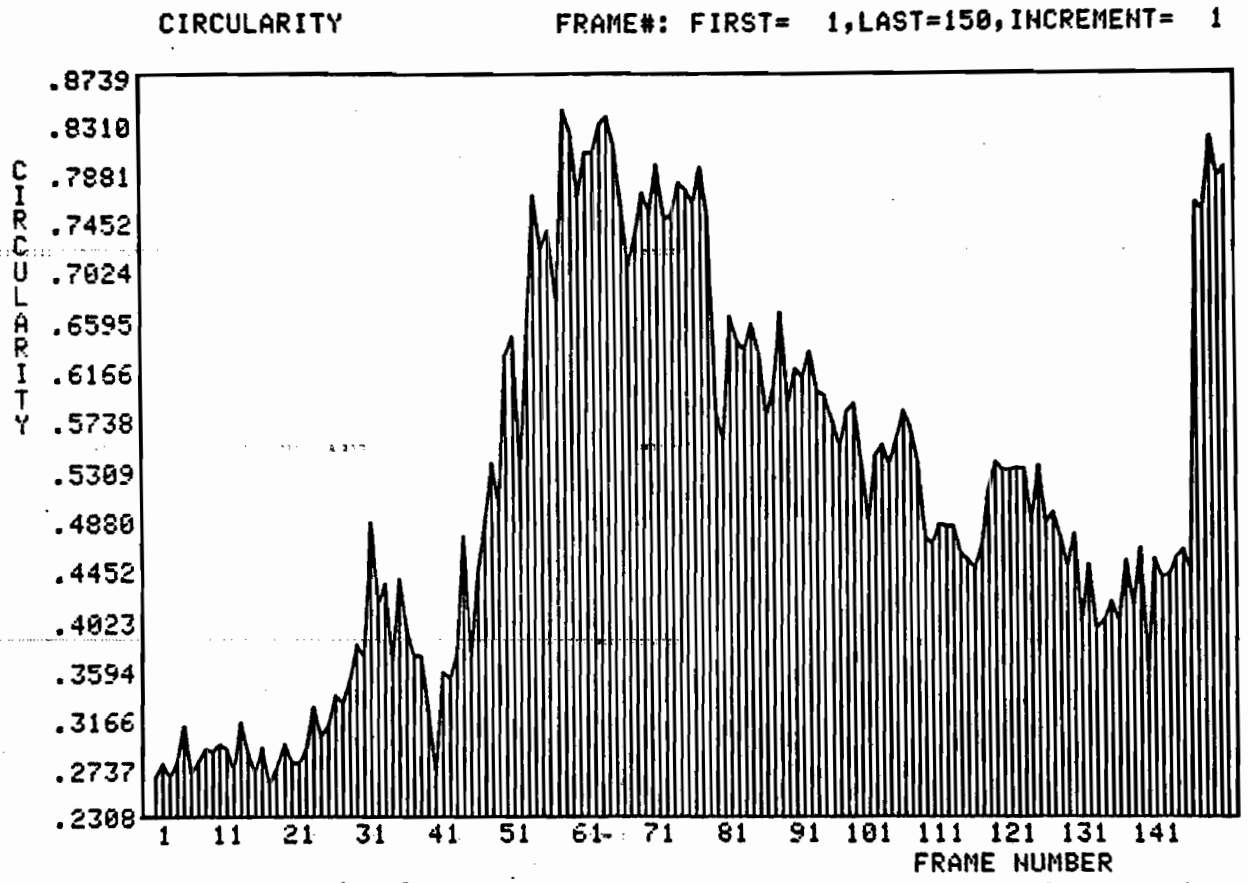
(c) Presenting all the above descriptions in a meaningful form to the user.

The problem of the incremental change detection and description was discussed in Chapter 5. The problem of detecting and characterizing the global changes in the shape will comprise the remainder of this chapter. Figure (7.1) shows the main processes and data structures used in global shape analysis and description. The input to this stage of the system consists of the static and incremental description of the shape properties in and between each frame. An example of the input data is shown in Figure (7.2). It consists of the static values of one of the shape properties (circularity) in 150 frames in sequence. The output consists of a summary describing the global changes of the cell shape and their characterization. A typical example of this characterization is given in Description (7.1). The methodologies and processes which are used to generate these descriptions are discussed in this chapter in the following order.

The basic methodologies and techniques for detecting the global changes from the static and incremental data are described in Section 7.2. Description of the global changes in each of the main shape properties will be given in




*Figure(7.1) Processes and data structure for global shape analysis.*




***Figure(7.2) The static description of one of the shape properties (circularity) used as input data for global shape analysis.***

Section 7.3. In Section 7.4, we will show experimentally that an individual shape property is not sufficient to describe an arbitrary shape. Also in the same section, we discuss the development of a mathematical expression for the membrane shape measure. In this way, the global changes in the cell membrane shape can be characterized and described. Finally, Section 7.5 is a summary of the chapter.

## 7.2 METHODOLOGY FOR DETECTING AND DESCRIBING GLOBAL SHAPE CHANGES

The technique for detecting and describing the shape changes in this thesis consists of three basic steps:
(a) Global change detection: to detect relevant changes in shape from the static and incremental data.
(b) Symbolical qualification: to transform the global data into symbolic qualifiers.
(c) Characterization and description: to analyze the symbolic qualification in order to summarize the characteristic behaviour of the changes in the cell shape.

The objective of the global change detection is to reduce the static and incremental data to those representing information relevant to the global changes. This can be achieved by detecting the points (frames) where a significant change has occured, called "key frames". There are two definitions for point of significant change. The first definition is a point where the dynamic behaviour changes. For example, from stationary (dP/dt = 0) to

increasing (dP/dt > 0) or decreasing (dP/dt < 0). The second refers to a time at which a significant change has occured in the qualification (description) level. This can be defined as:

$$Qi(Pk) \longrightarrow Qj(Pk),$$

where Qi(P) and Qj(P) are two different levels of qualification for the property (Pk). For example, from SIMPLE to COMPLEX, or from SMOOTH to JAGGED.

If the change in the qualification level is the only required description for a specific application, this can be accomplished by applying the representational rules directly to the static and incremental symbolic qualifications. However, in some applications, description of the changes in the dynamic behaviour (for example from increasing to decreasing or stationary) is also required.

The technique developed for this system is designed to detect and provide descriptions for both types of global changes. This will be described in the following two sections. First, in Section 7.2.1, detection of the changes in the dynamic behaviour will discussed. Second, Section 7.2.2 describes a methodology for detecting and summarizing the significant changes in the qualification level.

## 7.2.1 Dynamic Changes And Key Frames

The objective of this analysis is to detect the points where the dynamic behaviour of a specific property has changed. The frames where these changes have occured are referred to as key frames. To accomplish this, the static and incremental data is first normalized between 0-1, and represented by a curve, as shown in Figure (7.2). The amplitude of the curve at any point (frame) represents the static value of the property at that frame. The variations between the neighbouring points in the curve represent the incremental changes of the property between the sequential frames. Curve smoothing and approximation can be used to detect the points (key frames) in the curve where significant changes occur. This data can be used to characterize and describe the changes in the property under consideration. A detailed description of this computation is given below.

### NORMALIZATION

In the global shape analysis processes, normalization is employed as a preprocessing step for all the static and incremental data for the following reasons :

(a) The different properties have been measured in different units.

(b) The values of the input data have different scales (varing over several order of magnitude).

(c) The amplitude of the change for different properties varies widely.

In order to detect the global changes in a specific property and to be able to perform any mathematical operation between the different properties, we should present their data in a uniform pattern. The theoretical aspects and different methods of normalization are discussed in Section 4.6.4. In this operation, the values of a specific property in a sequence of frames P1, P2, ....., Pn are normalized to range between zero and one to give p1, p2, ....., pn, as follows:

$$pi = ( Pi - Pmin ) / ( Pmax - Pmin ), \qquad (7.1)$$

where pi is the normalized value of Pi, and

$$Pmin = Min (P1, P2, ..., Pn) \qquad (7.2)$$

$$Pmax = Max (P1, P2, ..., Pn). \qquad (7.3)$$

Normalizing the static and incremental data will assist the analysis of the following steps:

(1) Detecting the global changes, especially when the change is very small from frame to frame or almost negligible relative to the original value of the property.

(2) Describing the property value or the change in it, by a limited number of symbolic qualifiers.

(3) Comparing the amount of change between different properties.

(4) Forming higher level descriptors as a function of more than one property.

## CURVE APPROXIMATION AND SMOOTHING

Curve approximation or fitting is a popular technique in many branches of engineering and computer science for describing large volumes of data in a concise way. The objective is to extract the relevant information (points of significant change) from a set of irrelevant and noisy data. A mathematical background and different techniques for curve fitting can be found in [Pavlidis, 77]. From a theoretical point of view, techniques which are used for polygonal approximation can also be used for curve approximation. The difference between the two methods is that; in most cases, the data for polygonal approximation is given in two dimensions (f(x,y)), whereas for curve approximation the data is given in one dimension (f(y)). In Section 4.3 we described an algorithm for polygonal approximation based on a splitting technique developed by [Ramer, 72]. Using the same technique, we will describe below an iterative splitting algorithm for curve approximation.

Figure (7.2) shows an example of the input data to the curve approximation algorithm. The procedure of the algorithm consists of the following steps:

(1) Connect the first and the last point on the curve by a straight line AB.

(2) Find the point C on the curve at the maximum distance Lm

from the line AB. If Lm is greater than the approximation threshold E, then split the curve at point C.

(3) For each new segment, repeat steps (1) and (2).

Figure (7.3) shows the curve approximation of the data in Figure (7.2).

The output of the curve approximation consists of a set of vertices {KF} representing the key frames:

$$\{KF\} = \{KF1, KF2, \ldots, KF_j, \ldots KFm\}, \qquad (7.4)$$

where m the number of key frames. Each key frame is defined by time (frame number) and the property value at that time, as follows:

$$KF_j(Pi) = [t_j, Pi(t_j)]. \qquad (7.5)$$

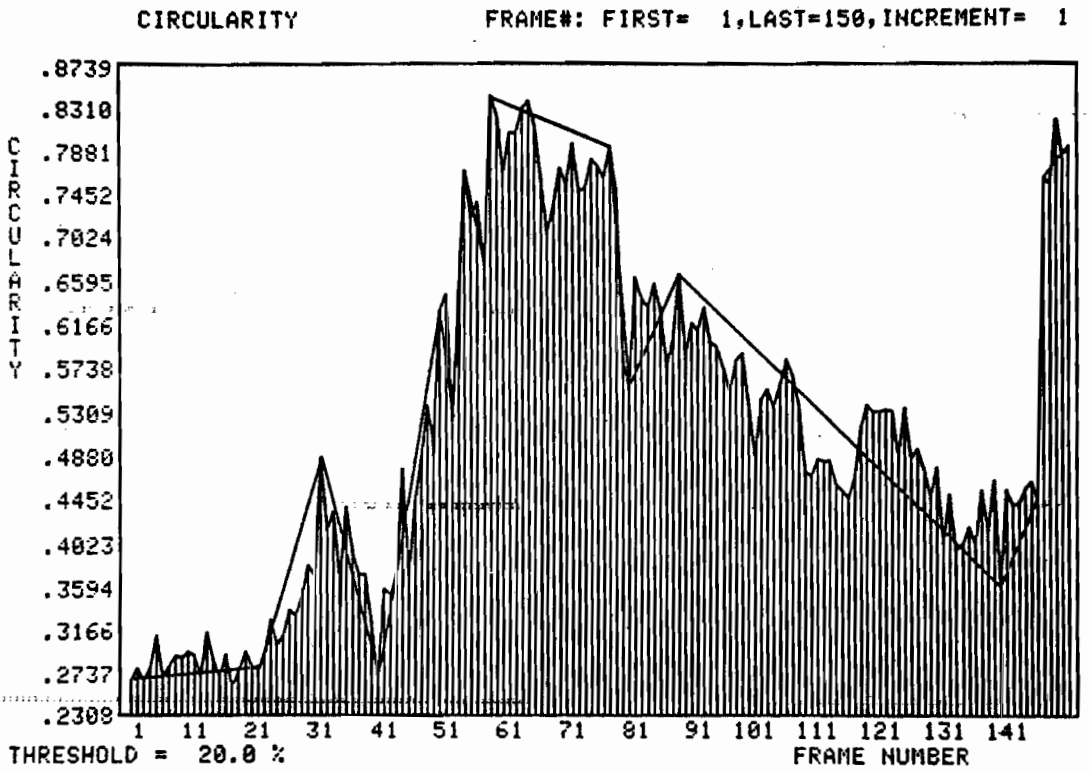
Pi(tj) is the value of the property (i) at time (tj).

The key frames of a specific property represent the first type of global change of the property under analysis. Thus, between each two sequential key frames (two sequential vertices on the curve approximation) the property has a constant dynamic behaviour (increasing, descreasing, or stationary). This behaviour can be described from the information included in the key frames. At this level, the following global data can be computed for each period between two sequential key frames:

TIME OF CHANGE
==========

$$TKF(i, i+1) = [t(i+1) - ti + to)\ to \quad \text{seconds}, \qquad (7.6)$$

where ti, t(i+1) are the times of key frames i and i+1 respectively, and to is the time interval between two




***Figure(7.3) Curve fitting of the static data shown in figure(7.2). An iterative splitting algorithm is used to compute a piecewise linear approximation to the curve.***

sequential frames.

AMOUNT OF CHANGE
==========

$$CKF(i,i+1) = Pi+1 - Pi \ , \qquad (7.7)$$

where Pi, Pi+1 are the property values at key frames KF(i) and KF(i+1) respectively.

RATE OF CHANGE
==========

$$RKF(i,i+1) = \tan^{-1} [\ CKF(i,i+1) \ / \ TKF(i,i+1)\ ]. \qquad (7.8)$$

In a similar fashion to that described in the preceding chapters, the values of TKF, CKF, and RKF can be qualified in order to summarize the dynamic changes in the property under analysis, as follows:

| TIME OF CHANGE | TYPE OF CHANGE | AMOUNT OF CHANGE | RATE OF CHANGE |
|---|---|---|---|
| TKF | CKF(0/+/-) | \|CKF\| | RKF |
| VERY SHORT | STATIONARY | NEGLIGIBLE | VERY SLOW |
| SHORT | INCREASING | SLIGHT | SLOW |
| AVERAGE | DECREASING | AVERAGE | AVERAGE |
| LONG | | CONSIDERABLE | FAST |
| VERY LONG | | SIGNIFICANT | VERY FAST |

The above analysis is meant to describe the global changes in the dynamic behaviour of a specific property. For example, the generated description of the ELONGATION property could be:

"For a SHORT time, the ELONGATION was INCREASING at a VERY SLOW rate, causing a SLIGHT INCREASE in the ELONGATION. Then for a LONG time it was STATIONARY. This was followed by a VERY FAST DECREASE in a VERY SHORT time."

Thus, at this level, the global description of a specific property can be given over a number of periods. Each period is bounded by key frames representing the initial and final frame numbers, and the property qualifiers (levels) at each key frame. From this data, the amount and rate of change in each period can be computed. Table (7.1) summarizes the numerical data resulting from the curve approximation of the data that is shown in Figure (7.3).

## 7.2.2 Qualification Of Level Changes

In order to summarize the global changes in the qualification levels of a specific property, two methods can be used: (a) applying the representational rules directly to the static and incremental symbolic qualifications; (b) applying the representational rules to the key frame data. Method (a) is recommended only if the generated description from curve approximation is not required. Otherwise, method (b) is obviously faster and will result in a more precise description. This is because, in the curve approximation, most of the irrelevant and noisy data has already been removed, and the information reduced to only that included in the key frames. In this case, the data associated with the key frames represent the input for higher level processes and rules. The objective is to split and merge these periods between the key frames in order to generate a summary of the quantification level changes. The procedures of these higher level processes are described below.

## SPLITTING AND MERGING THE PERIODS BETWEEN KEY FRAMES

In order to qualify and describe a shape property through a period of time with the appropriate qualifier (descriptor), the periods between the key frames, may be split or merged, according to specific rules. The objective is to eliminate the irrelevant, noisy, or artifactual changes, and generate a new sequence of periods. The property in each of these periods has the same qualifier.

### SPLITTING

The output of the curve approximation consists of a number of periods, each period being bounded by two key frames. If the symbolic qualifier of the property in these two key frames is different, we split the period bounded by them into a number of periods, such that, the property in each period has the same qualifier. For each new period, a key frame is initiated and the key frames set is updated. This can be achieved by applying the follwing rule :

RULE(7.1):

IF Q(Pk,KFi) .NE. Q(Pk,KFj)

==then==> 1) SPLIT Tij INTO Ti1, T12, T23, ..., Tmj

2) KF(u+m) <--- KF(u) , u=j,n

2) INITIATE KF(i+1), KF(i+2), ..., KF(i+m)

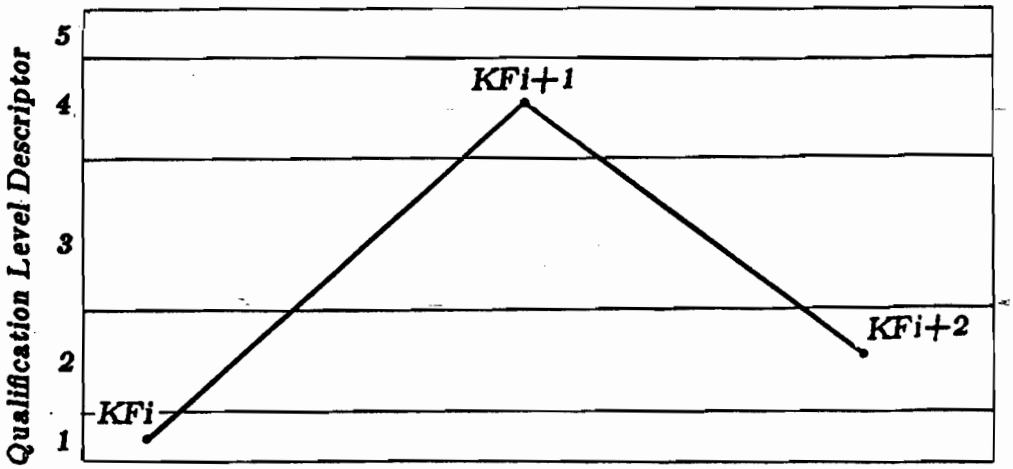
where Q(Pk,KFi) and Q(Pk,KFj) are the property qualifiers in key frames KF(i) and KF(j), respectively. For example, if Tij is the period bounded by key frames KF(i), KF(j)

respectively, where the descriptor is different at those key frames, and the period Tij is split into four periods Ti1, T12, T23, T3j, then the three key frames KF(i+1), KF(i+2), KF(i+3), should be initiated, and inserted in the key frame set: kF(j)-->KF(j+3), KF(j+1)-->KF(j+4), ..,KF(n)-->KF(n+3). Figure (7.4a) shows two sequential periods resulting from the curve approximation, and Figure (7.4b) shows the result of splitting them into a number of periods. Table (7.2) shows the output of applying the splitting rule to the data given in Table (7.1). The output of the splitting operation consists of a sequence of periods, with the property in each having the same qualifier.

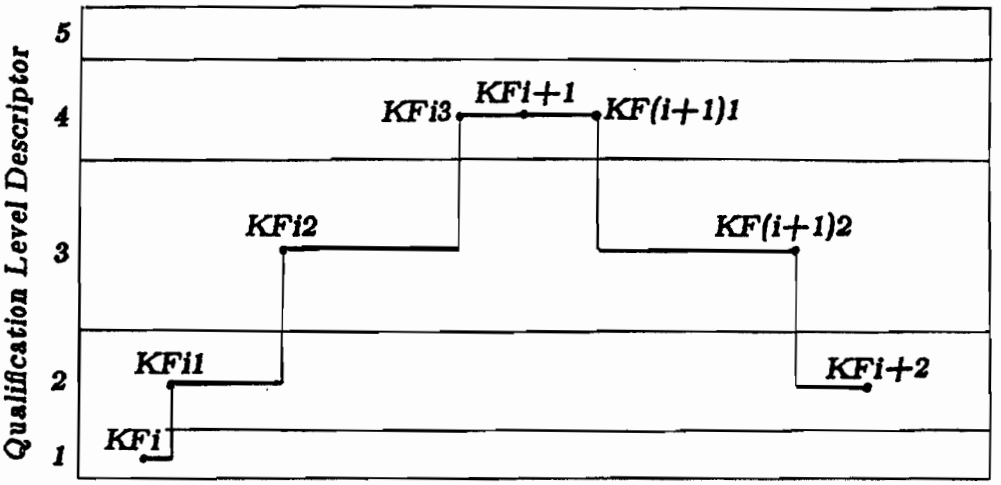
## MERGING

The splitting operation might result in some sequential periods that have the same qualifiers, as shown in Figure (7.4b), or it may produce very short periods (see Table 7.2). The latter are usually caused by noise or changes due to undesirable experimental conditions. These periods can be merged according to other rules. For example, if a specific property has the same qualifier for a sequence of periods, then these periods can be merged. The property during the merged period can be described by the same descriptor, and the key frames updated. This strategy may be modeled by the following rule:

RULE(7.2):




*Figure(7.4a) Two periods bounded by three sequential key frames, resulting from the curve approximation of the static data. In each period, the dynamic behaviour of the characteristic is constant. Between KF(i) and KF(i+1) the characteristic is increasing. During the next period it is decreasing.*




***Figure(7.4b) Splitting the above periods into a sequence of periods, where in each period the property has the same qualification descriptor. By further analysis, these periods may be merged using high level representational rules.***

*Figure(7.4) Splitting periods in order to generate a descriptive summary of the global changes in a specific property. (a) input, (b) output.*

```
  IF      Q(Pk,Ti,i+1) .EQ. Q(Pk,Ti+1,i+2) .EQ. ... .EQ.
          Q(Pk,Tj-1,j) .NE. Q(Pk,Tj,j+1)

=then=> 1) MERGE:
             Ti,i+1 + Ti+1,i+2 + ... + Tj-1,j  -INTO-> Tij
          2) Q(Pk,Tij)   <--- Q(Pk,Ti,i+1) ;   ! Assign a qualifier
                                                 to the merged period.
          3) KF(u-j+i+1) <--- KF(u),   u=j,n   !
                                               ! Updating
          4) DELETE  KF(u)  , u=[n-(j-i-1),n]  !
                                               ! key frames
          5) n <--- n-(j-i-1)                  !
```

where n is the number of key frames. Note that Tij is the merged period that includes the periods {Ti,i+1, Ti+1,i+2,...,Tj-1,j}, and the key frame KF(j) is updated to KF(i+1), KF(j+1) to KF(i+2),....and so on. Table (7.3) shows the result of applying the above rule to the data in Table (7.2).

The above is an example of a simple rule, based only on the dynamic data of the different properties. The second step in creating global periods, is concerned with distinguishing the significant changes from the irrelevent or noisy ones. In this case, a more sophisticated approach would utilize the dynamic description of the different properties, in conjunction with logical knowledge constraints. For example, an inference process could eliminate the very short events. Thus, suppose a specific property Pi is measured through three sequential periods T01, T12, and T23. If the time T12 is very short relative

to both T01 and T23 (T01,T23 >> T12), then we might eliminate T12 by one of two actions: first, by merging all three periods T01,T12,T23 into one (T03), or second, by merging T12 to either T01 or T23. Thus, if the cell was described in three sequential periods as SMALL, VERY SMALL, and SMALL, then a merging of these periods would result in SMALL as the description. This may be achieved by the following rule:

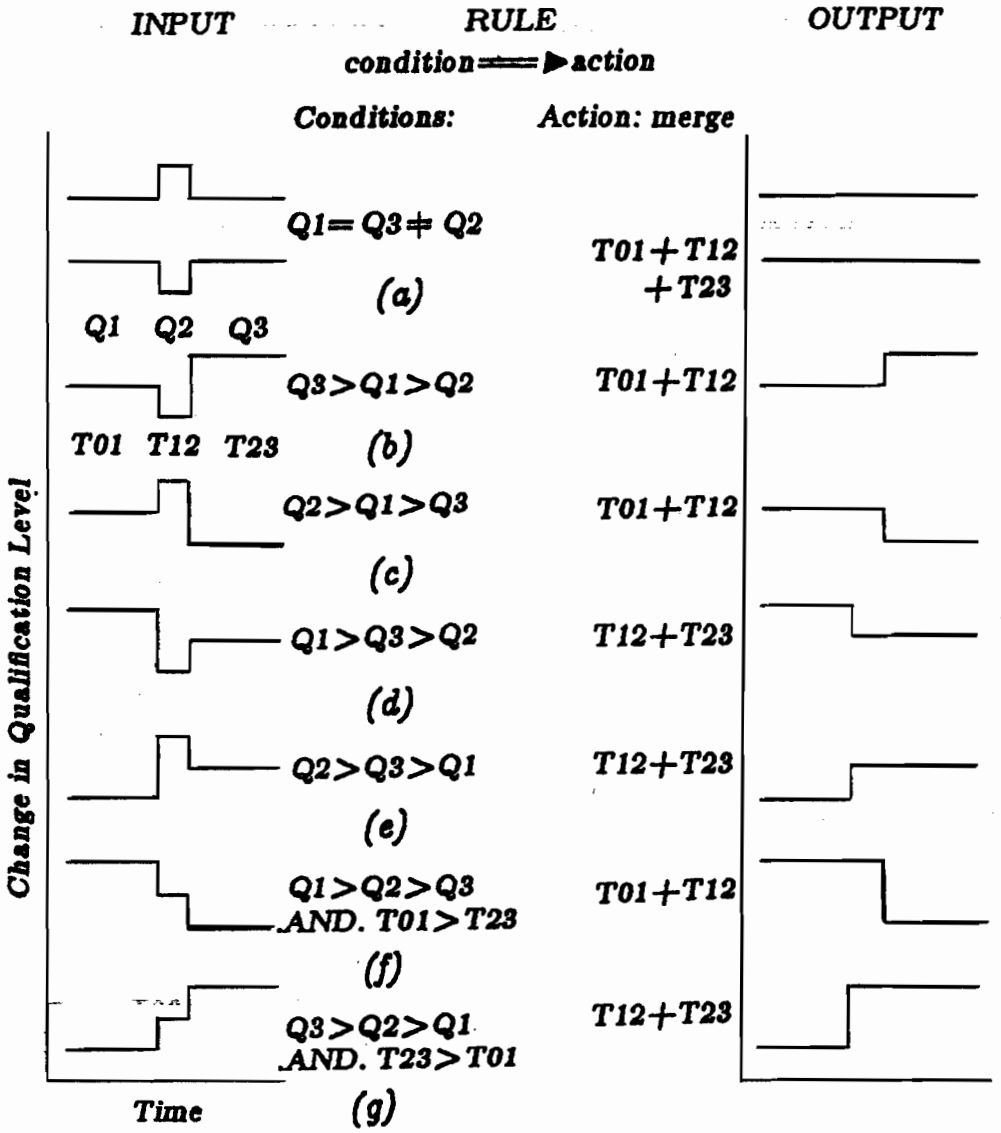
RULE(7.3):

IF (T01 .GTT. T12 .LTT. T23) .AND.
Q(Pi,T01) .EQ. Q(Pi,T23) .AND.
Q(Pi,T01) .NE. Q(Pi,T12),

=then=> 1) MERGE THE THREE PERIODS INTO T03
2) UPDATE THE KEY FRAMES

where T03 = T01 + T12 + T23, and Q(Pi,T13) = Q(Pi,T01) = Q(Pi,T23). Figure (7.5a) illustrates the function of this rule. In this figure, the values Q(Pi,T13), Q(Pi,T01), and Q(Pi,T23) are presented as Q1, Q2, and Q3, respectively. The details of the merging action, steps 1 and 2 in the above rule, is similar to that described in Rule 7.2.

A second action, in which T12 may be merged to either T01 or T23, is based on logical inference. For example, if the length of the cell was described in three sequential periods as being SHORT, VERY SHORT, MEDIUM, then one may logically deduce that it has changed from SHORT to MEDIUM, if the description of the very short period (T12) is the




*Note: In all the above figures, T01>>T12<<T23, Q is the qualifier*

***Figure(7.5)*** *Using condition===>action rules to eliminate irrelevant and noisy changes.*

same as that of either the preceding or following period. Thus, if the description of the VERY SHORT period is closer to the SHORT period rather than to MEDIUM period, period T12 may be merged to T01. This logical deduction may be accomplished by the following rule:

RULE(7.4):

```
      IF     (T01 .GTT. T12, .AND. T12 .LTT. T23)
        .AND.(Q1 .GT. Q2 .AND. Q2 .LT. Q3 .AND. Q1 .LT. Q3),
        .OR. (Q1 .LT. Q2 .AND. Q2 .GT. Q3 .AND. Q1 .GT. Q3),
   =then=> 1) MERGE PERIODS T01 AND T12 INTO T02,
           2) UPDATE KEY FRAMES
```

where T02 = T01 + T12, and Q(Pi,T02) = Q(Pi,T01). Figure (7.5b) illustrates the function of this rule. Other examples of similar rules are shown in Figures (7.5c-7.5g). Table (7.4) gives the final global periods after applying the merging rules on the data in Table (7.3). From this table one can see that the number of the global periods has decreased from 43 (Table 7.2) to 10 (Table 7.4) by applying the rules discussed. Also, all periods which have duration of one frame have been eliminated.

The final step in this analysis is to characterize the data and describe them symbolically in a fashion similar to the preceding chapters. For example, the time for each period Tij may be normalized and described symbolically as VERY SHORT, SHORT, MEDIUM, LONG, or VERY LONG, and the property during each period can be qualified as described in

Chapter 4. In the following section, we will show how to use the above methodology to describe the global changes in each individual shape property.

## 7.3 GLOBAL CHANGES IN THE DESCRIPTION of Individual Shape Properties

In Section 4.5 we defined the factors influencing the selection of properties used by the system for describing the different aspects of the dynamic behaviour. In addition to these factors, for shape description we require that the method be translation, rotation, and size independent. Based on these criteria and from our experimental work, we found that the most efficient properties for cell shape description are:

(1) Circularity

(2) Average Bending Energy

(3) Angle Regularity

(4) Elongation

(5) Number of Concave Angles

(6) Number of Subparts

Using the methodologies described in Sections 7.2.1 and 7.2.2, the global changes in each of these properties can be described. First, the static and incremental data associated with each property should be normalized to range between zero and one. Zero corresponds to the simplest shape and one to the most complex. For example, the definition of elongation EL is:

$$EL = Width / Length . \quad (7.9)$$

From this definition, one can see that EL = 1 for shapes resembling squares, and EL = 0 for filamentary-like shapes, in which the width is negligible relative to the length. Accordingly, square-like shapes are assumed to be simpler than elongated ones. Therefore, the computation of the elongation can be modified to:

$$EL = 1 - ( Width / Length )$$
$$= | Length - Width | / Length. \quad (7.10)$$

In this case EL = 0 for square-like shapes, and EL = 1 for filamentary shapes. Another example can be given for circularity, which is defined as:

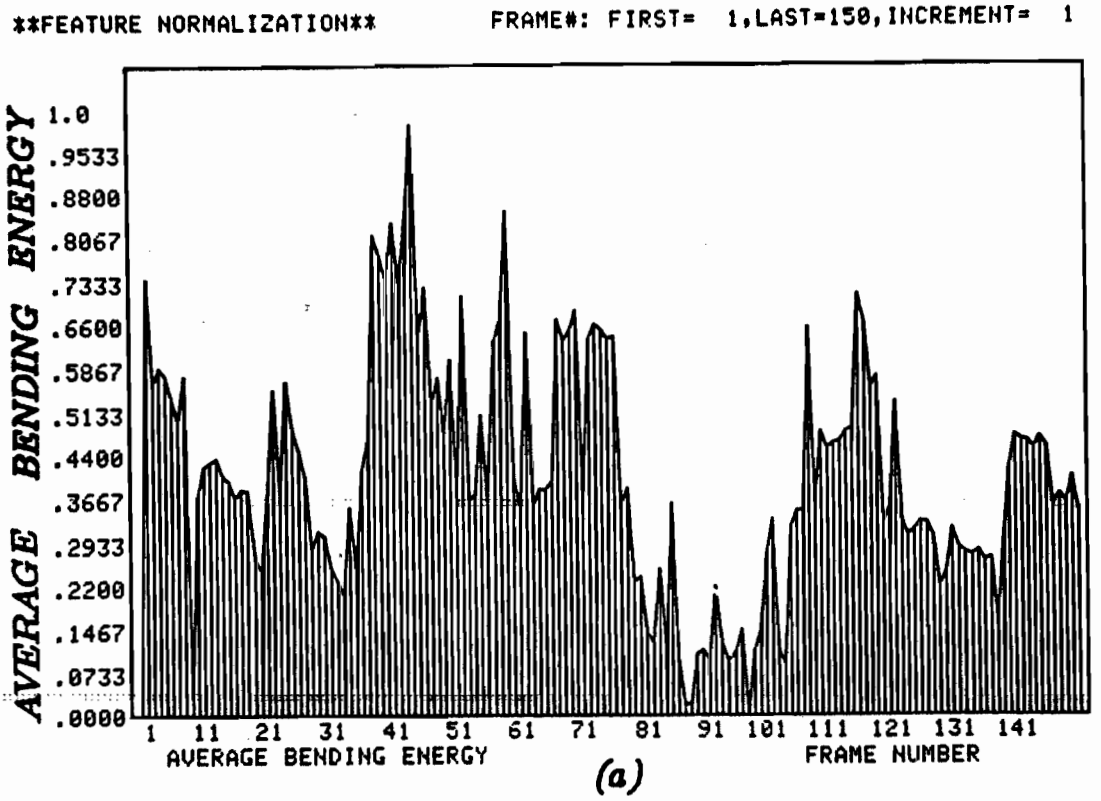
$$CR = P^2 / 4 \pi A \quad (7.11)$$

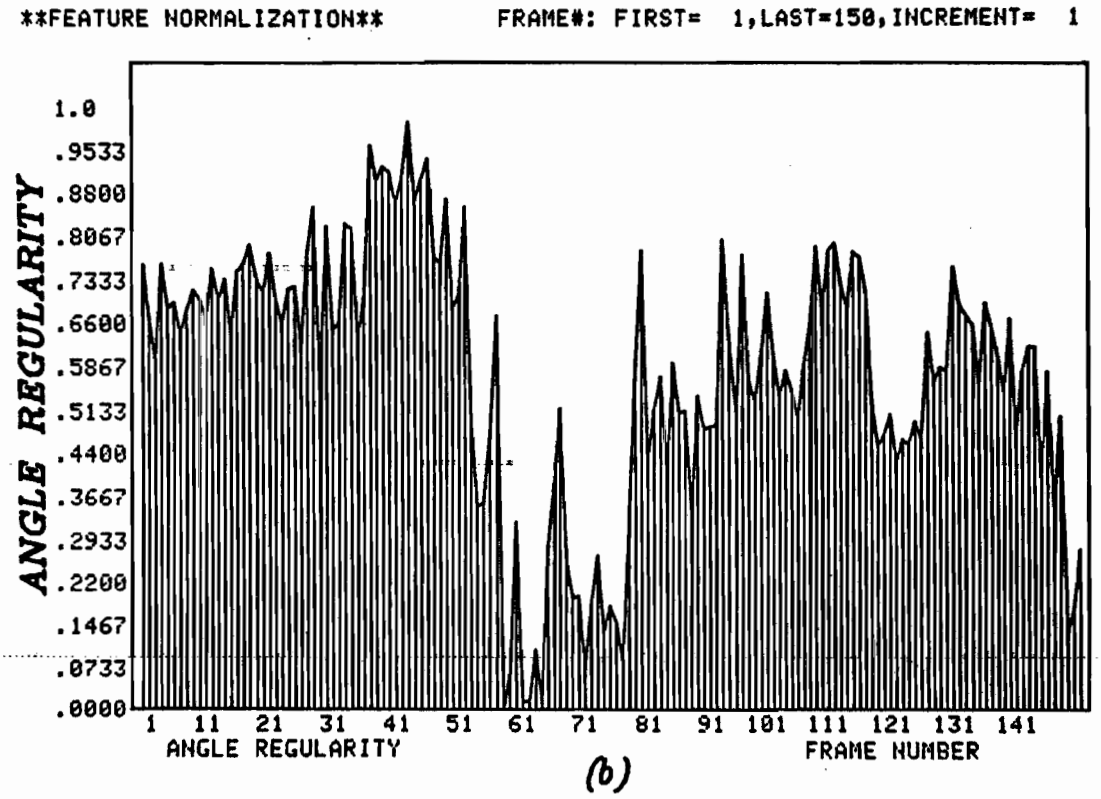
where P is the perimeter and A is the area. This expression can be normalized as:

$$CR = 1 - ( P^2 / 4 \pi A ). \quad (7.12)$$

Thus, CR = 0 for the most circular shapes, and CR = 1 for the least circular ones. Figures (7.3), (7.6a) and (7.6b) show examples of the normalized values of the static data for different shape properties computed for 150 frames in sequence.

The normalized static data for each property are analyzed according to Sections 7.2.1 and 7.2.2, using the same sequence of steps used for curve analysis. That is, splitting, merging, and finally generating summaries




***Figure(7.6) The normalized values of two shape properties in a sequence of 150 frames. (a) Average Bending Energy (b) Angle regularity***

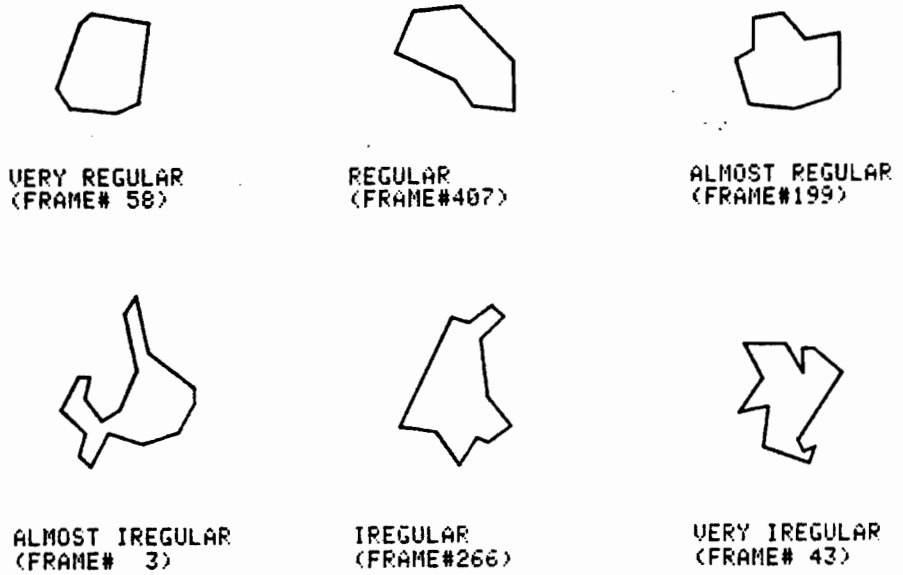
describing their global changes. Typical examples of these summaries are given in Description (7.1). The symbolic qualifiers which are used to describe the different shape properties are given in Table (3.2). Examples of cell shapes that are described by each qualifier are given in Figures (7.7a-7.7b).

## 7.4 MEMBRANE SHAPE: GLOBAL CHANGE DESCRIPTION

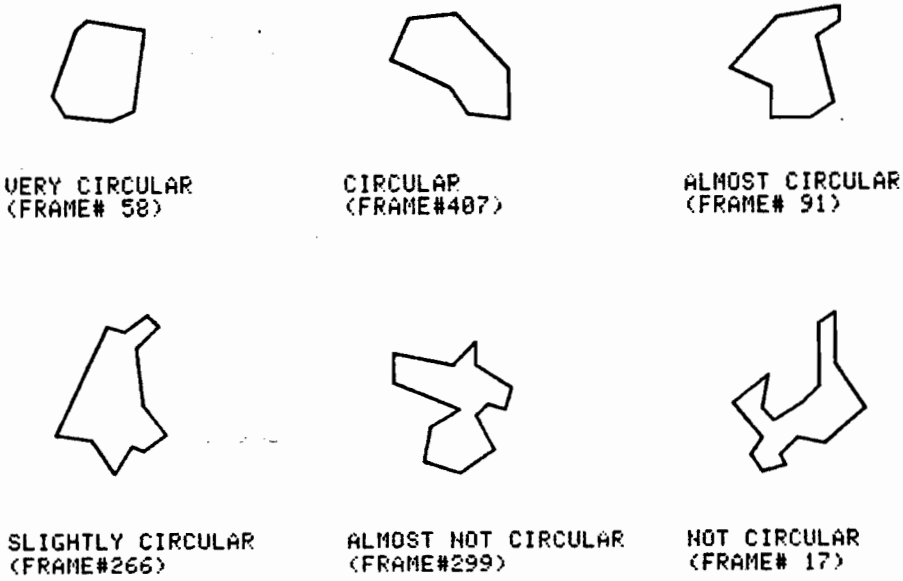
In the preceding sections we discussed the basic methodology for detecting and describing the global changes in each of the shape properties. In this section, we will discuss how to integrate these properties in order to describe the changes in the membrane shape.

The task of describing the cell membrane shape is similar to that of describing the silhouette (contour) of an arbitrary shape. This task may be considered one of the difficult issues in computer vision and pattern recognition. In spite of the enormous research and number of publications on the subject, there is as yet no established technique for solving this problem. However, we should recognize that we are trying to imitate one of the most complex processes of human visual perception.

Most previous work has attempted to solve the problem by describing the shape in terms of the properties of the object boundary. These techniques have achieved reasonable success in static scene analysis. However, in dynamic scene




***(a) ANGLE REGULARITY***




***(b) CIRCULARITY***

***Figure(7.7) Examples of cell shape property characterization and description. (a) Angle regularity (b) Circularity.***

analysis, it is necessary to describe the changes in shape as well. This problem has been almost completely neglected. In our research, besides the difficulties of describing an arbitrary static shape, we also want to describe the random changes in the shape of non-rigid moving objects.

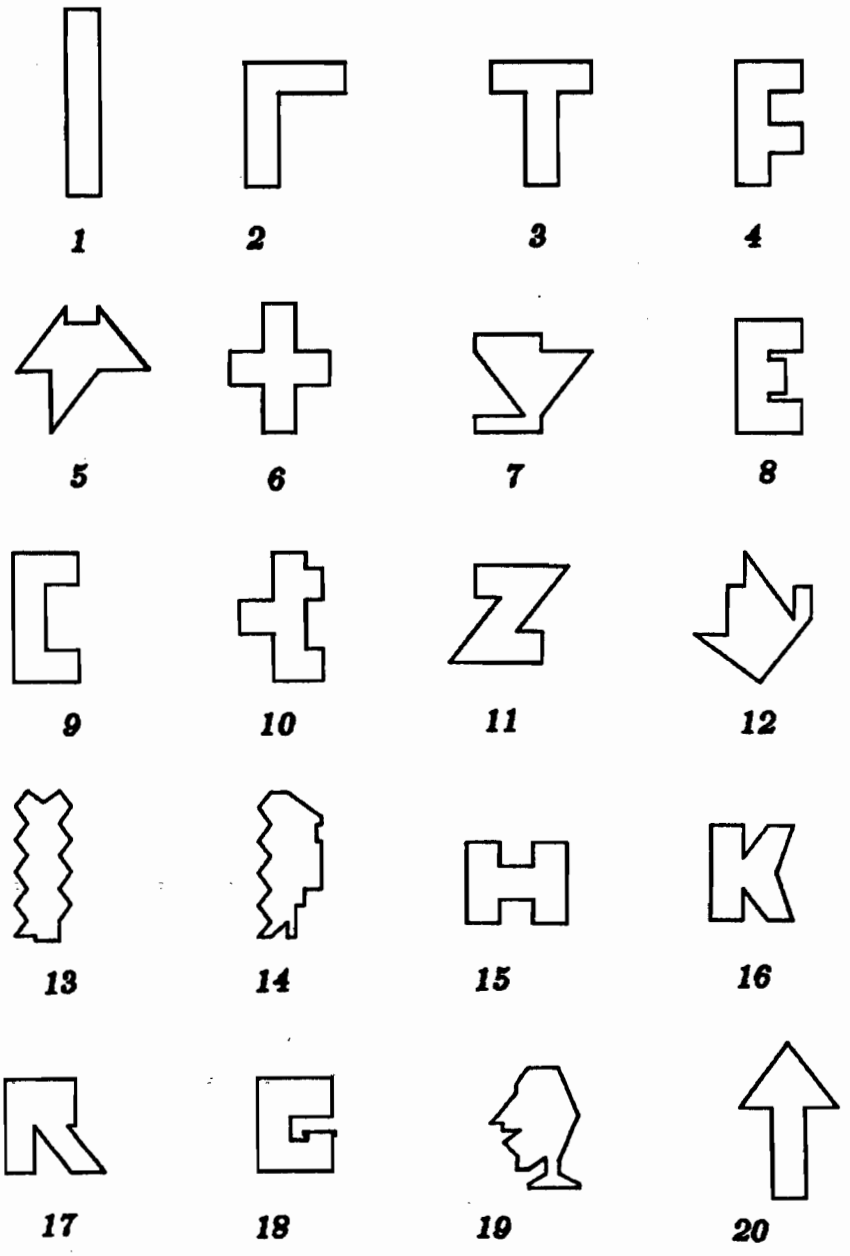
From the previous analysis of the individual shape properties, one can see that each of them describes only a specific shape property. But the change in object shape does not necessary match the equivalent change in a particular property. In other words, we may find that many different shapes have the same value for a specific property.

OBSERVATION: There is no known single shape property that gives a unique value for each different shape.

The shape properties indicated above are those referred to as information nonpreserving [Pavlidis, 76]. Examples are circularity, elongation, and regularity. On the other hand, there are some shape measurements that are unique for different shapes, and are termed information preserving. For example, if we take the chain code of a digital contour and string the digits together, the resulting number is unique for each shape. However, this representation is clearly not useful as a global description for the shape.

In order to illustrate the above observation, we use as an example, one of the earliest and most established shape descriptors, that is, circularity. Figure (7.8) shows 20 different shapes that are designed such that all of them have the same area and perimeter. From this figure, one can see that a wide variety of different shapes can have the same circularity value. In a similar fashion, we can show the above observation for other shape properties, such as for example, elongation, regularity, and average bending energy. Therefore, in order to generate different descriptions for different shapes, we must use more than one property. The perfect description is the one that fits only one shape. In order to achieve this, we may have to use an infinite number of descriptors. Since this is impossible, we must find the minimum and most efficient set of descriptors that generates complete informative descriptions for the different shapes.

For example, in order to choose descriptors for different classes of neutrophil cells, categorized as juvenile, banded, segmented, and hypresegmented, Liu [Liu, 76] studied the classification power of different descriptors. He discriminated between the power of the different shape descriptions as follows: "A shape description, D1, is more informative than another description, D2, if the set of shapes that can be described by D1, is included in the set of shapes described by D2". For example, the description of a given shape as a square or




***Figure(7.8) Different shapes having the same area and perimeter, and hence the same circularity.***

rectangle is more informative than describing it as quadrangle.

In dynamic scene analysis of non-rigid moving objects, the task of finding the set of properties that generates complete informative descriptions is more difficult. This is because, besides the above requirements (minimum number of properties and unique static descriptions for the different shapes), the change in the generated descriptions should correspond to the change in the shape, and be insensitive to noise. Our approach is based on forming a mathematical expression which can be computed from the individual shape properties. This expression represents the degree of membrane shape complexity. In this way, the change in its value can be used as a measurement for the change in the membrane shape.

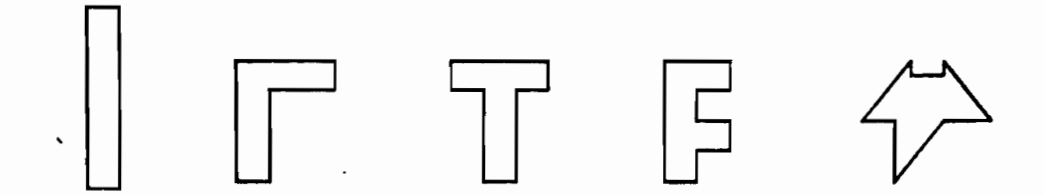
## Expression for Membrane Shape Desription

In order to form an expression for the measurement of an arbitrary shape, we choose the minimum set of properties based on two criteria: (a) maximum discrimination between the different shapes, and (b) descriptive power related to shape complexity. To satisfy these criteria, we have used the shapes given in Figure (7.8) as a training set. We computed the value of each shape property for every shape in the training set. Then, we selected the properties that: (a) produced the same value for a minimum number of shapes, and (b) sorted the complexity of the shapes in a manner

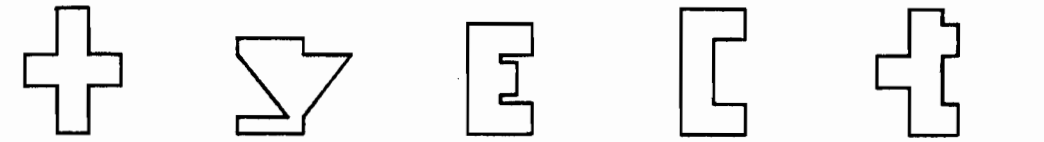
similar to that of human sorting. In this experiment, we did not include circularity, because the shapes were originally designed to give the same value for circularity. However, circularity was tested separately using some of the cell shapes (see Figure 7.7b).

From the above simple experiment, we selected the following shape properties in order to form an expression for the membrane shape measurement: circularity, average bending energy, and angle regularity. Table (7.5a) gives the result of computing the property values for the different shapes in the training set. Figure (7.9) shows the shapes associated with the values of the different properties, and Figure (7.10) is a sort of the shapes according to complexity, using each individual property.

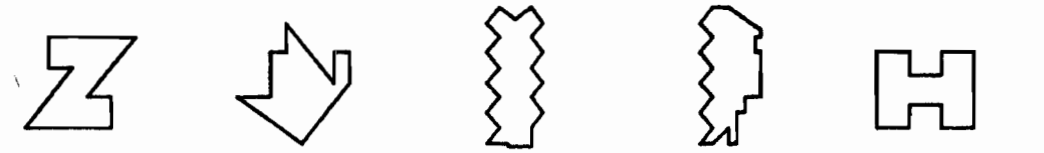
To combine the three selected properties into one expression, we have repeated the above experiment, using the arithmetic and geometric means of the properties. The result of this experiment is given in Table (7.5b) and is shown in Figure (7.11). From this data, we can see that the mean of more than one property is more efficient than an individual one. Based on this experiment, we have selected the geometric mean of circularity, average bending energy, and angle regularity, as a measure for the cell membrane shape.



| | | | | |
|---|---|---|---|---|
| PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 |
| AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 |
| CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 |
| ANGR: 0.000 | ANGR: 0.417 | ANGR: 0.500 | ANGR: 0.525 | ANGR: 0.767 |
| AVBE: 0.007 | AVBE: 0.007 | AVBE: 0.013 | AVBE: 0.022 | AVBE: 0.021 |
| AVGM: 0.001 | AVGM: 0.124 | AVGM: 0.160 | AVGM: 0.192 | AVGM: 0.216 |
| AVAM: 0.207 | AVAM: 0.346 | AVAM: 0.376 | AVAM: 0.387 | AVAM: 0.468 |



| | | | | |
|---|---|---|---|---|
| PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 |
| AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 |
| CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 |
| ANGR: 0.533 | ANGR: 0.502 | ANGR: 0.533 | ANGR: 0.500 | ANGR: 0.536 |
| AVBE: 0.032 | AVBE: 0.010 | AVBE: 0.065 | AVBE: 0.013 | AVBE: 0.030 |
| AVGM: 0.219 | AVGM: 0.145 | AVGM: 0.277 | AVGM: 0.157 | AVGM: 0.216 |
| AVAM: 0.394 | AVAM: 0.376 | AVAM: 0.405 | AVAM: 0.376 | AVAM: 0.394 |

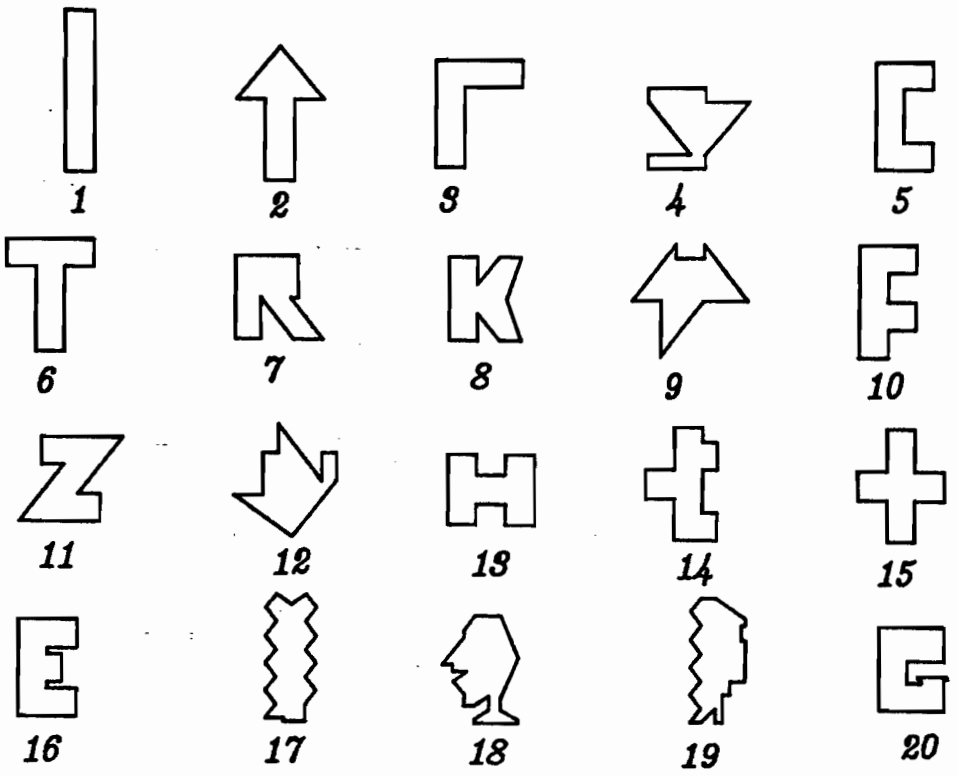


| | | | | |
|---|---|---|---|---|
| PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 |
| AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 |
| CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 |
| ANGR: 0.637 | ANGR: 0.599 | ANGR: 0.462 | ANGR: 0.467 | ANGR: 0.533 |
| AVBE: 0.024 | AVBE: 0.025 | AVBE: 0.084 | AVBE: 0.121 | AVBE: 0.029 |
| AVGM: 0.212 | AVGM: 0.212 | AVGM: 0.216 | AVGM: 0.216 | AVGM: 0.219 |
| AVAM: 0.425 | AVAM: 0.413 | AVAM: 0.387 | AVAM: 0.401 | AVAM: 0.393 |

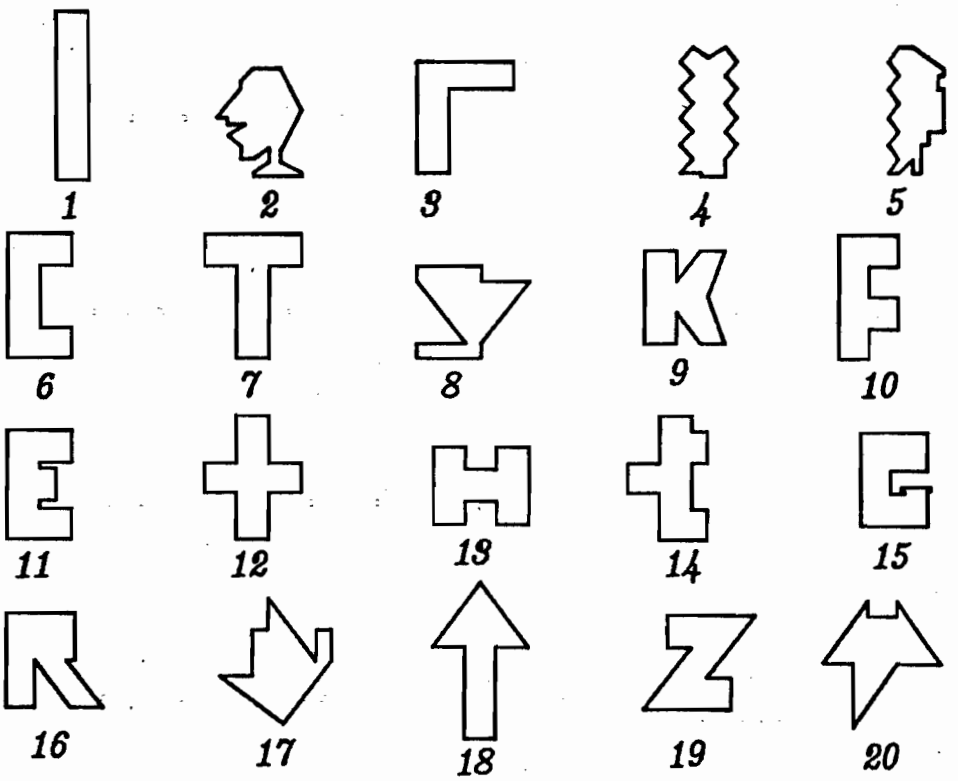
| | | | | |
|---|---|---|---|---|
| PERM: 283.2 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 | PERM: 280.0 |
| AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 | AREA:2400.0 |
| CIRC: 0.624 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 | CIRC: 0.615 |
| ANGR: 0.520 | ANGR: 0.556 | ANGR: 0.536 | ANGR: 0.417 | ANGR: 0.629 |
| AVBE: 0.021 | AVBE: 0.013 | AVBE: 0.593 | AVBE: 0.096 | AVBE: 0.007 |
| AVGM: 0.277 | AVGM: 0.288 | AVGM: 0.291 | AVGM: 0.326 | AVGM: 0.580 |
| AVAM: 0.388 | AVAM: 0.395 | AVAM: 0.581 | AVAM: 0.376 | AVAM: 0.417 |

***Figure(7.9) Different property values of shapes having the same circularity.***

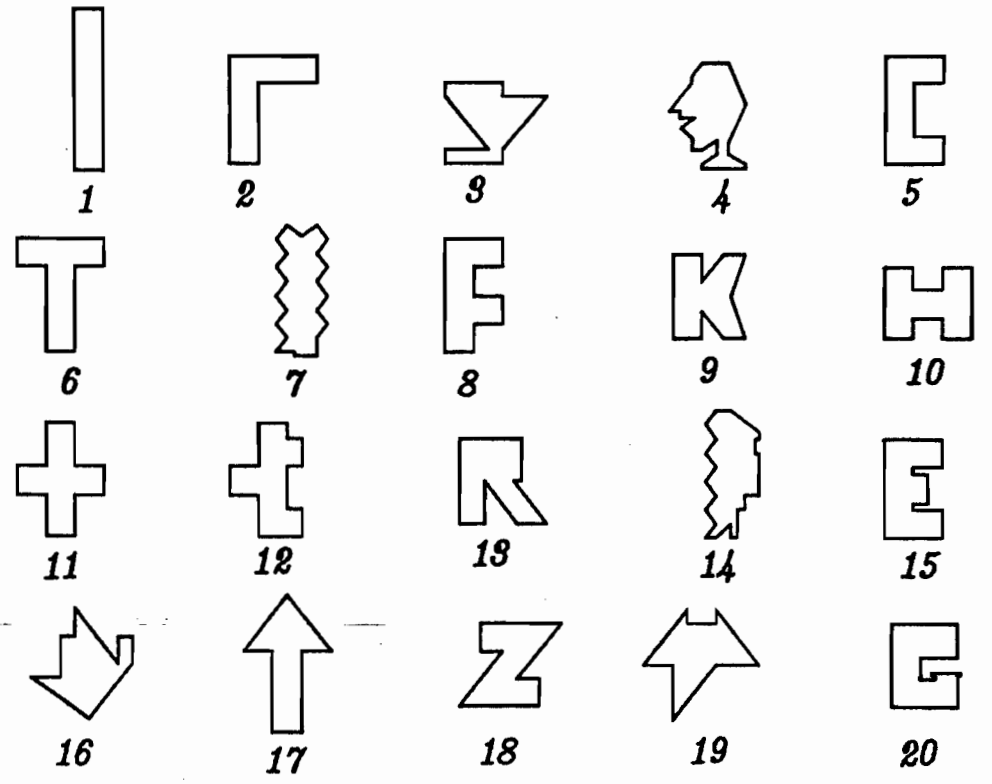
***PERM: Perimeter,*** ***AVBE: Average Bending Energy,***
***CIRC: Circularity,*** ***AVAM: Arithmetic Mean,***
***ANGR: Angle Regularity,*** ***AVGM: Geometric Mean.***



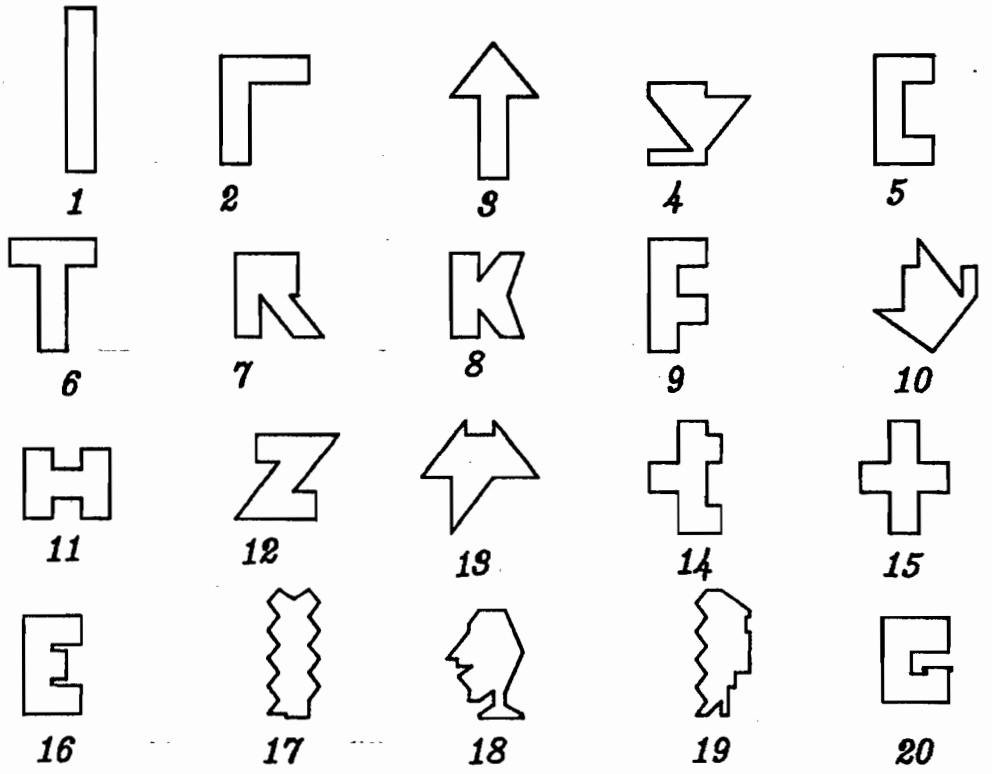

*Figure(7.10a) Different shapes sorted according to their complexity using average bending energy.*




*Figure(7.10b) Different shapes sorted according to their complexity using angle regularity.*




*(a) Sorting using arithemetic mean.*



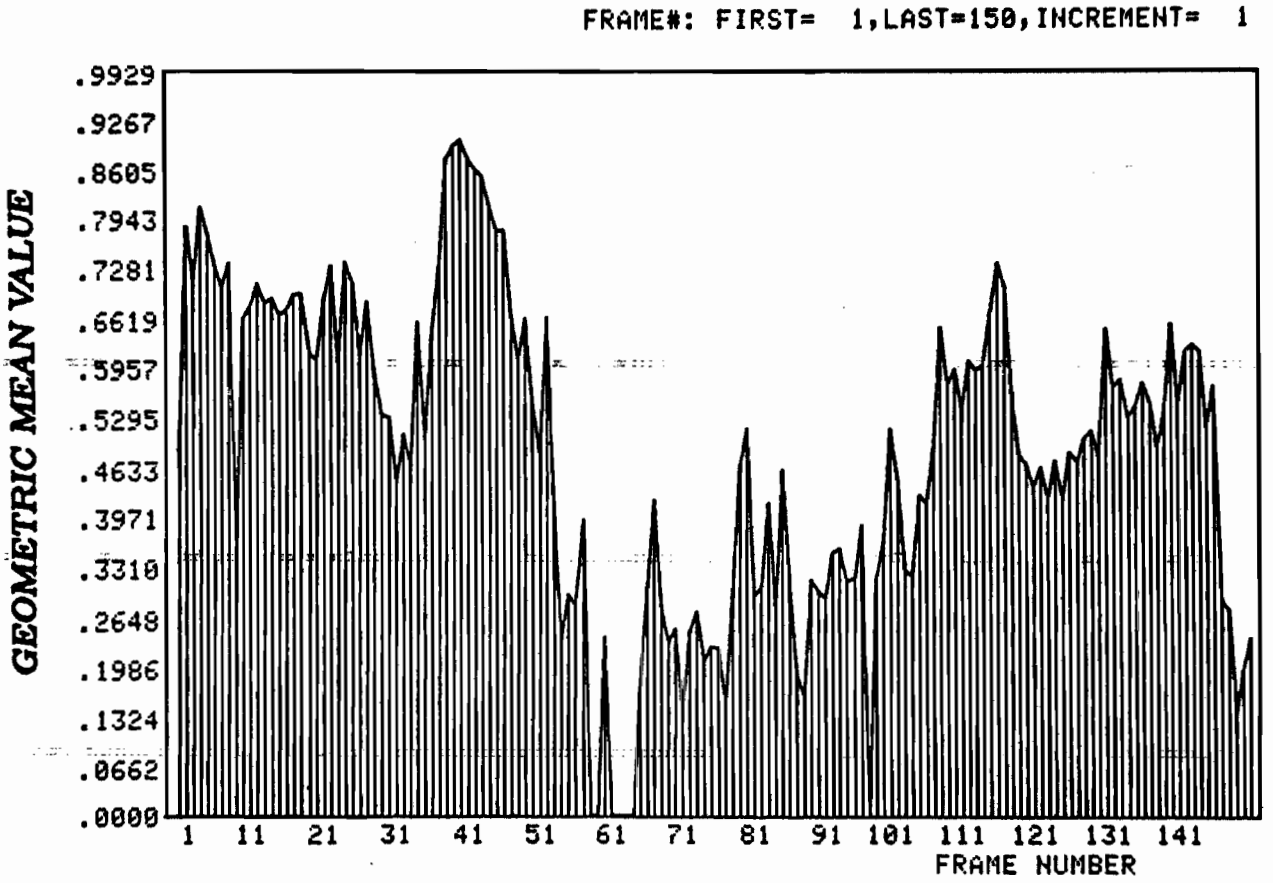

*(b) Sorting using geometric mean.*

*Figure(7.11) Different shapes sorted according to their complexity using the: (a) arithemetic mean (b) geometric mean, of the average bending energy and angle regularity.*

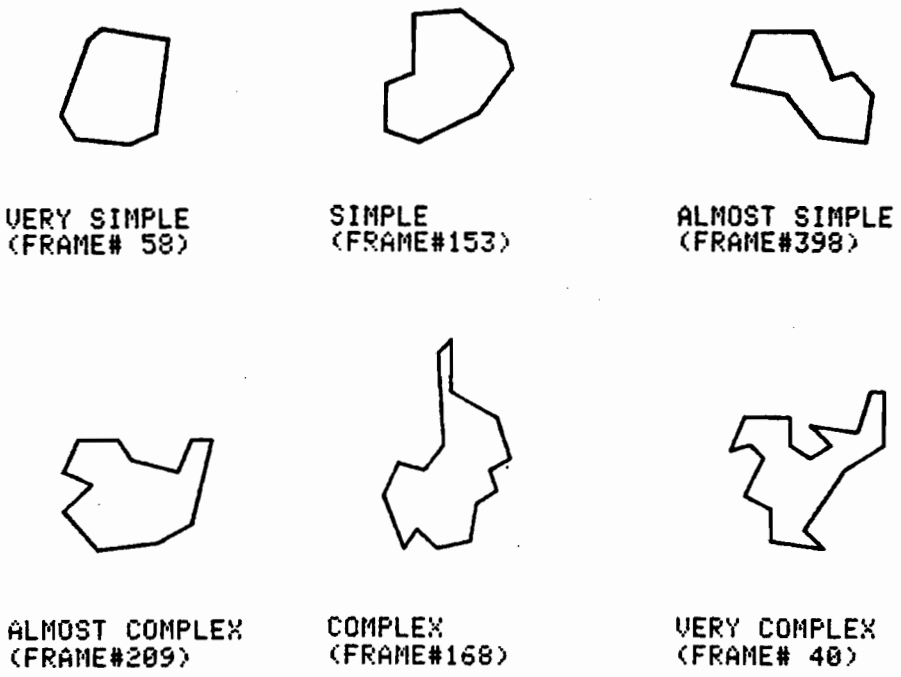
The geometric or arithmetic mean of the selected properties can be computed from static data. The result represents the static values of the membrane shape in the sequential frames as shown in Figure (7.12). This can be analyzed in a similar fashion to the individual shape properties in order to generate a summary of the global changes in the cell membrane. This will include detecting key frames and assigning descriptors to both the dynamic trends and the actual values of membrane shape. The different symbolic qualifiers that are used to describe the membrane shape are: VERY SIMPLE, SIMPLE, ALMOST SIMPLE, ALMOST COMPLEX, COMPLEX, VERY COMPLEX. Note that here we are using six categories for the symbolic qualification in order to give a more precise description near the average level. In fact, the qualifiers ALMOST SIMPLE and ALMOST COMPLEX combined, are the same as the qualifier AVERAGE that is given in most of the preceding descriptions. Figure (7.13) shows examples of cell shapes that are characterized by the different descriptors. A typical example of the generated summary of the global membrane shape changes is given in Description (7.1).

## 7.5 SUMMARY

Shape description represents the high level stage of shape perception by the system. The objective is to analyze the data resulting from the low level shape analysis processes in order to generate a summary containing all the




***Figure(7.12) Geometric mean of circularity, average bending energy, and angle regularity computed for cell shapes in a sequence of 150 frames.***




***Figure(7.13) examples of cell shape characterization.***

relevant information pertaining to the shape of the object under analysis. In spite of the difficulties in describing the arbitrary shape of an object in a static scene, in our research we wish to describe the significant changes in the shape of a non-rigid moving object.

In this chapter, we have demonstrated a methodology for detecting and describing the changes in shape of a moving cell from a sequence of images. The input to these processes consists of the static description of the cell shape properties in each frame of the sequence. The output is a summary that characterizes the changes in each of the cell shape properties individually, as well as the global membrane shape itself. The description is given as a sequence of periods bounded by key frames. The latter represent the transitions where significant changes have occured.

A global change is defined as a significant change in the dynamic behaviour or the level of the symbolic qualifications. The former occur when a specific shape property changes its dynamic behaviour; for example, from being stationary to increasing or decreasing. The key frames that define the times of these changes can be computed from the curve that represents the static data of the property under consideration. In order to achieve this, a curve approximation technique similar to that of the polygonal approximation is used to detect the relevant information from the irrelevant and noisy data.

The global changes in the level of the qualification descriptors can be accomplished by using the high level LTM representational rules. The latter utilize the dynamic data and constraint knowledge of the cell shape properties. The representational rules can be applied directly to the symbolic qualifications of the static shape properties, or their curve approximation. The latter has the advantage that the data is reduced to those represented by the key frames, and most of the irrelevant and noisy changes are removed by the curve approximation. The processes that detect the global changes in the qualification levels use splitting and merging of the periods between sequential key frames.

We have show experimentally that a single shape property cannot be used to describe the changes in the cell membrane shape. Therefore, we have developed an expression for measuring the complexity of an arbitrary shape pattern based on a group of selected shape properties that are location, rotation, and size independent. This expression is used to measure the complexity of the membrane shape in each frame of the sequence. The resulting data is represented by a curve that can be analyzed using techniques similar to the analysis of individual shape properties. In this way, the changes in the membrane shape can be detected, qualified, and described.

Finally, a summary of the global changes in cell shape and its individual properties is generated in meaningful symbolic terminology. A typical example of this summary is given in Description (7.1).

DESCRIPTION <7.1>

# GLOBAL SHAPE ANALYSIS AND DESCRIPTION

## INTRODUCTION

A summary of the global changes in the cell shape characteristics are described below. The description is given in two sections. First, the characterization of each of the main cell shape properties, and then, the global changes in the cell membrane shape are described. The description given is for the period including frame number(1) to frame number(150), a duration of 75.0 seconds.

## SHAPE PROPERTIES

### AVERAGE BENDING ENERGY

| PERIOD NUMBER | FRAME NUMBER | TIME | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 2 | VERY SHORT | SMOOTH |
| 2 | 3 --> 18 | SHORT | ALMOST SMOOTH |
| 3 | 19 --> 20 | VERY SHORT | JAGGED |
| 4 | 21 --> 30 | SHORT | ALMOST SMOOTH |
| 5 | 31 --> 35 | VERY SHORT | JAGGED |
| 6 | 36 --> 37 | VERY SHORT | ALMOST SMOOTH |
| 7 | 38 --> 44 | VERY SHORT | SMOOTH |
| 8 | 45 --> 78 | MEDIUM | ALMOST SMOOTH |
| 9 | 79 --> 85 | VERY SHORT | JAGGED |
| 10 | 86 --> 88 | VERY SHORT | VERY JAGGY |
| 11 | 89 -->103 | SHORT | JAGGED |
| 12 | 104 -->127 | MEDIUM | ALMOST SMOOTH |
| 13 | 128 -->138 | SHORT | JAGGED |
| 14 | 139 -->150 | SHORT | ALMOST SMOOTH |

## CIRCULARITY

| PERIOD NUMBER | FRAME NUMBER | TIME | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 25 | MEDIUM | NOT CIRCULAR |
| 2 | 26 --> 43 | SHORT | SLIGHTLY CIRCULAR |
| 3 | 44 --> 53 | SHORT | ALMOST CIRCULAR |
| 4 | 54 --> 57 | VERY SHORT | CIRCULAR |
| 5 | 58 --> 78 | SHORT | VERY CIRCULAR |
| 6 | 79 -->131 | LONG | ALMOST CIRCULAR |
| 7 | 132 -->137 | VERY SHORT | SLIGHTLY CIRCULAR |
| 8 | 138 -->145 | SHORT | ALMOST CIRCULAR |
| 9 | 146 -->149 | VERY SHORT | CIRCULAR |
| 10 | 150 -->150 | VERY SHORT | VERY CIRCULAR |

## ANGLE REGULARITY

| PERIOD NUMBER | FRAME NUMBER | TIME | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 13 | SHORT | ALMOST REGULAR |
| 2 | 14 --> 21 | SHORT | IRREGULAR |
| 3 | 22 --> 23 | VERY SHORT | ALMOST REGULAR |
| 4 | 24 --> 28 | VERY SHORT | IRREGULAR |
| 5 | 29 --> 32 | VERY SHORT | ALMOST REGULAR |
| 6 | 33 --> 36 | VERY SHORT | IRREGULAR |
| 7 | 37 --> 46 | SHORT | VERY IRREGULAR |
| 8 | 47 --> 52 | VERY SHORT | IRREGULAR |
| 9 | 53 --> 57 | VERY SHORT | ALMOST REGULAR |
| 10 | 58 --> 64 | VERY SHORT | VERY REGULAR |
| 11 | 65 --> 67 | VERY SHORT | ALMOST REGULAR |
| 12 | 68 --> 70 | VERY SHORT | REGULAR |
| 13 | 71 --> 77 | VERY SHORT | VERY REGULAR |
| 14 | 78 -->109 | MEDIUM | ALMOST REGULAR |
| 15 | 110 -->116 | VERY SHORT | IRREGULAR |
| 16 | 117 -->147 | MEDIUM | ALMOST REGULAR |
| 17 | 148 -->150 | VERY SHORT | REGULAR |

# MEMBRANE SHAPE DESCRIPTION

The following is a summary of the global shape of the cell membrane based on the GEOMETRIC MEAN of the following properties: AVERAGE BENDING ENERGY, CIRCULARITY, and ANGLE REGULARITY.

| PERIOD NUMBER | FRAME NUMBER | TIME | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 18 | SHORT | COMPLEX |
| 2 | 19 --> 20 | VERY SHORT | ALMOST SIMPLE |
| 3 | 21 --> 27 | VERY SHORT | COMPLEX |
| 4 | 28 --> 36 | SHORT | ALMOST SIMPLE |
| 5 | 37 --> 44 | SHORT | VERY COMPLEX |
| 6 | 45 --> 49 | VERY SHORT | COMPLEX |
| 7 | 50 --> 57 | SHORT | ALMOST SIMPLE |
| 8 | 58 --> 60 | VERY SHORT | SIMPLE |
| 9 | 61 --> 64 | VERY SHORT | VERY SIMPLE |
| 10 | 65 --> 70 | VERY SHORT | ALMOST SIMPLE |
| 11 | 71 --> 77 | VERY SHORT | SIMPLE |
| 12 | 78 --> 86 | SHORT | ALMOST SIMPLE |
| 13 | 87 --> 88 | VERY SHORT | SIMPLE |
| 14 | 89 -->113 | MEDIUM | ALMOST SIMPLE |
| 15 | 114 -->116 | VERY SHORT | COMPLEX |
| 16 | 117 -->145 | MEDIUM | ALMOST SIMPLE |
| 17 | 146 -->150 | VERY SHORT | SIMPLE |

# TABLE <7.1>

# GLOBAL SHAPE ANALYSIS

## ANALYSIS OF INDIVIDUAL SHAPE PROPERTY

## PROPERTY = CIRCULARITY

FIRST FRAME NUMBER = 1
LAST FRAME NUMBER = 150
TOTAL TIME = 75.0 Seconds
PERCENTAGE OF CURVE APPROXIMATION = 20.0 %
QUALIFICATION THRESHOLDS = .10, .30, .70, .90,

CURVE APPROXIMATION

NUMBER OF PERIODS =15

| PERIOD NUMBER | INITIAL FRAME No. | FINAL FRAME No. | INITIAL LEVEL | FINAL LEVEL | SLOPE(RATE OF CHANGE) |
|---|---|---|---|---|---|
| 1 | 1 | 21 | 1 | 1 | 3 |
| 2 | 21 | 31 | 1 | 3 | 5 |
| 3 | 31 | 40 | 3 | 1 | 1 |
| 4 | 40 | 50 | 1 | 3 | 5 |
| 5 | 50 | 52 | 3 | 3 | 1 |
| 6 | 52 | 54 | 3 | 4 | 5 |
| 7 | 54 | 57 | 4 | 4 | 1 |
| 8 | 57 | 58 | 4 | 5 | 5 |
| 9 | 58 | 77 | 5 | 5 | 2 |
| 10 | 77 | 80 | 5 | 3 | 1 |
| 11 | 80 | 88 | 3 | 3 | 4 |
| 12 | 88 | 139 | 3 | 2 | 2 |
| 13 | 139 | 145 | 2 | 3 | 4 |
| 14 | 145 | 146 | 3 | 4 | 5 |
| 15 | 146 | 150 | 4 | 5 | 4 |

Table (7.1) The output data for the curve approximation in Figure 7.3. This data are represented by a squence of key frames.

# TABLE <7.2>
# GLOBAL SHAPE ANALYSIS

NUMBER OF PERIODS = 43

| PERIOD NUMBER | INITIAL FRAME No. | FINAL FRAME No. | LEVEL |
|---|---|---|---|
| 1 | 1 | 21 | 1 |
| 2 | 21 | 22 | 1 |
| 3 | 23 | 23 | 2 |
| 4 | 24 | 25 | 1 |
| 5 | 26 | 30 | 2 |
| 6 | 31 | 31 | 3 |
| 7 | 31 | 31 | 3 |
| 8 | 32 | 34 | 2 |
| 9 | 35 | 35 | 3 |
| 10 | 36 | 39 | 2 |
| 11 | 40 | 40 | 1 |
| 12 | 40 | 40 | 1 |
| 13 | 41 | 43 | 2 |
| 14 | 44 | 44 | 3 |
| 15 | 45 | 45 | 2 |
| 16 | 46 | 50 | 3 |
| 17 | 50 | 52 | 3 |
| 18 | 52 | 53 | 3 |
| 19 | 54 | 54 | 4 |
| 20 | 54 | 57 | 4 |
| 21 | 57 | 57 | 4 |
| 22 | 58 | 58 | 5 |
| 23 | 58 | 77 | 5 |
| 24 | 77 | 77 | 5 |
| 25 | 78 | 78 | 4 |
| 26 | 79 | 80 | 3 |
| 27 | 80 | 88 | 3 |
| 29 | 88 | 129 | 3 |
| 30 | 131 | 131 | 3 |
| 31 | 132 | 135 | 2 |
| 32 | 136 | 136 | 3 |
| 33 | 137 | 137 | 2 |
| 34 | 138 | 138 | 3 |
| 35 | 139 | 139 | 2 |
| 36 | 139 | 139 | 2 |
| 37 | 140 | 145 | 3 |
| 38 | 145 | 145 | 3 |
| 39 | 146 | 146 | 4 |
| 40 | 146 | 147 | 4 |
| 41 | 148 | 148 | 5 |
| 42 | 149 | 149 | 4 |
| 43 | 150 | 150 | 5 |

Splitting the periods resulting from the curve approximations, given in Table#<7.1> into periods having the same level.

# TABLE <7.3>

# GLOBAL SHAPE ANALYSIS

NUMBER OF PERIODS = 29

| PERIOD NUMBER | INITIAL FRAME No. | FINAL FRAME No. | LEVEL |
|---|---|---|---|
| 1 | 1 | 22 | 1 |
| 2 | 23 | 23 | 2 |
| 3 | 24 | 25 | 1 |
| 4 | 26 | 30 | 2 |
| 5 | 31 | 31 | 3 |
| 6 | 32 | 34 | 2 |
| 7 | 35 | 35 | 3 |
| 8 | 36 | 39 | 2 |
| 9 | 40 | 40 | 1 |
| 10 | 41 | 43 | 2 |
| 11 | 44 | 44 | 3 |
| 12 | 45 | 45 | 2 |
| 13 | 46 | 53 | 3 |
| 14 | 54 | 57 | 4 |
| 15 | 58 | 77 | 5 |
| 16 | 78 | 78 | 4 |
| 17 | 79 | 129 | 3 |
| 18 | 130 | 130 | 2 |
| 19 | 131 | 131 | 3 |
| 20 | 132 | 135 | 2 |
| 21 | 136 | 136 | 3 |
| 22 | 137 | 137 | 2 |
| 23 | 138 | 138 | 3 |
| 24 | 139 | 139 | 2 |
| 25 | 140 | 145 | 3 |
| 26 | 146 | 147 | 4 |
| 27 | 148 | 148 | 5 |
| 28 | 149 | 149 | 4 |
| 29 | 150 | 150 | 5 |

Table 7.3 The result of merging the periods given in Table 7.2

## TABLE <7.4>

## GLOBAL SHAPE ANALYSIS

NUMBER OF PERIODS = 10

| PERIOD NUMBER | INITIAL FRAME NO. | FINAL FRAME NO. | LEVEL |
|---|---|---|---|
| 1 | 1 | 25 | 1 |
| 2 | 26 | 43 | 2 |
| 3 | 44 | 53 | 3 |
| 4 | 54 | 57 | 4 |
| 5 | 58 | 78 | 5 |
| 6 | 79 | 131 | 3 |
| 7 | 132 | 137 | 2 |
| 8 | 138 | 145 | 3 |
| 9 | 146 | 149 | 4 |
| 10 | 150 | 150 | 5 |

Table 7.4 The final result of merging the periods given in Table 7.3 by using Rules 7.3 and 7.4.

# TABLE <7.5A>

## SHAPE ANALYSIS

| FIGURE NUMBER | ANGLE REGULARITY | AVERAGE BENDING ENERGY | ARITHMETIC MEAN | GEOMETRIC MEAN |
|---|---|---|---|---|
| 1 | 0.0000 | 0.0065 | 0.2073 | 0.0011 |
| 2 | 0.4167 | 0.0074 | 0.3464 | 0.1237 |
| 3 | 0.5000 | 0.0133 | 0.3762 | 0.1600 |
| 4 | 0.5250 | 0.0219 | 0.3874 | 0.1921 |
| 5 | 0.7669 | 0.0212 | 0.4678 | 0.2156 |
| 6 | 0.5333 | 0.0321 | 0.3936 | 0.2191 |
| 7 | 0.5024 | 0.0099 | 0.3759 | 0.1454 |
| 8 | 0.5333 | 0.0650 | 0.4046 | 0.2774 |
| 9 | 0.5000 | 0.0125 | 0.3759 | 0.1568 |
| 10 | 0.5357 | 0.0304 | 0.3938 | 0.2156 |
| 11 | 0.6366 | 0.0244 | 0.4254 | 0.2122 |
| 12 | 0.5988 | 0.0246 | 0.4129 | 0.2085 |
| 13 | 0.4621 | 0.0843 | 0.3872 | 0.2883 |
| 14 | 0.4672 | 0.1210 | 0.4012 | 0.3264 |
| 15 | 0.5333 | 0.0289 | 0.3925 | 0.2117 |
| 16 | 0.5201 | 0.0208 | 0.3883 | 0.1889 |
| 17 | 0.5556 | 0.0133 | 0.3947 | 0.1657 |
| 18 | 0.5357 | 0.5928 | 0.5813 | 0.5803 |
| 19 | 0.4167 | 0.0957 | 0.3757 | 0.2906 |
| 20 | 0.6286 | 0.0074 | 0.4171 | 0.1418 |

Table(7.5a) The computed values of two different shape properties, and their arithmetic and geometric means for the different shapes given in Figure (7.8).

# TABLE <7.5B>

## SHAPE ANALYSIS

| ANGLE REGULARITY | | AVERAGE BENDING ENERGY | | ARTHIMETIC MEAN | | GEOMETRIC MEAN | |
|---|---|---|---|---|---|---|---|
| NO. | VALUE | NO. | VALUE | NO. | VALUE | NO. | VALUE |
| 1 | 0.0000 | 1 | 0.0065 | 1 | 0.2073 | 1 | 0.0011 |
| 19 | 0.4167 | 20 | 0.0074 | 2 | 0.3464 | 2 | 0.1237 |
| 2 | 0.4167 | 2 | 0.0074 | 7 | 0.3759 | 20 | 0.1418 |
| 13 | 0.4621 | 7 | 0.0099 | 19 | 0.3759 | 7 | 0.1454 |
| 14 | 0.4672 | 9 | 0.0125 | 9 | 0.3759 | 9 | 0.1568 |
| 9 | 0.5000 | 3 | 0.0133 | 3 | 0.3762 | 3 | 0.1600 |
| 3 | 0.5000 | 17 | 0.0133 | 13 | 0.3872 | 17 | 0.1657 |
| 7 | 0.5024 | 16 | 0.0208 | 4 | 0.3874 | 16 | 0.1889 |
| 16 | 0.5201 | 5 | 0.0212 | 16 | 0.3883 | 4 | 0.1921 |
| 4 | 0.5250 | 4 | 0.0219 | 15 | 0.3925 | 12 | 0.2058 |
| 8 | 0.5333 | 11 | 0.0244 | 6 | 0.3936 | 15 | 0.2117 |
| 6 | 0.5333 | 12 | 0.0246 | 10 | 0.3938 | 11 | 0.2122 |
| 15 | 0.5333 | 15 | 0.0289 | 17 | 0.3947 | 5 | 0.2156 |
| 10 | 0.5357 | 10 | 0.0304 | 14 | 0.4012 | 10 | 0.2156 |
| 18 | 0.5357 | 6 | 0.0321 | 8 | 0.4046 | 6 | 0.2191 |
| 17 | 0.5556 | 8 | 0.0650 | 12 | 0.4129 | 8 | 0.2774 |
| 12 | 0.5988 | 13 | 0.0843 | 20 | 0.4171 | 13 | 0.2883 |
| 20 | 0.6286 | 19 | 0.0957 | 11 | 0.4254 | 19 | 0.2906 |
| 11 | 0.6366 | 14 | 0.1210 | 5 | 0.4678 | 14 | 0.3264 |
| 5 | 0.7669 | 18 | 0.5928 | 18 | 0.5813 | 18 | 0.5803 |
| Figure(7.10a) | | Figure(7.10a) | | Figure(7.10a) | | Figure(7.10a) | |

Table(7.5b) Sorting different shapes (given in Figure 7.8) according to their complexity as measured by the average bending energy, angle regularity, and the arithemetic and geometric means of both.

# CHAPTER 8

# GLOBAL STRUCTURAL ANALYSIS AND DESCRIPTION OF DYNAMIC BEHAVIOUR

## 8.1 INTRODUCTION

Most of the work in dynamic scene analysis has considered change in location as the major aspect in understanding the dynamic behaviour of a moving object. Recently, some, albeit few, efforts have considered the change in shape of a rigid moving object due to change in the viewing conditions (see Section 2.2.3). In the present research, we designed a system for understanding and describing the dynamic behaviour of a non-rigid moving object. The major difference from the previous work, is that the changes in shape are due to changes in the morphology (spatial structure) of the primitive components of the object. Thus, we analyze, quantify, and describe the structural changes of a non-rigid moving object, hitherto neglected in all the previous work done in image sequence analysis.

The dynamic behaviour of a non-rigid moving object can be described in terms of changes in locomotion, shape, and structure. The locomotion analysis is described in Chapter 6, and the global shape changes are discussed in Chapter 7. Two main issues will be addressed in this chapter, global structural analysis, and characterization of global dynamic behaviour. The objective of the first issue is to analyze the static and incremental structural descriptions in order to generate a summary of the global structural changes. The second part of this chapter is concerned with the integration of the three global aspects pertaining to locomotion, shape, and structure, in order to understand and characterize the dynamic behaviour of the moving object.

## 8.2 GLOBAL STRUCTURAL ANALYSIS

The main goal of analyzing the structural changes in a moving cell is to characterize and describe the dynamic behaviour of the different pseudopods that are formed during cell locomotion. Also, we need to study the relationships between the changes in the shape of subparts, their movement, and the global locomotion of the cell. For example, in the study of the dynamic behaviour of a moving cell, a developmental biologist might be interested in the answers to the following three basic questions. First, how can one recognize a subpart developed on or by the membrane as a "pseudopod"? And if it is a pseudopod, is it

stationary, growing, or contracting?. Second, is a pseudopod "dominant" or not? A dominant pseudopod is one which leads the locomotion of the cell. If so, what is its degree of domination? Finally, if all of this information is at hand, the third question would be the interpretation of this data. Why is a specific pseudopod dominant and another not?

Our approach to the analysis of the structural changes is based on the same philosophy as syntactic pattern recognition techniques. These analyze patterns by a parsing process of hierarchical decomposition. Such techniques appear to be quite amenable as a strategy for cell shape description, including the global structural and membrane shape changes (see Section 2.3.10).

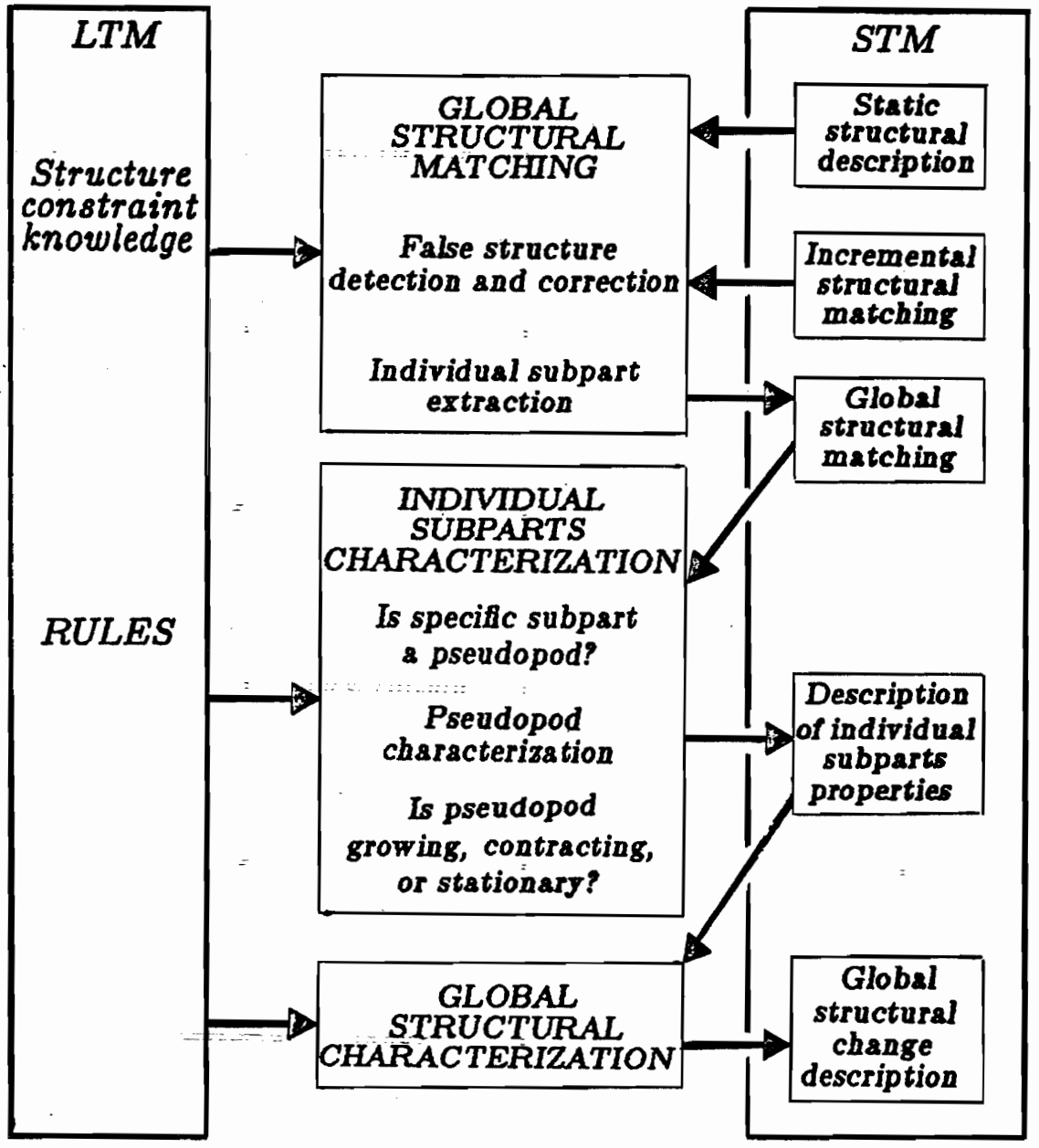
Quantification and characterization of the structural changes of a moving cell can be accomplished in three stages of analysis: static, incremental, and global. In static analysis, the cell is decomposed into its primitive subparts. The latter can be represented as a labeled graph that conveys the topological properties of the cell. This stage of analysis is described in Chapter 4. The incremental analysis is responsible for detecting and quantifying the structural changes between two sequential frames. The processes that accomplish this are discussed in Chapter 5. In these processes, the labeled graphs in both frames are matched in order to detect the correspondence between the different subparts. Then, the incremental

changes in the corresponding subparts are quantified and described. Cell structure can be matched in two frames, and described on a scale beginning with VERY DIFFERENT and ending with VERY SIMILAR. Global structural analysis is the final stage which will be discussed below.

The objective of global structural analysis is to generate a summary of the significant structural changes, from which questions pertaining to the subpart and/or pseudopod characteristics can be answered. The processes and data structure of global structural analysis are shown in Figure (8.1). In these processes, we first detect the frames where false structure has occurred. This is accomplished by an analysis of the incremental structural changes. These use representational rules that incorporate both dynamic data and constraint knowledge pertaining to the cell under consideration. Most of the cases of false structure are corrected by feedback to the low level decomposition process. Then, observable changes in the properties of each subpart are analyzed and described. The final step in this analysis is to generate a description of the significant changes in the structure of the cell and its primitive subparts.

## 8.2.1 GLOBAL STRUCTURAL MATCHING

Two main objectives are cited: (a) To analyze the static structure data and the incremental structural matching data in order to detect and correct (if possible)




*Figure(8.1) Global structural analysis, processes and data structure.*

false structure resulting from the low level decomposition. (b) To find the sequence of frames in which individual subparts appeared. These two issues will be discussed in the following two sections.

### 8.2.1.1 Detection And Correction Of False Structure

Errors in the low level decomposition can produce false structures. This is mainly caused by artifactual shape changes due to registeration noise, irrelevant changes, and/or the three-dimensionality of the cell. These artifactual situations cannot be detected by the static analysis stage. However, at the incremental analysis stage, false structures can be interpreted as ambiguous situations. This can be detected through a low value of the cell structural match between two sequential frames. On the other hand, if a false decomposition appears in several sequential frames, it cannot be detected from the incremental structural matching.

Our approach to detecting the cases of false structure uses representational rules that utilize both the dynamic data and constraint knowledge pertaining to the structure of the cell under consideration. For example, if the structural match SM(i-1, i) (see equations (5.12) and (5.15)) between two sequential frames (i-1) and (i) is less than a specific threshold value Em, this indicates either significant structural change or a false decomposition at frame (i). This ambiguous situation can be encountered by

the incremental analysis at frames (i) and (i-1), by using the following rule:

RULE(8.1): IF SM(i-1,i) .LT. Em

==then==> SIGNIFICANT STRUCTURAL CHANGE

.OR. FALSE DECOMPOSITION

However, this can be clarified at the global level by the follwing rule:

RULE(8.2): IF SM(i-1,i) .AND. SM(i,i+1) .LT. Em

==then==> FALSE DECOMPOSITION AT FRAME(i)

where SM(i-1,i), SM(i,i+1) are the structural matches between frames (i-1),(i) and (i),(i+1) respectively. These rules are based on the cell structural data which are derived from frames (i-1), (i), (i+1), and the constraint knowledge pertaining to the cell dynamic behaviour. For example, a pseudopod or subpart of the cell membrane cannot be formed and deformed in three sequential frames (1.5 seconds in this particular application).

The threshold value Em in the above rule specifies the acceptable structural match SM. The latter is computed based on two factors: the correspondence between the different subparts in both frames and the changes in their properties (see equation 5.15). With regard to the former, it seen from equation (5.15) that the value of SM is bounded by:

$$0 < SM(i-1,i) < [\, K(i-1,i) / NSP(i) \,], \qquad (8.1)$$

where K(i-1,i) is the number of the corresponding subparts between frames (i) and (i-1), and NSP(i) is the number of

subparts in frame (i). For the latter, The change in the value of SM depends on the changes in the properties of the corresponding subparts. The effect of these changes on the value of SM is less than that caused by the change in the number of corresponding subparts. A lower bound on the value of SM has been found experimentally to be governed by:

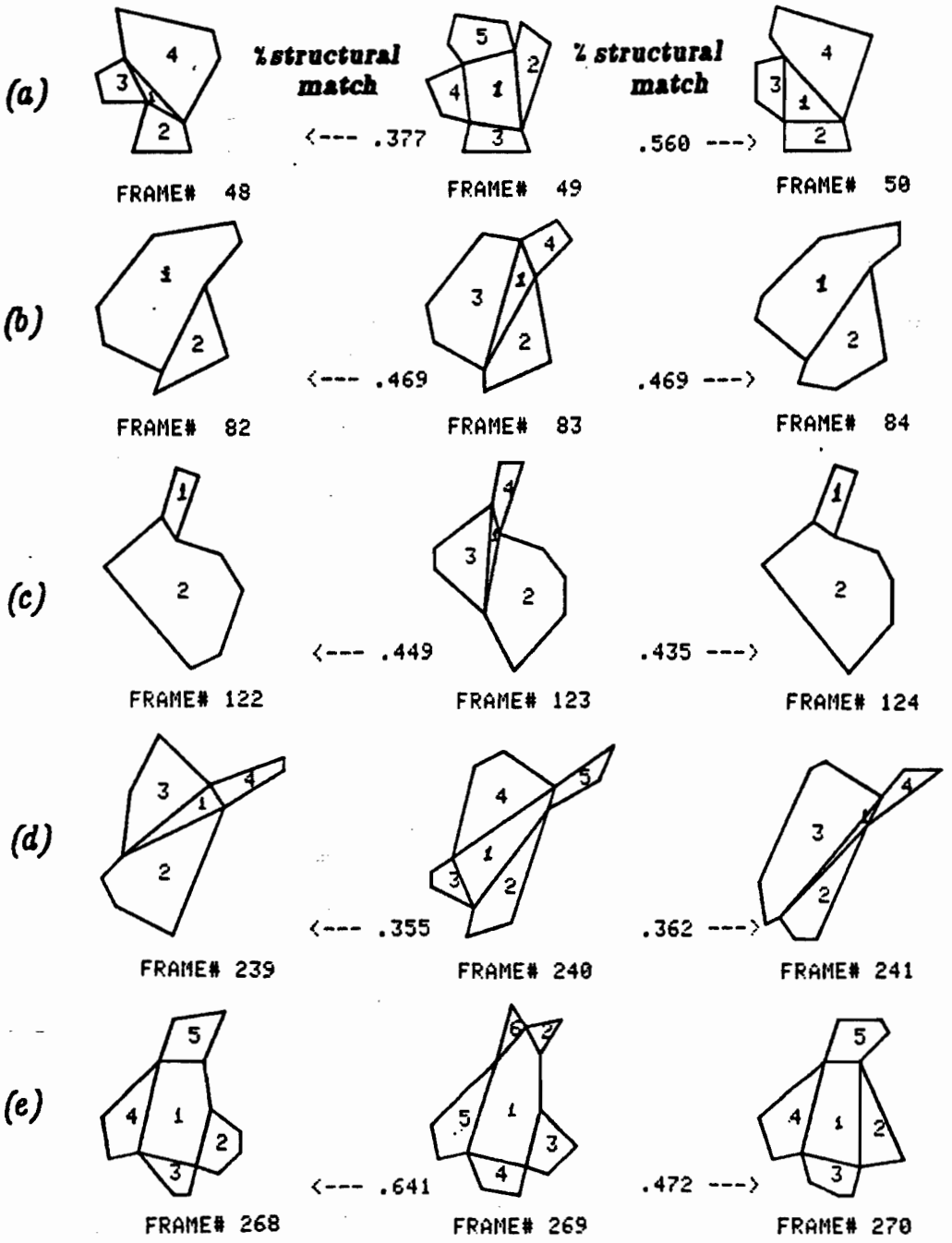
$$SM(i-1,i) > [K(i-1,i) - 1] / NSP(i) \qquad (8.2)$$

From this inequality, we can compute the threshold value of the minimum acceptable structural matching as:

$$Em = [K(i-1,i) - 1] / NSP(i) \qquad (8.3)$$

Using this value in Rule (8.2), the cases of false structure can be detected. Figure (8.2) shows examples of sequential frames where false decompositions have been detected. In this figure, the structural matching of the cell in the false frames, with the previous and the successive ones is indicated. Using the bound defined by equation (8.3) yielded satisfactory distinctions between false and appropriate decompositions using Rules (8.1) and (8.2).

When a false structure is detected, the attention of the system will be directed to the frame where the fault has occurred. More information about the cell structure and its match with the previous and successive ones is extracted. For example, information such as OVERDECOMPOSITION or UNDERDECOMPOSITION can be extracted from the global analysis, and utilized by the low level decomposition processes to correct the false decomposition. Examples of the rules that accomplish this objective are:




***Figure(8.2) Examples of frames having false structure (decomposition) detected at the global structural analysis stage.***

***Note that in each example the middle frames, where the false structure appears, is associated with a low value of structural match compared with both the previous and successive frames.***

RULE(8.3): IF FALSE DECOMPOSITION AT FRAME(i)
. AND. NSP(i-1) .EQ. NSP(i+1) .LT. NSP(i)
==then==> OVERDECOMPOSITION AT FRAME(i)

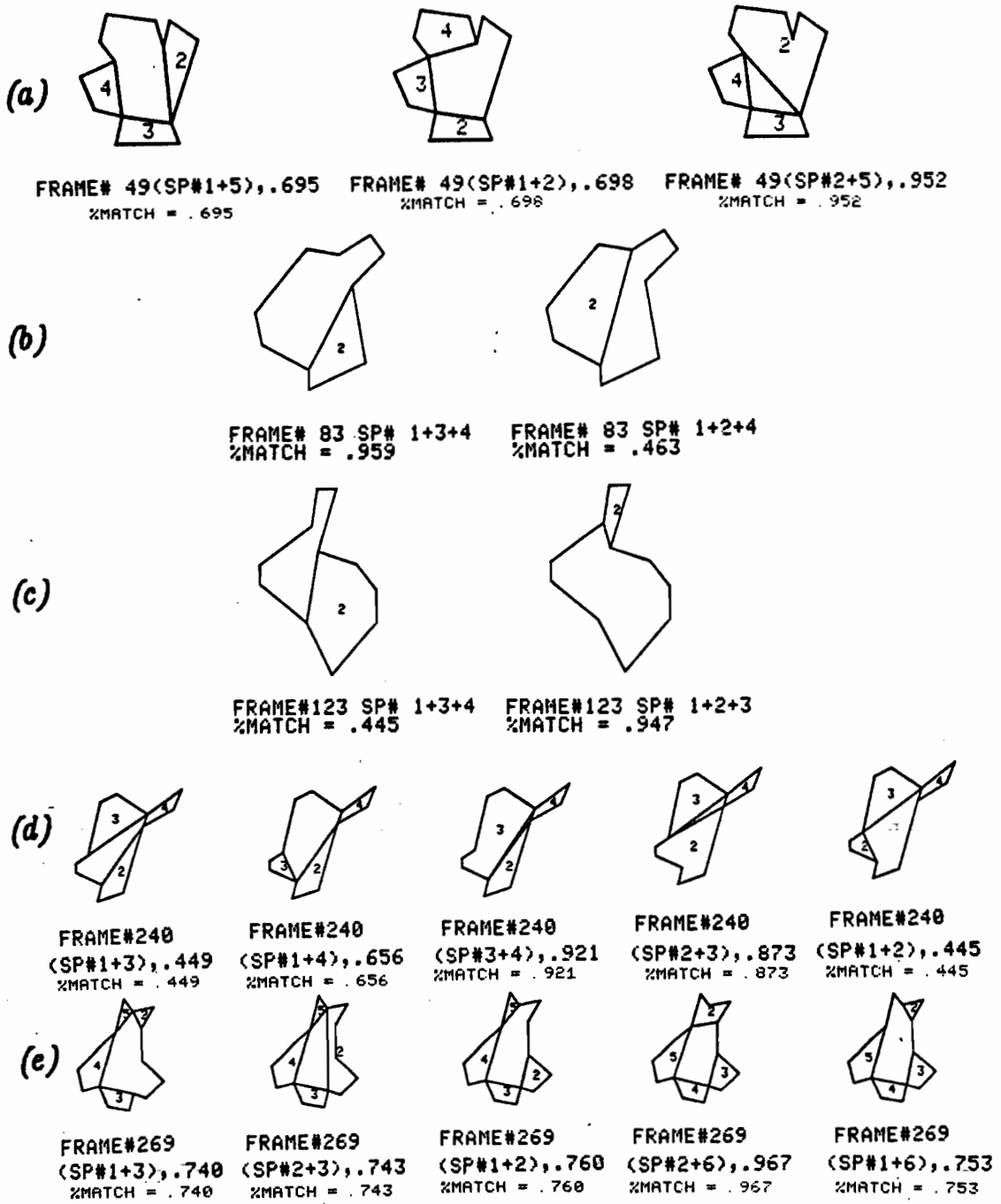
RULE(8.4): IF FALSE DECOMPOSITION AT FRAME(i)
. AND. NSP(i-1) .EQ. NSP(i+1) .GT. NSP(i)
==then==> UNDERDECOMPOSITION AT FRAME(i)

where NSP(i-1), NSP(i), NSP(i+1) are the number of subparts at frames i-1, i, and i+1, respectively. For example, in Figure (8.2) an overdecomposition is detected in each of the middle frames.

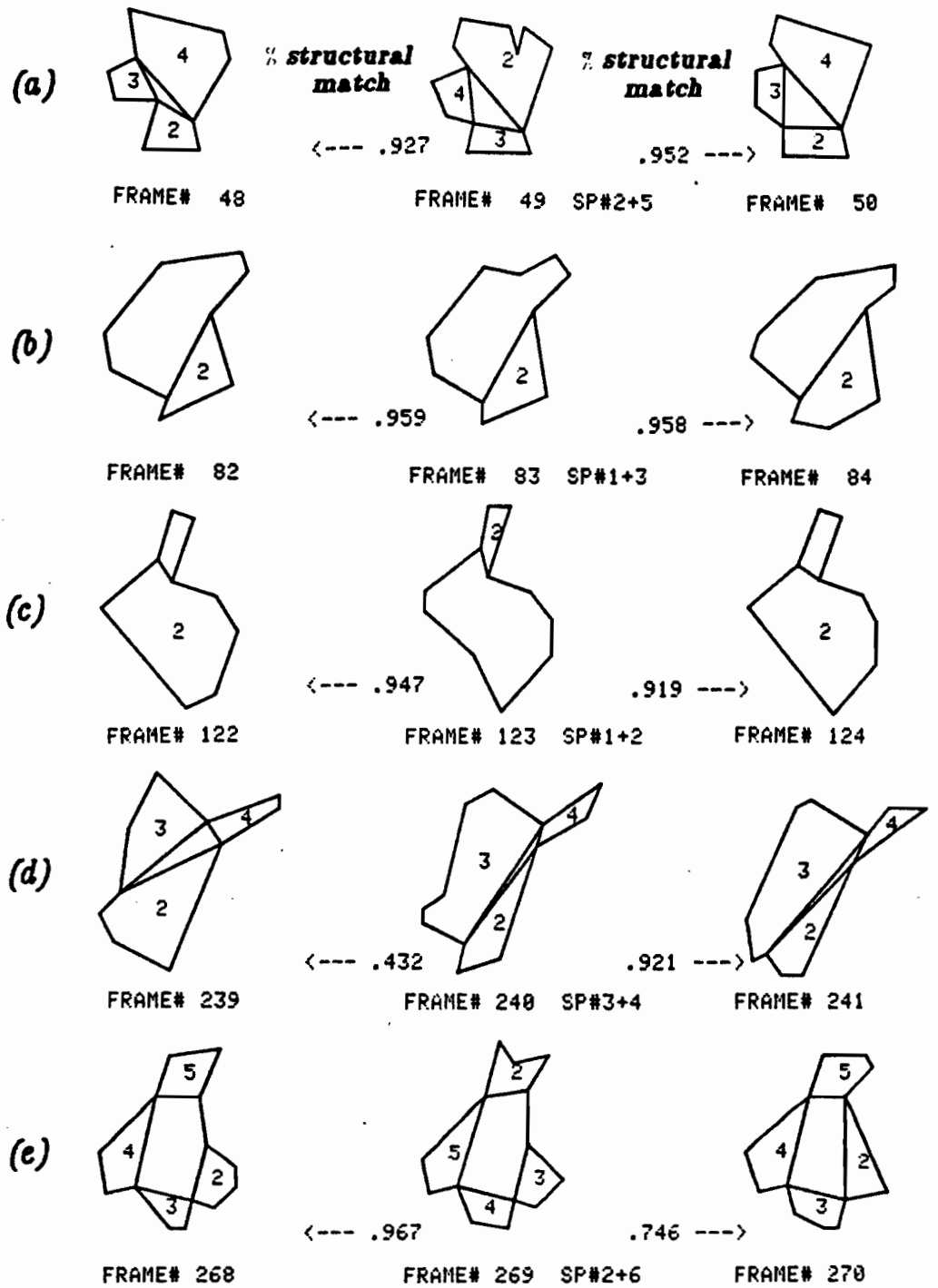
Using this type of feedback data, specific rules are activated in order to modify the result of the original low level analysis. For example, in the case of the overdecomposition, the following rules will be activated:

RULE(8.5):
IF OVERDECOMPOSITION AT FRAME(i)
==then==> 1) MERGE SPj+SPk j=1,2,...,NSP(i-1),
k=j+1,j+2,...,NSPi
2) FOR EACH MERGE, MEASURE SM(i-1,i) AND SM(i,i+1)
3) SELECT THE MERGE THAT RESULTS IN
MAX. OF SM(i-1,i) .OR. SM(i,i+1).

Figure (8.3) shows the result of activating Rule 8.5 on the cases shown in Figure (8.2). The corrected structure and the resulting modified structural matches are shown in Figure (8.4). We may note in Figure (8.2d) that a false structural change has occurred between frames 240 and 241;




***Figure(8.3) Possible variations in the decompositions of the cells having false structure in Figure(8.2), (see Rule 8.5).***




*Figure(8.4) Correcting the false structure of the examples shown in Figure(8.2) using Rule 8.5.*

***Note that in case (d), the structural match has only been corrected between frames 240 and 241. There still remains a significant structural discrepancy between frames 239 and 240. Thus, subpart 3 in frame 239 has merged with the cell body, and in frame 240, the new subpart 2 appears.***

a significant change in structure has occurred between frames 239 and 240.

Using the above rule, most cases of false structure can indeed be corrected. However, in some situations it may fail, especially if the false decomposition is due to noise or artifactual changes. In this case, another rule should be activated in order to describe the situation and action taken. For example:

RULE(8.6):

IF THE ACTION SPECIFIED BY RULE(8.5) DOES NOT IMPROVE THE STRUCTURAL MATCHING BETWEEN FRAMES (i) AND (i-1) .OR. (i) AND (i+1)

==then==> (1) MEASURE SM(i-1,i+1)

(2) ACTIVATE RULE(8.7)

RULE(8.7) IF SM(i-1,i+1) .GT. SM(i-1,i) .OR. SM(i,i+1)

==then==> DELETE FRAME(i) FORM THE SEQUENCE

where SM(i-1,i+1) is the structural match between frames i-1 and i+1.

The result of activating the above set of rules can be one of the following:

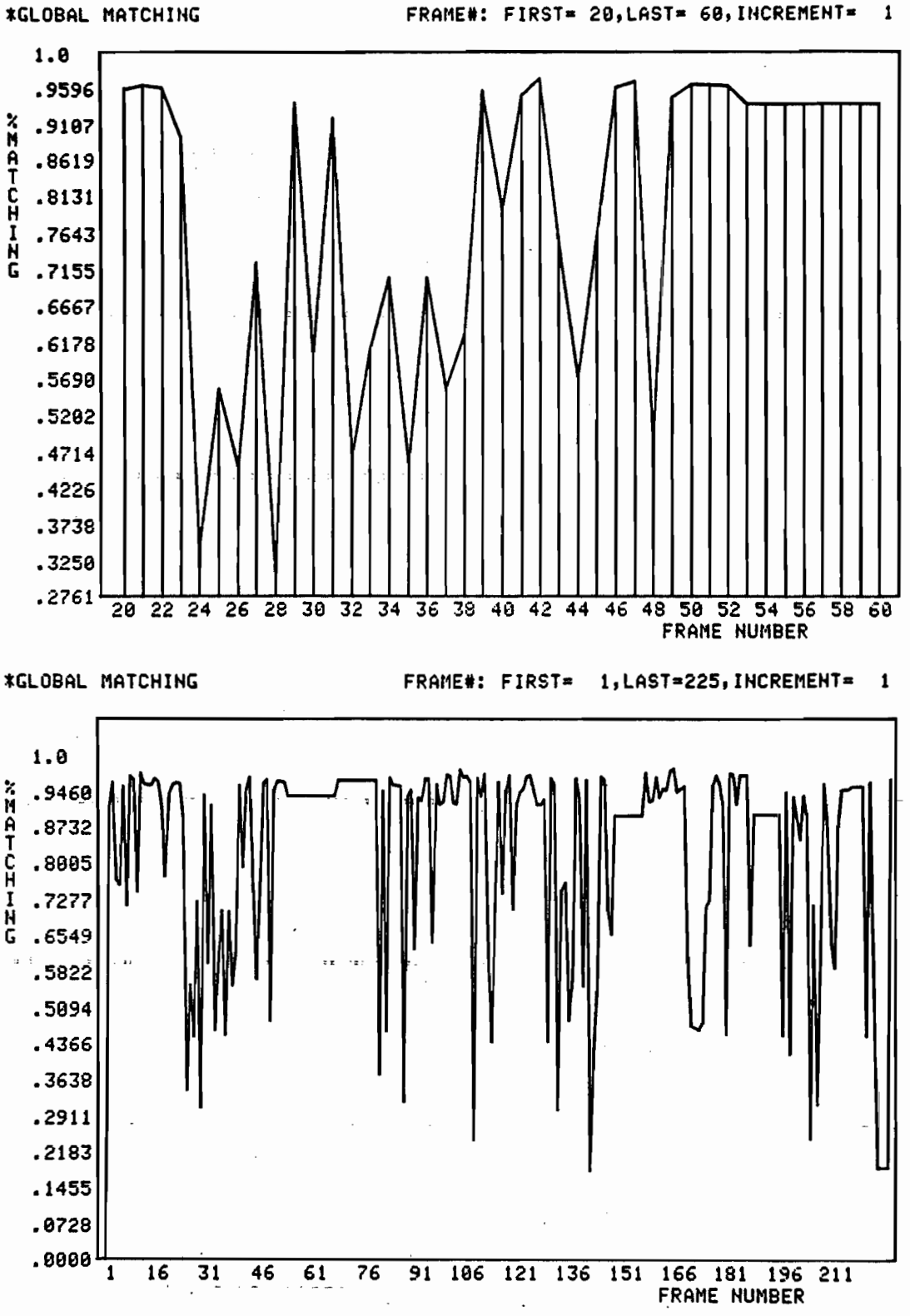
(a) The structure of frame(i) is corrected and the matching is improved; this is the most common case. Examples of this case are shown in Figure (8.2), and the result of correcting them is shown in Figure (8.4).

(b) The false decomposition of frame(i) cannot be corrected,

but frames (i-1) and (i+1) exhibit an acceptable structural match. In this case, frame(i) is considered as noisy data and deleted from the sequence. We may mention here, that deleting some frames from the sequence does not affect the global result. On the contrary, deleting the frames that include noisy or irrelevant information improves the global result.

(c) The structure of frame(i) cannot be corrected, and the decomposition in frames (i-1) and (i+1) do not match. This case is not very common. However, if it does occur, it will cause noise which can be removed by additional rules to be described below.

The above example demonstrates the detection of a false decomposition that has occurred in a single frame. In order to detect false decompositions in a sequence of frames, a global analysis strategy can be used. In this case, first, curve approximation and analysis is used in a fashion similar to that described in Chapter 7 for global shape analysis. The objective of the curve approximation here is to detect the points (key frames) where significant structural changes have occurred. Thus, the structural matching between the sequential frames (SM) can be represented by a curve, as shown in Figure (8.5). The amplitude of the curve at any point (i) represents the structural matching between two sequential frames SM(i-1, i). Therefore, the variations in neighbouring points on the curve indicate the structural changes of the cell. A global




*Figure(8.5) Curves representing the structural matching of the cell between each pair of sequential frames.*
*(a) 60 frames in sequence,*
*(b) 225 frames in sequence*

structural change is defined as a point where a subpart has apppeared or disappeared. The output of the curve approximation is a set of key frames. Then, a further analysis (splitting ang merging) of the periods between the key frames, will result in a summary of the structural changes. This analysis is similar to that described in Chapter 7 for the global shape analysis.

The output of the global structural matching is a set of key frames. Each key frame represents the time where a subpart has appeared or disappeared. This is utilized to detect and characterize the global structural changes of the cell. However, in this particular application, we are also interested in characterizing the dynamic behaviour of the different subparts. Therefore, each subpart is extracted and its properties analyzed as follows.

### 8.2.1.2 Extraction Of Individual Subparts

In order to characterize the dynamic behaviour of the different subparts of the cell, first, we initiate a record for each individual subpart. This record includes the following information: (a) the initial and final frame numbers (time of appearing and disappearing), (b) the subpart label in each frame of the sequence where it has appeared, and (c) its duration. The latter is normalized and described in a fashion similar to that used for other variables, i.e., as VERY SHORT, SHORT, MEDIUM, LONG, or VERY LONG. The number of subparts classified by each of these

descriptors can be utilized as one of the parameters that characterize the overall cell dynamic behaviour.

The second step involves the analysis of the different properties of each subpart, in order to characterize it as: (a) irrelevant change or noise, (b) part of or the cell body, and (c) pseudopod. However, we are specifically interested in those subparts which may be characterized as pseudopods. Therefore, we only analyze those that seem to be good candidates for pseudopods. The selection is based on an initial guess utilizing expert knowledge. For example, the subparts that appeared for MEDIUM, LONG, or VERY LONG period are selected. Also, those which started after the cell has a VERY SIMPLE shape (one part) are likely candidates.. In fact, these are the criteria which we are currently using. This is because, at this stage of research, our knowledge about the pseudopod characteristics is limited. However, using the present system, we expect to gain more knowledge pertaining to these characteristics. Hence, more sophisticated rules could be developed to improve the selection of the pseudopod candidates.

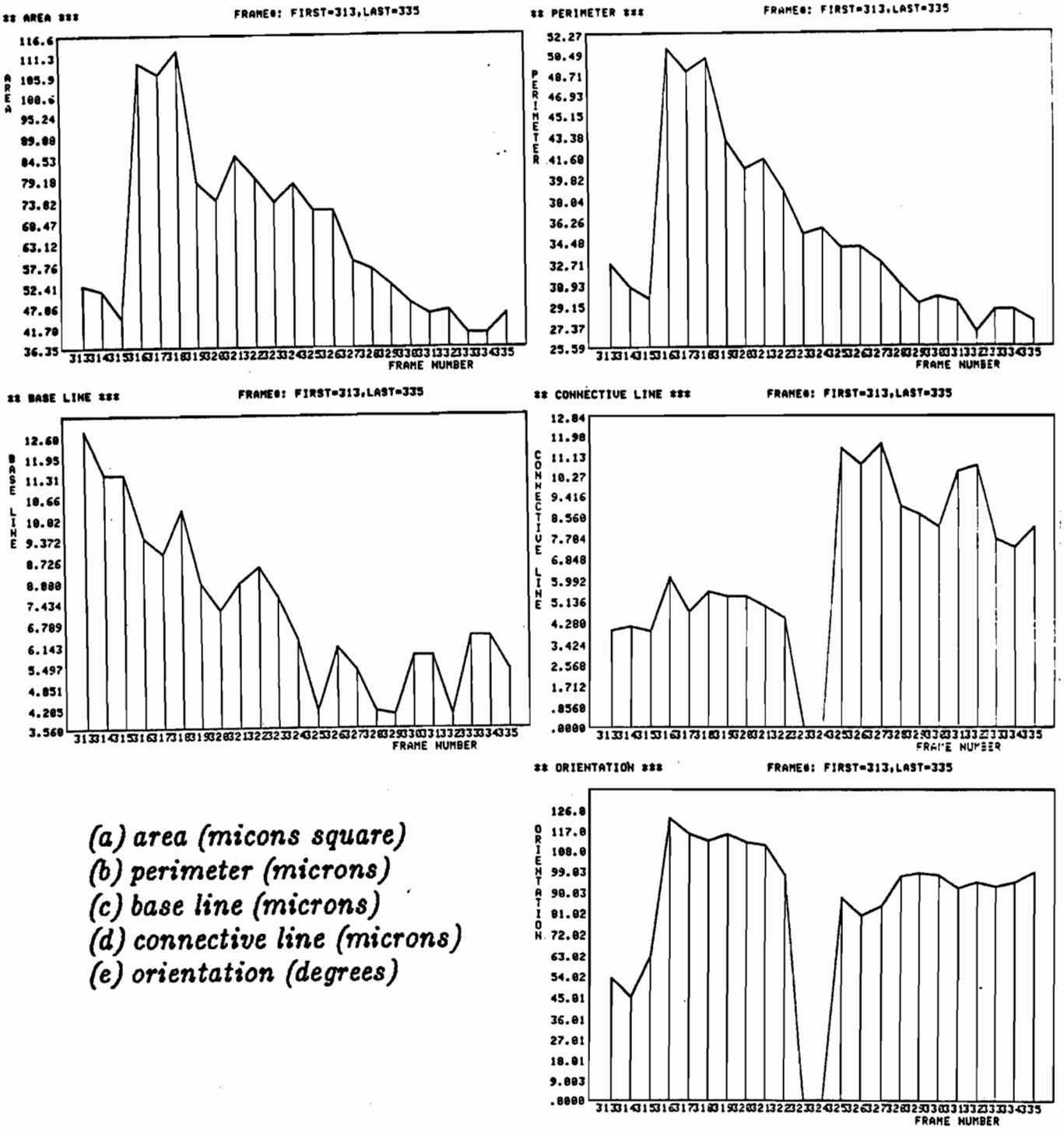
Analysis and characterization of the subparts will be described in the following section.

## 8.2.2 Individual Subpart Characterization

The main goal of characterizing the individual subparts of the cell is to know whether a specific subpart is a pseudopod or not. In order to accomplish this, each subpart is treated individually as a moving object. The properties pertaining to the location, shape, and the topological structure of each subpart are analyzed using similar techniques to those described for the entire cell in previous chapters.

The main properties that are available for subpart characterization are given in Table 4.1. However, the actual properties which are employed depend on the purpose of the analysis. For example, in the present system, we have used the area, base line, connective line, perimeter, and orientation (the angle between the connective line and the X axis) in order to characterize a subpart as either the cell body or a pseudopod.

Changes in the values of a property of a specific subpart under analysis can be reprsented by a curve, as shown in Figure (8.6). This curve is analyzed using the methodology described in Chapter 7 for global shape analysis. This analysis consists of two main steps. First, a curve approximation is used in order to detect the key frames where the significant changes occurred. This can be utilized to describe the global changes in the dynamic behaviour of the property under analysis as increasing,




*(a) area (micons square)*
*(b) perimeter (microns)*
*(c) base line (microns)*
*(d) connective line (microns)*
*(e) orientation (degrees)*

*Figure(8.6) Curves representing the changes in subpart properties.*

decreasing, or stationary (see Section 7.2.1). Then a further analysis, that includes splitting and merging the periods between the key frames, is used in order to generate the final description of the changes in the property (VERY SMALL, SMALL, MEDIUM, LARGE, OR VERY LARGE) (see Section 7.2.2).

The output is summarized in a number of periods. Each period is bounded by key frames, where the dynamic behaviour of the property between sequential key frames is constant (increasing, decreasing, or stationary). The amount of change in each period is expressed in terms of numerical and symbolical qualifiers. The former represents changes in the number of qualification levels (+ or - 0,1,2,3,4). The latter gives the change in the symbolic qualifier as MEDIUM ---> LARGE or SHORT ---> VERY SHORT. A typical example of a summary of the global changes in the properties of a specific subpart is given in Description (8.1a), and the result of analyzing a specific property is given in more detail in Description (8.1b). This data is utilized in order to determine whether a specific subpart is a pseudopod or not, as described in the next section.

### 8.2.2.1 Is A Subpart A Pseudopod?

A pseudopod is a protrusion forming around the cell membrane. The study of the pseudopod characteristics is essential in understanding the role that the membrane plays in regulating the social behaviour of the cell. In order to

characterize a subpart as a pseudopod, we use production representational rules that utilize the dynamic data associated with it, as well as constraint knowledge pertaining to pseudopods in gemeral. The function of these rules is to compare the description of specific subpart properties to the pseudopod constraint knowledge. For example, a pseudopod has an area that should not exceed half of the cell area, has elongated shape, concave corners, short base line, and its main axis is almost normal to the cell body. However, these properties change gradually through the growing and contracting of the pseudopod. For example, a pseudopod starts with a relatively long base line, very small area, and main axis parallel to the cell body. Thus, we cannot use the properties of a specific subpart in a static frame to decide whether it is a pseudopod or not. We have used the normalized values of the base line and relative area to characterize a subpart as a pseudopod or part of the cell body, by using the following rule:

RULE(8.8):

```
      IF     BASE LINE (SHORT .OR. VERY SHORT)
             .AND. AREA (SMALL .OR. VERY SMALL)
  ==then==> SUBPART <--- PSEUDOPOD

      IF     BASE LINE (LONG .OR. VERY LONG)
             .AND. AREA (LARGE .OR. VERY LARGE)
  ==then==> SUBPART <--- CELL BODY
```

In the above rule, the symbolic qualification of two of the subpart properties is used to characterize it as a pseudopod or the cell body. However, in many cases this decision is not clear; in such cases more properties should be used, such as elongation and regularity.

In the present analysis, a subpart is characterized by a percentage value PCB indicating the confidence of its interpretation as a pseudopod or cell body. This value is computed as:

$$PRA(j,i) = PA(j,i) / A(i) \qquad (8.4)$$

$$PRB(j,i) = BL(j,i) / PP(j,i) \qquad (8.5)$$

where A(i) is the cell area in frame (i), and PA(j,i), BL(j,i), PP(j,i), PRA(j,i) and PRB(j,i) are the area, base line, perimeter, relative area and relative baseline of pseudopod (j) in frame (i), respectively. Using the values of PRA and PRB, the value of PCB can be computed as:

$$PCB(i) = [\, PRA(i) \cdot (1-PRB(i)) \,]^{1/2} \qquad (8.6)$$

Then, using global analysis of the curve representing these values, we obtained the global characterization of the subpart. An example of this characterization is given in Description (8.2a).

### 8.2.2.2 Pseudopod Description

In the preceding section, we described how to characterize a subpart of the cell as pseudopod or cell body. In this section, we will discuss the changes in the morphology of a pseudopod in order to understand and describe its dynamic behaviour. In this way, questions pertaining to the pseudopod kinetics can be answered. For example, in this particular application, we are interested in whether a pseudopod is growing, contracting, or stationary?.

Suppose a specific subpart is characterized as a pseudopod during the period Tij (ti and tj are the initial and final frame numbers of the pseudopod period). Then the different properties which describe the changes in location, shape and structural relationships during this period are analyzed to generate a description of its dynamic behaviour.

At this stage of study, expert knowledge about growing, contracting, or stationary pseudopods is limited to a definition based on the change in its size. However, in many cases, although a pseudopod may be stationary, the size of the entire cell may change, which consequently alters the pseudopod size. The effect of this can be avoided by using as a feature the change in the relative area of the pseudopod compared to the entire cell. Using this parameter and the change in the absolute area of the pseudopod, the change in the pseudopod size can be quantified as a

percentage value (PCA), such that:

$$PCA = \frac{PA(i) - PA(i-1)}{A(i-1) - A(i)} \qquad (8.7)$$

where PA(i-1), PA(i) and A(i-1), A(i) are the areas of the pseudopod and cell in frames (i-1) and (i), respectively. This value is used by the following rule in order to characterize a pseudopod as GROWING, CONTRACTING, or STATIONARY:

RULE(8.9):

IF PCA .LT. E1 ==then==> CONTRACTING

IF PCA .GE. E1 .AND .LT. E2 ==then==> STATIONARY

IF PCA .GE. E2 ==then==> GROWING

Description (8.2) is a typical example of this characterization. It represents the incremental description of the pseudopod characterization. By analyzing this description, a summary of the pseudopod characterization is obtained.

## 8.3 DYNAMIC BEHAVIOUR DESCRIPTION

The observable changes in the three basic aspects of the dynamics of a non-rigid moving object (locomotion, shape, and structure) have been studied individually. However, in most cases, the change in any of these aspects is related to change in the others. For example, the shape and structure of an object are functions of each other. Therefore, in order to understand the dynamic behaviour of a non-rigid moving object, it is not enough to study the

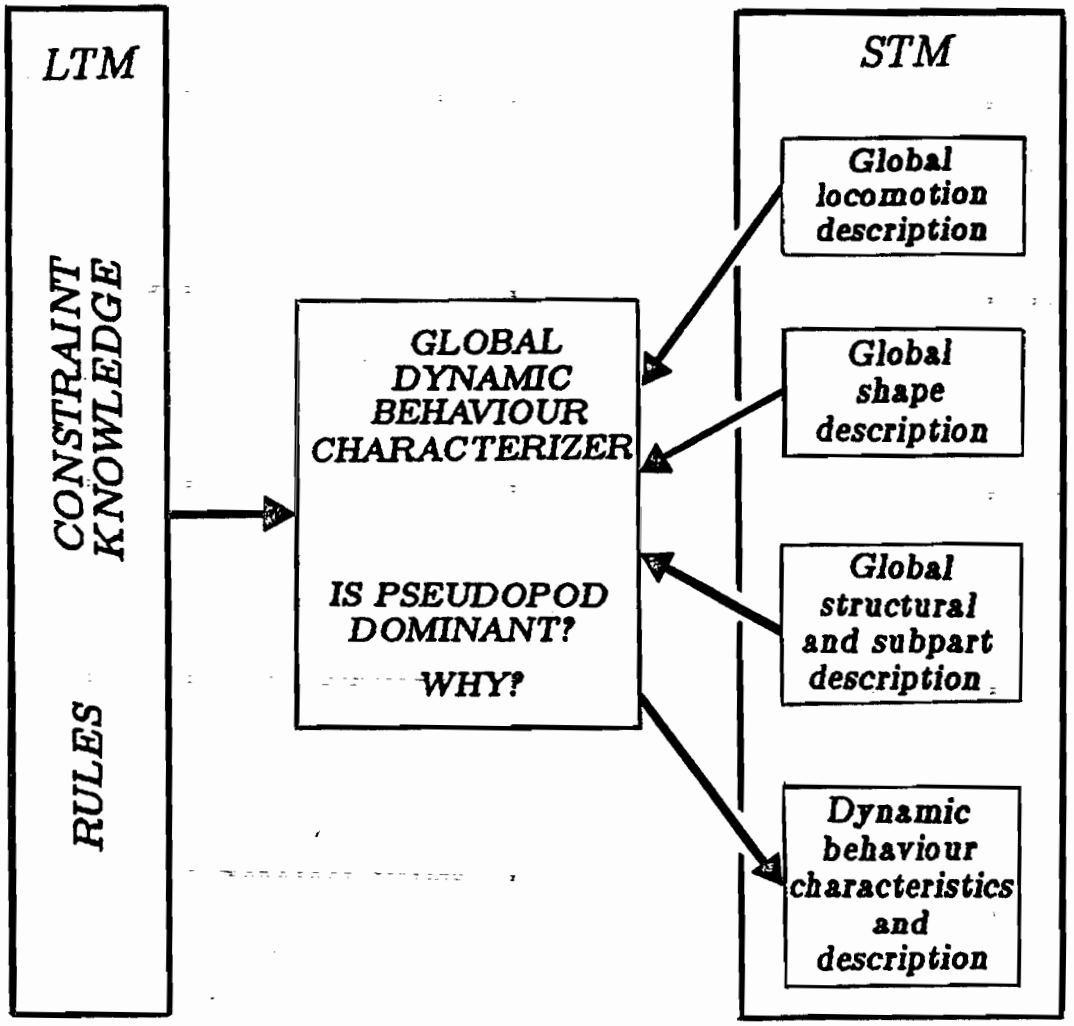
changes in each aspect individually. We should also examine the relationship between the changes in all the dynamic aspects. In fact, this is one of our main objectives. We wish to determine the relationship between the changes in the membrane shape, pseudopod characteristics, and the global locomotion of the cell. In this section, we will discuss the behaviour of a moving cell in order to answer questions pertaining to the domination of the different pseudopods in locomoting the cell. Figure (8.7) shows the processes and data structures of this stage of analysis.

A pseudopod can be defined as dominant if it leads the motion. We have employed two measures to compute the domination. The first is based on two parameters, and the second on three parameters. The first approach includes the following steps:

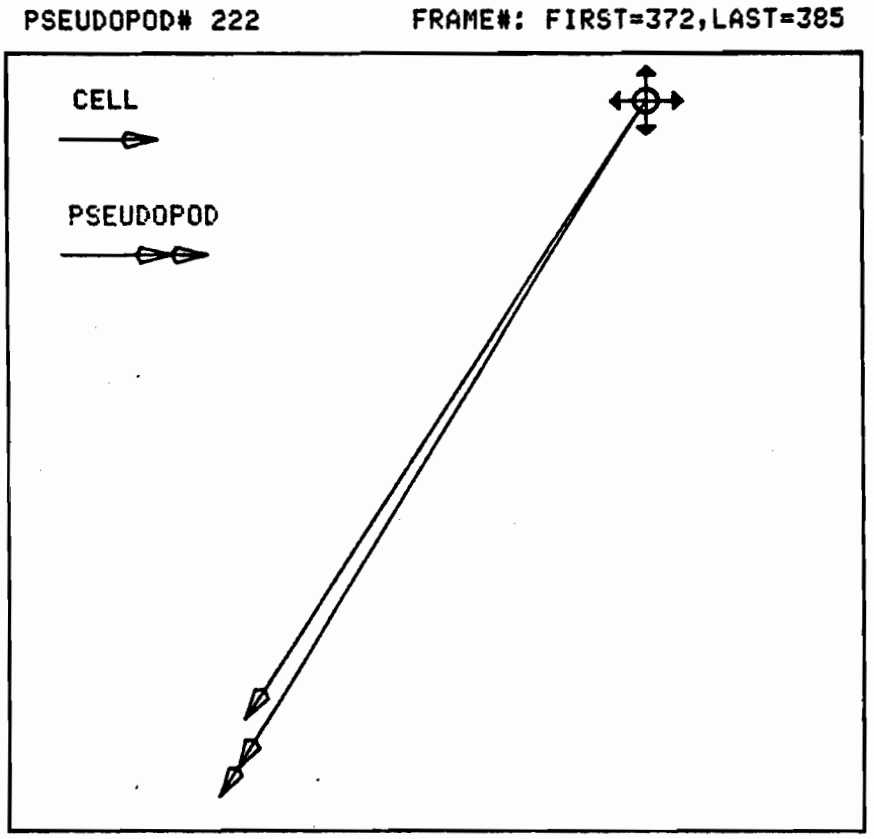
(a) Represent the dynamic motion of a pseudopod by a vector PV. This vector is the resultant of two vectors; the first representing the locomotion of the centroid of the pseudopod, and the second, the direction of growth or contraction. The latter can be computed from the increase or decrease in the pseudopod elongation in the direction of the main axis of the pseudopod.

(b) Represent the total cell locomotion during the period pseudopod existence by the vector CV.

(c) Initiate the vectors PV and CV from a point ti, where ti is time (the initial frame number) where the pseudopod commenced to appear. This is illustrated in Figure (8.8).




*Figure(8.7) Global dynamic behaviour understanding and description.*




*Figure(8.8) A vector diagram representing the dynamic changes (location and shape) of the pseudopod and the total locomotion of the cell.*

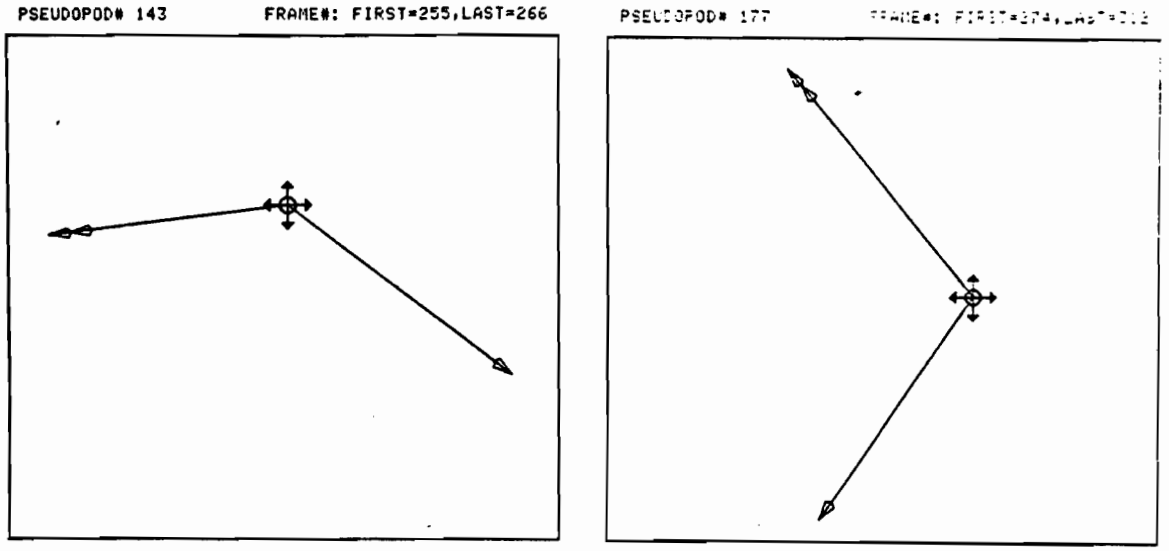
*The vectors represent the resultant motion during the existence of the pseudopod.*

(d) From the analysis of the vectors PV and CV, we can characterize the pseudopod domination. This analysis is similar to that described in Chapter 6 for the characterization of the chemotaxis behaviour.

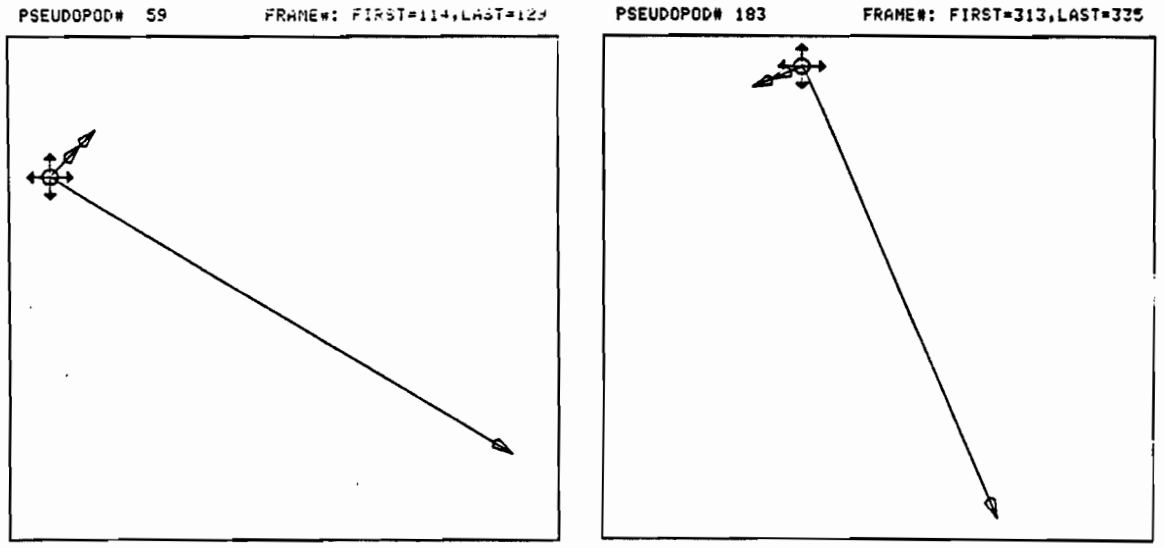
In this case, the direction of the vector PV is the direction of influence, and the vector CV will be used as the vector sum of the cell locomotion. Then, by using equation (6.38), the domination of a pseudopod in locomoting the cell is quantified and characterized as NOT DOMINANT, SLIGHTLY DOMINANT, ALMOST DOMINANT, DOMINANT, or VERY DOMINANT. Figures (8.9a-8.9e) show examples of different pseudopods and their degree of domination of the cell locomotion. A typical example of the pseudopod characterization using these two vectors is given in Description (8.3).

In the second approach, an additional factor can be considered in the pseudopod characterization. This parameter is the relative location of the pseudopod with respect to the entire cell. This can be presented by the vector PSV, which represents the connective line between the centroid of the pseudopod and that of the entire cell. This is shown in Figure (8.10). In this case, the domination DOM of a pseudopod is a function of three vectors PV, PSV, and CV:

$$DOM = f\ (\ PV,\ PSV,\ CV) \qquad (8.8)$$




*Figure(8.9a) NOT DOMINANT*

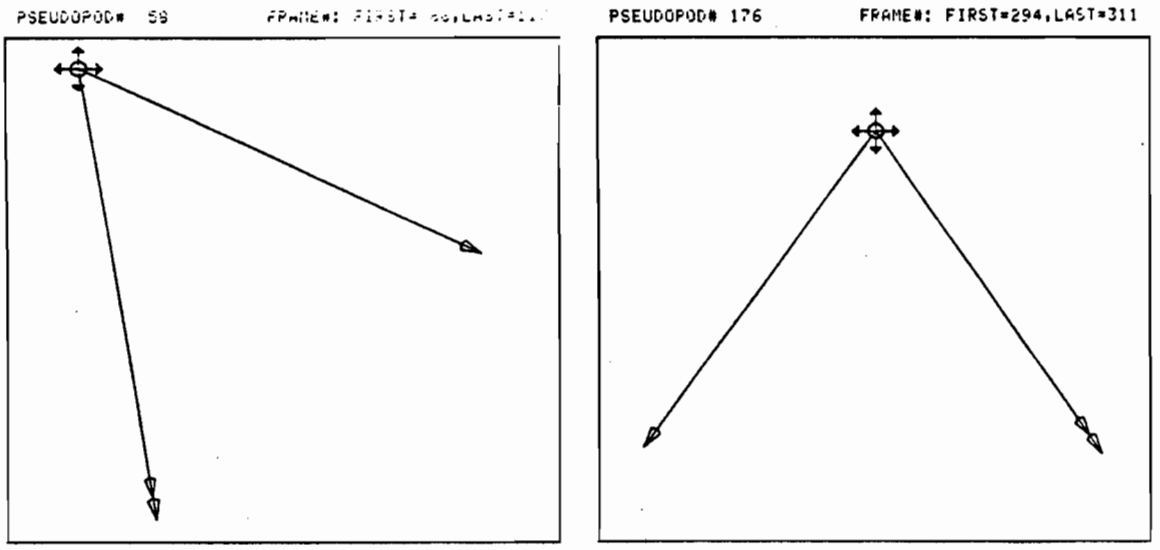



*Figure(8.9b) SLIGHTLY DOMINANT*

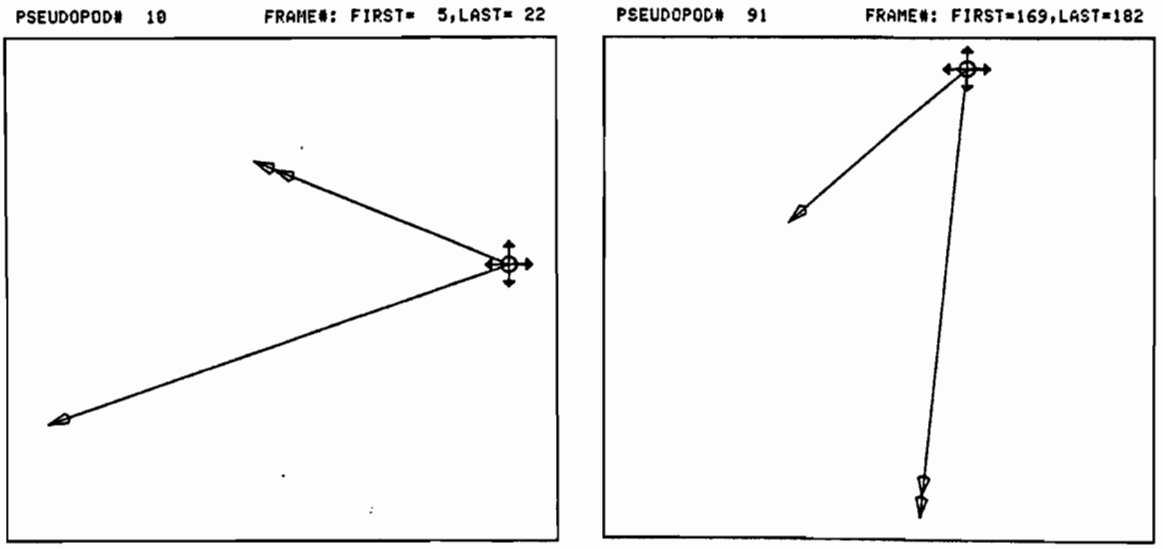
*CELL LOCOMOTION*

*PSEUDOPOD DYNAMIC CHANGES*

*Figures(8.9a-8.9b) Vector diagrams of pseudopod behaviour characterized as: (a) NOT DOMINANT*
*(b) SLIGHTLY DOMINANT*

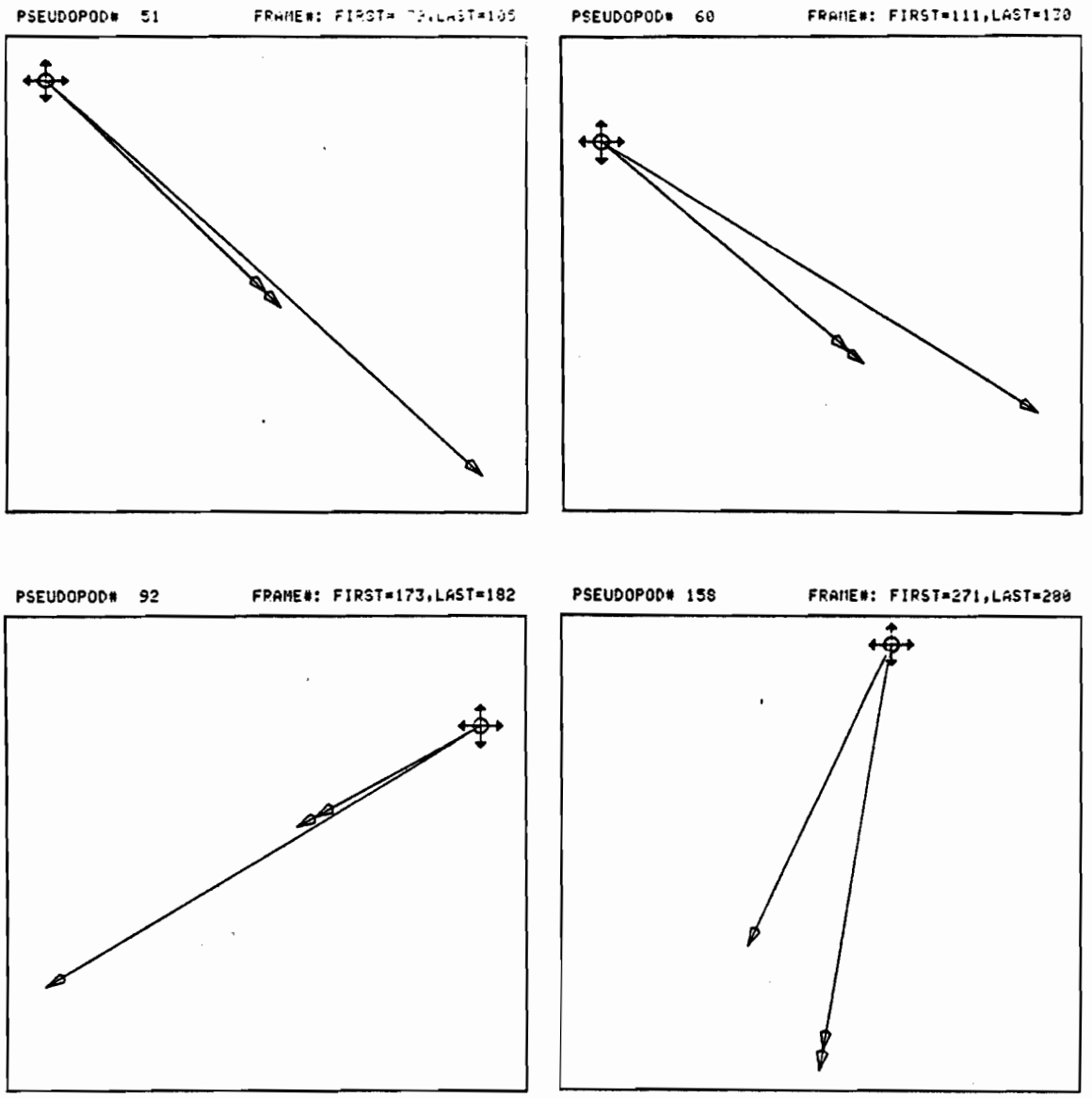



*Figure(8.9c) ALMOST DOMINANT*

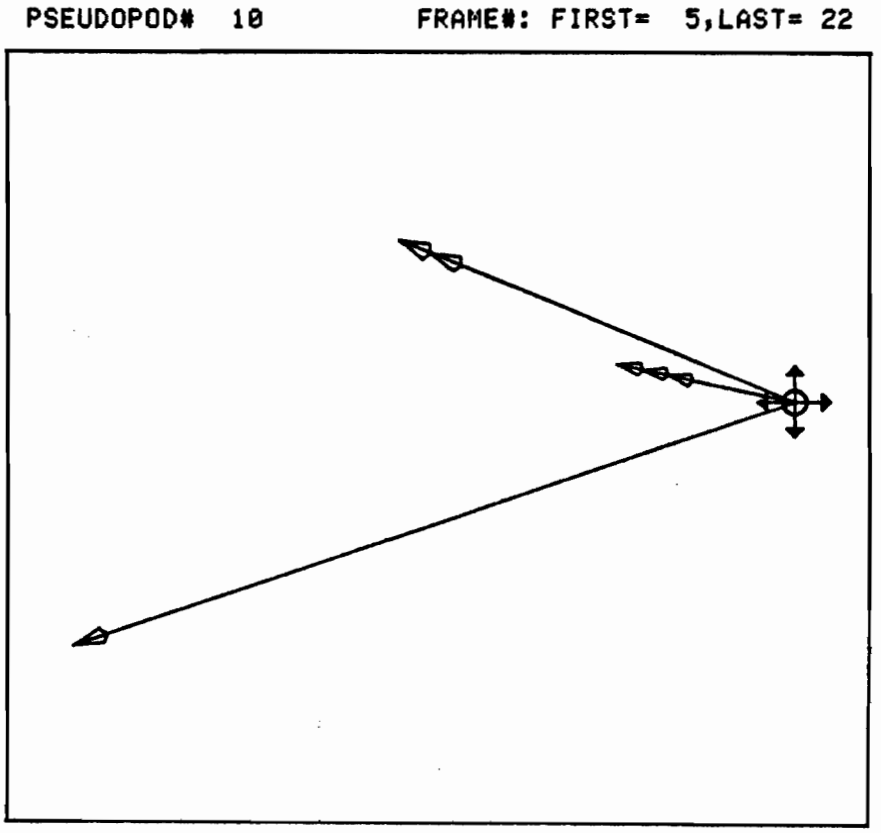



*Figure(8.9d) DOMINANT*

*Figures(8.9c-8.9d) Vector diagrams of pseudopod behaviour characterized as: (c) ALMOST DOMINANT (d) DOMINANT*



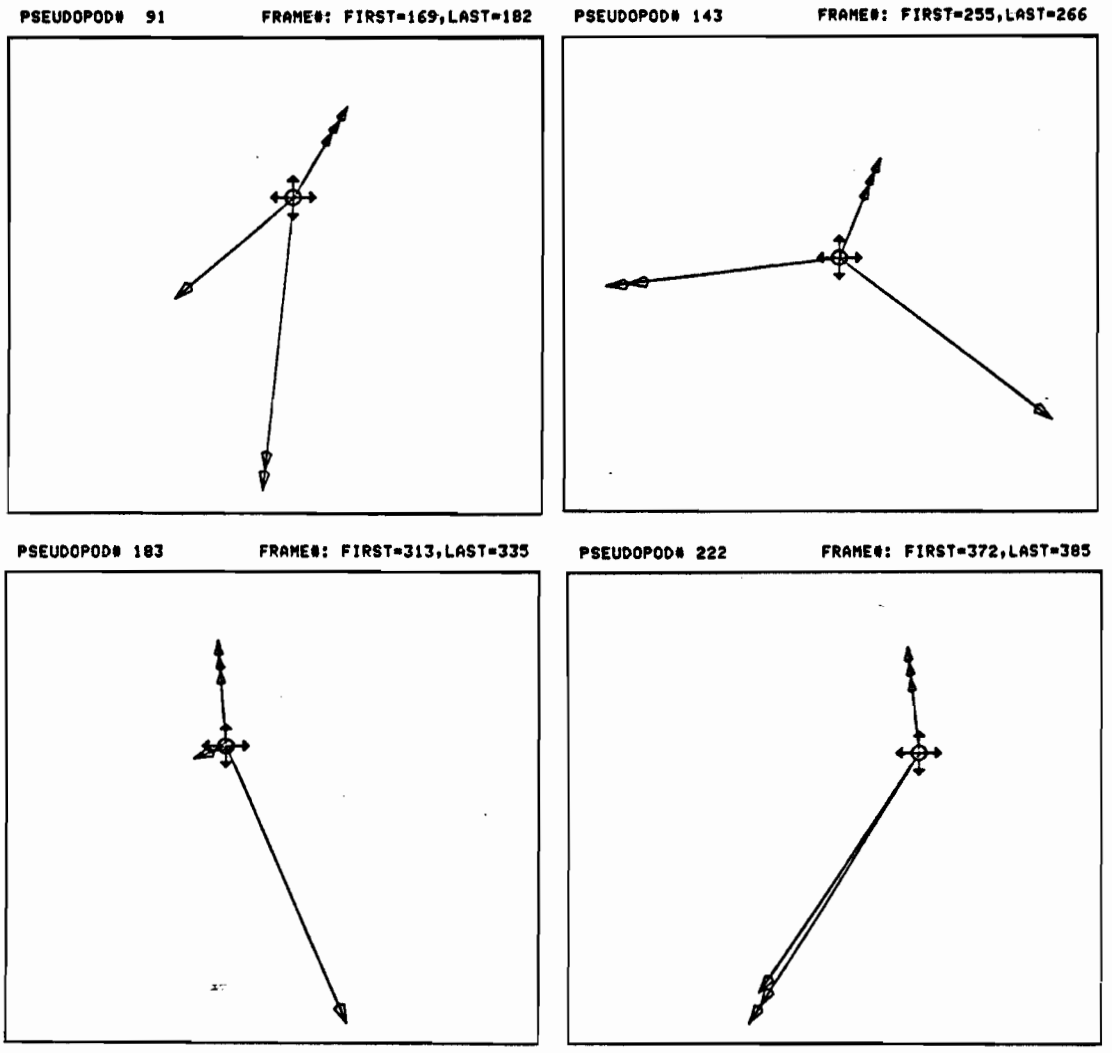

*Figure(8.9e) Vector diagrams of pseudopod behaviour characterized as VERY DOMINANT.*




*Figure(8.10) A vector diagram representing the dynamic changes of a pseudopod, its relative location, and the total locomotion of the cell.*

Thus, if we consider the cell locomotion, we assume that specific patterns in the pseudopod dynamics and location result in cell movement. Analyzing these vectors a pseudopod can be characterized as VERY DOMINANT, DOMINANT, ALMOST DOMINANT, SLIGHTLY DOMINANT, or NOT DOMINANT. These symbolic qualifications are used to discriminate between the different classes of pseudopod domination. However, the terminology is not established and may be changed in the future when the characteristic behaviour of pseudopods is better understood. Description (8.4) gives a typical example of characterizing different pseudopods using these three vectors. Examples are shown in Figures (8.11a-8.11e).

A final step pertaining to pseudopod characterization is to study the global rate of formation and deformation of the different pseudopods, and the effectiveness of each in dominating the cell locomotion. In order to accomplish this, we examine the cell locomotion path and the vectors representing the dynamics of the different pseudopods in one vector diagram, as shown in Figures (8.12a) and (8.12b). The vectors that represent the pseudopods branch from the cell path at the points where each pseudopod started to appear. From this figure, one can see the rate of pseudopod formation and the effectiveness of each in dominating the cell path. The cell locomotion in this particular figure was under the influence of bacteria located in the bottom left corner of the plane (South-West). Cell locomotion was characterized as POSITIVE CHEMOTAXIS (see Description 5.1).

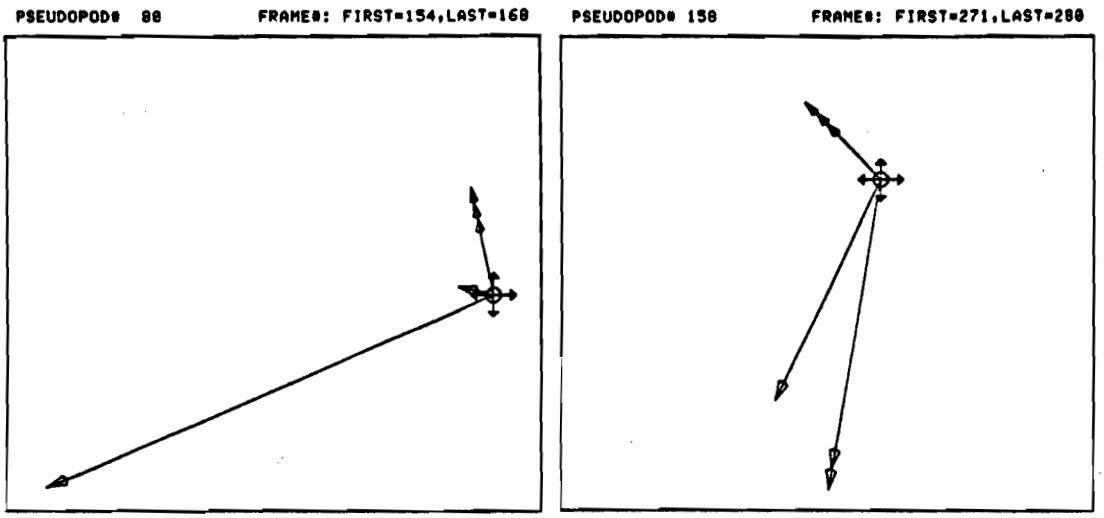



*Figure(8.11a) Vector diagrams of pseudopods that are characterized as NOT DOMINANT, using three vectors:*

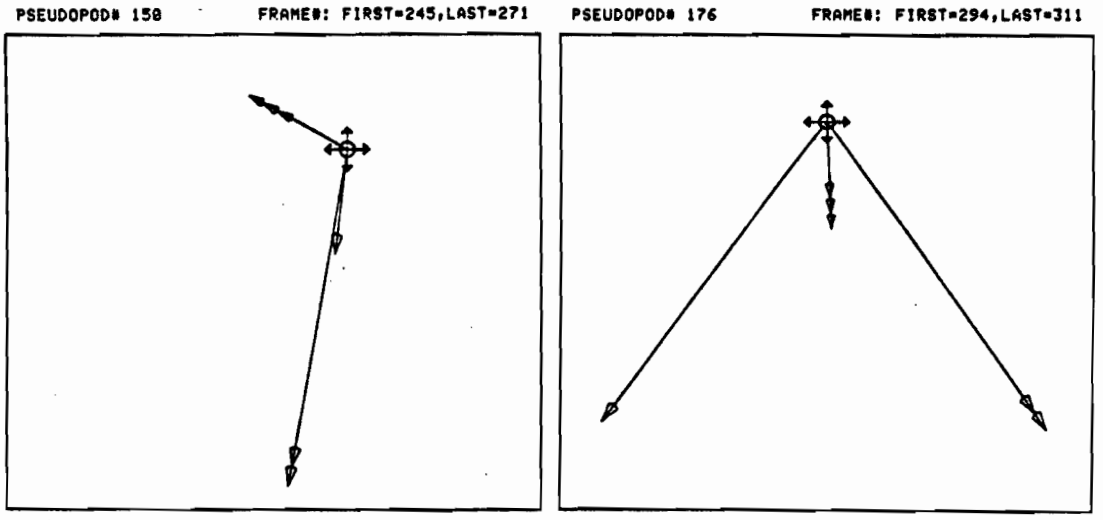
→ *CELL LOCOMOTION*

→→ *PSEUDOPOD DYNAMIC CHANGES*

→→→ *PSEUDOPOD RELATIVE LOCATION*



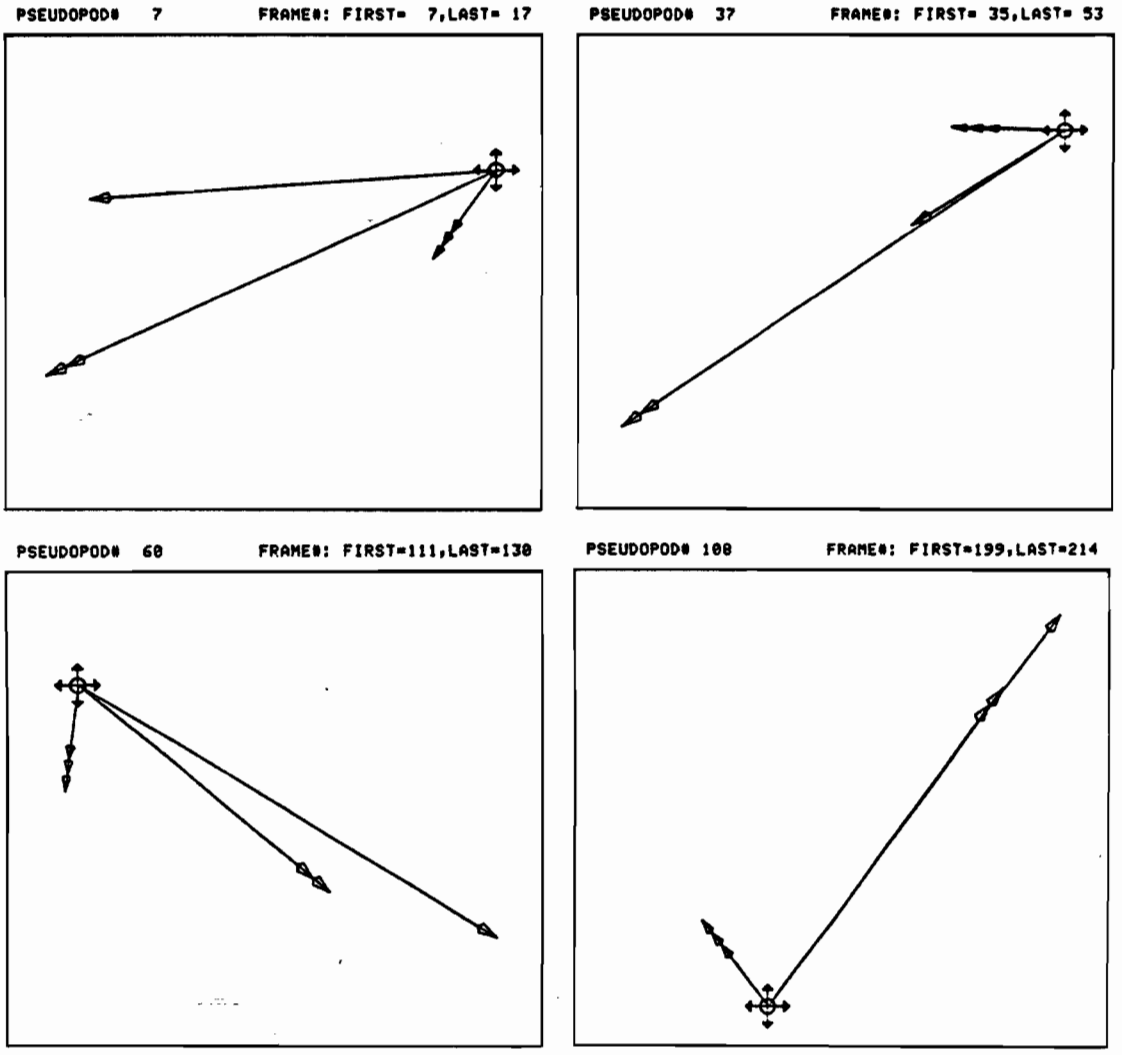

*Figure(8.11b) SLIGHTLY DOMINANT*



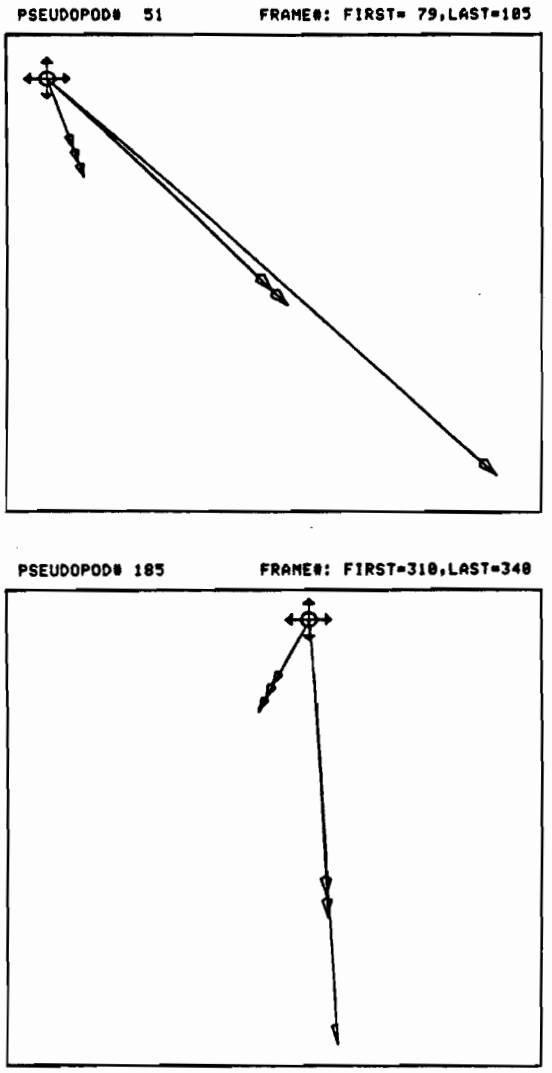

*Figure(8.11c) ALMOST DOMINANT*

*Figures(8.11b-8.11c) Vector diagrams of pseudopods that are characterized using three vectors as:*

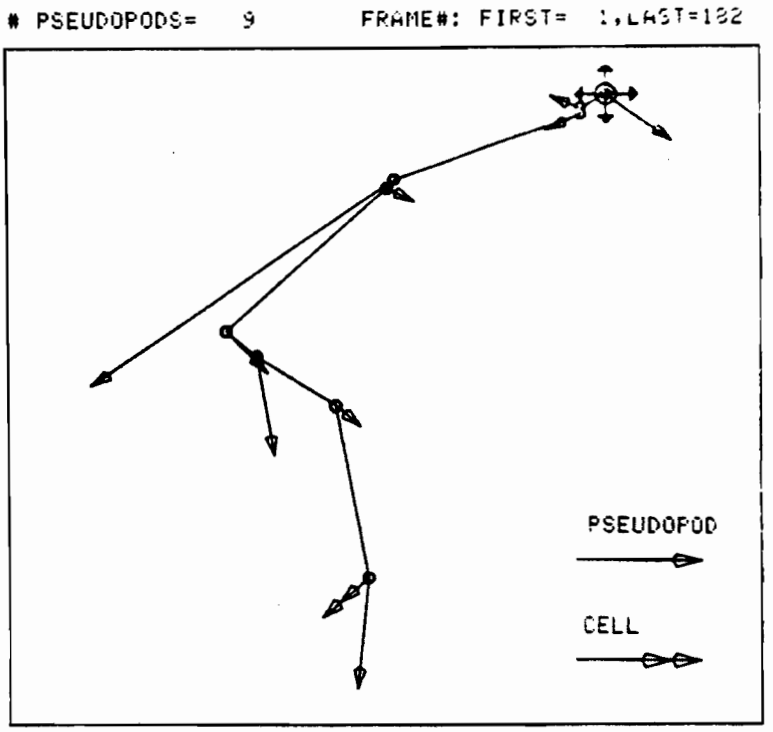
*(b) SLIGHTLY DOMINANT,*
*(c) ALMOST DOMINANT.*



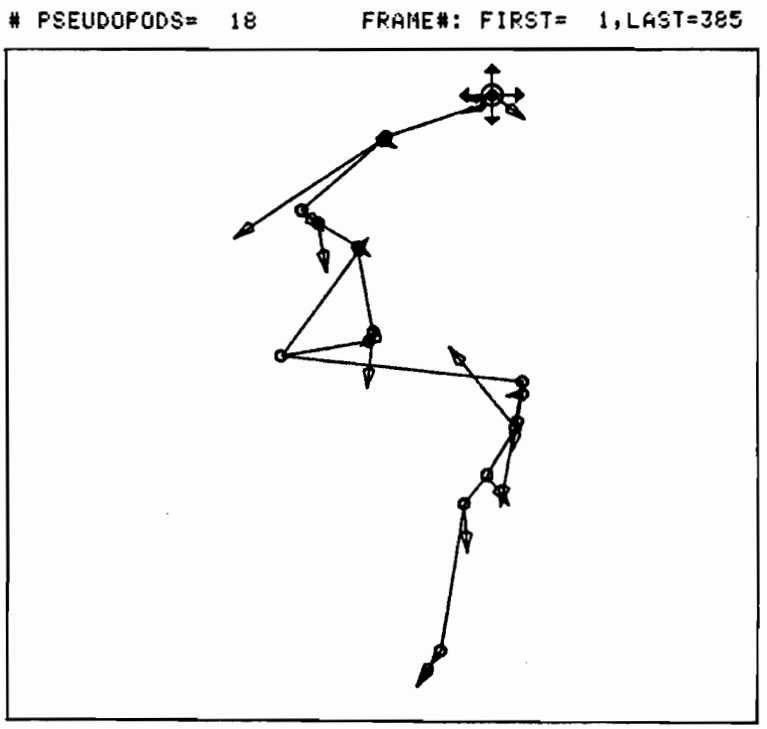

*Figure(8.11d) Vector diagrams of pseudopods that are characterized as DOMINANT, using three vectors.*




*Figure(8.11e) Vector diagrams of pseudopods that are characterized as VERY DOMINANT, using three vectors.*




*Figure(8.12a)*




*Figure(8.12b)*

*Figure(8.12) A vector diagram representing the cell locomotion path and the different pseudopods that formed during a sequence of:*
*(a) 182 frames, (b) 385 frames.*

One can see from Figure (8.12) that the dynamic behaviour of the cell is due to the effect of the pseudopod dynamics. Also, we note that the pseudopods guide the cell towards the bacteria. Thus, if we consider the cell starting at the origin of the plane, in order to reach the bacteria, it should move in the South-West direction. As we can see from Figure (8.12), whenever the cell started to lose this direction, a pseudopod formed and guided the cell in the right direction.

The accuracy of the quantification and characterization of the changes in location, shape, and structure of a moving cell that obtained by this system, are tested and evaluated based on a comparison to those obtained by a physiologist. The result of this evaluation is reported in Chapter 9.

## 8.4 SUMMARY

Understanding and describing the dynamic behaviour of a moving cell can be achieved through analysis of the global changes in its location, shape, and structure. However, the change that occurs in any of these aspects is due to and may cause change in the others. Therefore, it is not enough to study the changes in each aspect individually, but we should study the relationships between them as well. Detection and description of the global changes in locomotion and shape of a moving cell from the static and incremental data that are extracted for a sequence of images are discussed in Chapters 6 and 7, respectively. In this chapter, we have

discussed two basic issues: (a) detection and description of global structural changes, and (b) integration of the locomotion, shape, and structure descriptions in order to understand and describe the global dynamic behaviour of the cell and its pseudopods.

In static scene analysis, the cell structure was described in each frame of the sequence through a labeled graph that represents the primitive subparts of the cell. However, this structure may be false in some individual or sequence of frames, due to noise, irrelevant changes, and/or the three-dimensionality of the cell. In this chapter, we have described a rule-based method for detecting and correcting these false cases. This is accomplished by using data-driven representational and control rules that utilize information extracted from high level processes, and constraint knowledge about the cell under consideration. The objective of these rules is to describe the different situations, and provide this information to the low level processes in order to correct specific false structures.

The different properties pertaining to the location, motion, shape, and the relative location of each subpart are extracted and analyzed. In this way, the dynamic behaviour of each subpart is described. These descriptions were then utilized in order to characterize a subpart as (a) cell body, and (b) stationary, growing, or contracting pseudopod.

The interaction between the external factors and the internal cell processes occurs at or within the cell membrane by forming and deforming pseudopods. In order to understand the mechanisms that regulate the formation of these pseudopods, we studied, in the second part of this chapter, the relationship between the cell locomotion and the characteristics of the different pseudopods. In this study, a pseudopod is defined as dominant if it leads the cell locomotion. Thus, the cell locomotion can be considered as a cause and effect action. The cause is the dynamic behaviour of a pseudopod, and the effect is the cell locomotion. Based on a vector analysis of the global cell locomotion, pseudopod dynamics (location and shape), and pseudopod relative location, the domination of a pseudopod is quantified and characterized as VERY DOMINANT, DOMINANT, ALMOST DOMINANT, SLIGHTLY DOMINANT, or NOT DOMINANT.

Finally, the global locomotion path and the vectors that represent the formation of the different pseudopods during the observation period, are presented simultaneously in a vector diagram, as shown in Figure (8.12). Hence, questions about pseudopod formation, and the effectiveness of each on the cell locomotion, are answered.

# DESCRIPTION (8.1 a)

## SUBPART PROPERTY GLOBAL ANALYSIS

SUBPART NUMBER = 58

| FRAME NUMBER : | GLOBAL | LOCAL |
|---|---|---|
| FIRST | = 86 | 1 |
| LAST | = 117 | 32 |

DURATION = 32
CURVE APPROXIMATION THR. = 20.0 %
CLASSIFICATION THRESHOLDS = .100 ,.300 ,.700 ,.900 ,

### PROPERTY : AREA

NUMBER OF PERIODS = 5

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1 --> 3 | 1.5 | <-2>DECREASE | VERY LARGE ->MEDIUM |
| 2 | 4 --> 6 | 1.5 | < 1>INCREASE | MEDIUM ->LARGE |
| 3 | 7 --> 8 | 1.0 | <-1>DECREASE | LARGE ->MEDIUM |
| 4 | 9 --> 28 | 10.0 | < 0>STATIONARY | MEDIUM ->MEDIUM |
| 5 | 29 --> 32 | 2.0 | <-2>DECREASE | MEDIUM ->VERY SMALL |

### PROPERTY : BASE_LINE

NUMBER OF PERIODS = 4

| PERIOD NUMBER | FRAME NUMBERS | TIME (Sec) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1 --> 20 | 10.0 | <-3>DECREASE | VERY LONG ->SHORT |
| 2 | 21 --> 22 | 1.0 | < 1>INCREASE | SHORT ->MEDIUM |
| 3 | 23 --> 25 | 1.5 | <-1>DECREASE | MEDIUM ->SHORT |
| 4 | 26 --> 32 | 3.5 | < 3>INCREASE | SHORT ->VERY LONG |

### PROPERTY : RELATIVE BASE_LINE

NUMBER OF PERIODS = 2

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1 --> 28 | 14.0 | <0>STATIONARY | SHORT ->SHORT |
| 2 | 29 --> 32 | 2.0 | <1>INCREASE | SHORT ->MEDIUM |

## PROPERTY : PERIMETER

NUMBER OF PERIODS = 3

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1 --> 5 | 2.5 | <-2>DECREASE | VERY LONG ->MEDIUM |
| 2 | 6 --> 2 | 11.5 | < 0>STATIONARY | MEDIUM ->MEDIUM |
| 3 | 29 --> 32 | 2.0 | <-2>DECREASE | MEDIUM ->VERY SHORT |

## PROPERTY : CONNECTIVE LINE

NUMBER OF PERIODS = 3

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1 --> 5 | 2.5 | <-2>DECREASE | MEDIUM ->VERY LONG |
| 2 | 6 -->25 | 10.0 | < 4>INCREASE | VERY SHORT ->VERY LONG |
| 3 | 26 -->32 | 3.5 | <-2>DECREASE | VERY LONG ->MEDIUM |

## PROPERTY : RELATIVE AREA

NUMBER OF PERIODS = 1

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | FEATURE DESCRIPTION |
|---|---|---|---|---|
| 1 | 1--> 32 | 16.0 | <0>STATIONARY | MEDIUM ->MEDIUM |

# DESCRIPTION (8.1 b)

## SUBPART PROPERTY ANALYSIS

SUBPART NUMBER = 58

| FRAME NUMBER : | GLOBAL | LOCAL |
|---|---|---|
| FIRST | = 86 | 1 |
| LAST | = 117 | 32 |

DURATION = 32
CURVE APPROXIMATION THR. = 20.0 %
CALSSIFICATION THRESHOLD = .100 ,. 300 ,. 700 ,. 900 ,

### PROPERTY : AREA

NUMBER OF PEROIDS = 5

| PERIOD NUMBER | FRAME NUMBERS | | | TIME (SEC) | DESCRIPTION |
|---|---|---|---|---|---|
| 1 | 1 | --> | 3 | 1.5 | VERY LARGE |
| 2 | 4 | --> | 6 | 1.5 | MEDIUM |
| 3 | 7 | --> | 8 | 1.0 | LARGE |
| 4 | 9 | --> | 28 | 10.0 | MEDIUM |
| 5 | 29 | --> | 32 | 2.0 | VERY LARGE |

NUMBER OF PEROIDS = 5

| PERIOD NUMBER | FRAME NUMBERS | | | TIME (SEC) | DESCRIPTION |
|---|---|---|---|---|---|
| 1 | 1 | --> | 3 | 1.5 | <-2>DECREASE |
| 2 | 4 | --> | 6 | 1.5 | < 1>INCREASE |
| 3 | 7 | --> | 8 | 1.0 | <-1>DECREASE |
| 4 | 9 | --> | 28 | 10.0 | < 0>STATIONARY |
| 5 | 29 | --> | 32 | 2.0 | <-2>DECREASE |

NUMBER OF PEROIDS = 5

| PERIOD NUMBER | FRAME NUMBERS | TIME (SEC) | CHANGE DESCRIPTION | PROPERTY DESCRIPTION | |
|---|---|---|---|---|---|
| 1 | 1--> 3 | 1.5 | <-2>DECREASE | VERY LARGE | ->MEDIUM |
| 2 | 4--> 6 | 1.5 | < 1>INCREASE | MEDIUM | ->LARGE |
| 3 | 7--> 8 | 1.0 | <-1>DECREASE | LARGE | ->MEDIUM |
| 4 | 9-->28 | 10.0 | < 0>STATIONARY | MEDIUM | ->MEDIUM |
| 5 | 29-->32 | 2.0 | <-2>DECREASE | MEDIUM | ->VERY SMALL |

## CONCLUSION :

| | | |
|---|---|---|
| % INCREASE | = | 7.69 % |
| % DECREASE | = | 41.03 % |
| % STATIONARY | = | 51.28 % |

## GLOBAL CHANGE DESCRIPTION :

SLIGHTLY INCREASE -MODERATELY DECREASE -MODERATELY STATIONARY

## DESCRIPTION (8.2a)

# GLOBAL STRUCTURE ANALYSIS

## SUBPART CHARACTERIZATION :

SUBPART NUMBER = 58

| FRAME NUMBER : | | GLOBAL | LOCAL |
|---|---|---|---|
| | FIRST | = 86 | 1 |
| | LAST | = 117 | 32 |

DURATION = 32

CURVE APPROXIMATION

| PERIOD NUMBER | FRAME NUMBERS | TIME (sec) | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 15 | 7.5 | CELL BODY |
| 2 | 16 --> 29 | 7.0 | PSEUDOPODE |
| 3 | 30 --> 32 | 1.5 | PSEUDOPODE |

SUMMARY OF SUBPART CHARACTERIZATION AFTER ANALYSIS

NUMBER OF PERIODS = 2

| PERIOD NUMBER | FRAME NUMBERS | TIME (sec) | DESCRIPTION |
|---|---|---|---|
| 1 | 1 --> 15 | 7.5 | CELL BODY |
| 2 | 16 --> 32 | 8.5 | PSEUDOPOD |

# DESCRIPTION (8.2b)

# GLOBAL STRUCTURE ANALYSIS

## PSEUDOPOD CHARACTERIZATION :

SUBPART NUMBER = 58

| FRAME NUMBER : | GLOBAL | LOCAL |
|---|---|---|
| FIRST | = 86 | 1 |
| LAST | = 117 | 32 |

DURATION = 32

NUMBER OF PERIODS = 18

| PERIOD NUMBER | FRAME NUMBER | TIME (sec) | CHARACTERIZATION |
|---|---|---|---|
| 1 | 1 --> 15 | 7.5 | CELL BODY |
| 2 | 16 --> 16 | 0.5 | STATIONARY PSEUDOPOD |
| 3 | 17 --> 17 | 0.5 | STATIONARY PSEUDOPOD |
| 4 | 18 --> 18 | 0.5 | STATIONARY PSEUDOPOD |
| 5 | 19 --> 19 | 0.5 | STATIONARY PSEUDOPOD |
| 6 | 20 --> 20 | 0.5 | STATIONARY PSEUDOPOD |
| 7 | 21 --> 21 | 0.5 | STATIONARY PSEUDOPOD |
| 8 | 22 --> 22 | 0.5 | STATIONARY PSEUDOPOD |
| 9 | 23 --> 23 | 0.5 | STATIONARY PSEUDOPOD |
| 10 | 24 --> 24 | 0.5 | STATIONARY PSEUDOPOD |
| 11 | 25 --> 25 | 0.5 | STATIONARY PSEUDOPOD |
| 12 | 26 --> 26 | 0.5 | STATIONARY PSEUDOPOD |
| 13 | 27 --> 27 | 0.5 | STATIONARY PSEUDOPOD |
| 14 | 28 --> 28 | 0.5 | STATIONARY PSEUDOPOD |
| 15 | 29 --> 29 | 0.5 | CONTRACTED PSEUDOPOD |
| 16 | 30 --> 30 | 0.5 | STATIONARY PSEUDOPOD |
| 17 | 31 --> 31 | 0.5 | STATIONARY PSEUDOPOD |
| 18 | 32 --> 32 | 0.5 | STATIONARY PSEUDOPOD |

SUMMARY AFTER ANALYSIS

NUMBER OF PERIODS = 4

| PERIOD NUMBER | FRAME NUMBER | TIME (sec) | CHARACTERIZATION |
|---|---|---|---|
| 1 | 1 --> 15 | 7.5 | CELL BODY |
| 2 | 16 --> 28 | 6.5 | STATIONARY PSEUDOPOD |
| 3 | 29 --> 29 | 0.5 | CONTRACTED PSEUDOPOD |
| 4 | 30 --> 32 | 1.5 | STATIONARY PSEUDOPOD |

# DESCRIPTION (8.3)

## PSEUDOPOD CHARACTERIZATION AND DESCRIPTION

The following results are obtained using the vectors for global cell locomotion and pseudopod dynamics as parameters for characterization.

| PSEUDOPOD NUMBER | DURATION (SEC) | DOMINATION PERCENTAGE | DESCRIPTION |
|---|---|---|---|
| 13 | 11.5 | 32.13 | NOT DOMINANT |
| 10 | 9.0 | 76.66 | DOMINANT |
| 7 | 5.5 | 88.23 | DOMINANT |
| 37 | 9.5 | 99.23 | VERY DOMINANT |
| 32 | 5.5 | 37.39 | NOT DOMINANT |
| 51 | 13.5 | 98.76 | VERY DOMINANT |
| 58 | 16.0 | 69.27 | ALMOST DOMINANT |
| 60 | 10.0 | 95.24 | VERY DOMINANT |
| 59 | 8.0 | 57.60 | SLIGHTLY DOMINANT |
| 80 | 7.5 | 80.04 | DOMINANT |
| 91 | 7.0 | 76.20 | DOMINANT |
| 92 | 5.0 | 98.66 | VERY DOMINANT |
| 108 | 8.0 | 99.91 | VERY DOMINANT |
| 150 | 13.5 | 98.33 | VERY DOMINANT |
| 143 | 6.0 | 24.88 | NOT DOMINANT |
| 158 | 5.0 | 91.33 | VERY DOMINANT |
| 177 | 19.5 | 40.46 | NOT DOMINANT |
| 176 | 9.0 | 60.65 | ALMOST DOMINANT |
| 185 | 15.5 | 99.81 | VERY DOMINANT |
| 183 | 11.5 | 50.22 | SLIGHTLY DOMINANT |
| 222 | 7.0 | 99.13 | VERY DOMINANT |

# DESCRIPTION (8.4)

## PSEUDOPOD CHARACTERIZATION AND DESCRIPTION

The following results are obtained using the vectors for global cell locomotion, pseudopod dynamics, and pseudopod relative location as parameters for characterization.

| PSEUDOPOD NUMBER | DURATION (SEC) | D O M I N A T I O N PERCENTAGE | DESCRIPTION |
|---|---|---|---|
| 13 | 11.5 | 31.61 | NOT DOMINANT |
| 10 | 9.0 | 79.57 | DOMINANT |
| 7 | 5.5 | 79.48 | DOMINANT |
| 37 | 9.5 | 89.34 | DOMINANT |
| 32 | 5.5 | 39.54 | NOT DOMINANT |
| 51 | 13.5 | 91.25 | VERY DOMINANT |
| 58 | 16.0 | 34.14 | NOT DOMINANT |
| 60 | 10.0 | 78.06 | DOMINANT |
| 59 | 8.0 | 45.63 | NOT DOMINANT |
| 80 | 7.5 | 58.62 | SLIGHTLY DOMINANT |
| 91 | 7.0 | 28.61 | NOT DOMINANT |
| 92 | 5.0 | 93.80 | VERY DOMINANT |
| 108 | 8.0 | 77.34 | DOMINANT |
| 150 | 13.5 | 60.05 | ALMOST DOMINANT |
| 143 | 6.0 | 32.07 | NOT DOMINANT |
| 158 | 5.0 | 58.37 | SLIGHTLY DOMINANT |
| 177 | 19.5 | 55.36 | SLIGHTLY DOMINANT |
| 176 | 9.0 | 69.12 | ALMOST DOMINANT |
| 185 | 15.5 | 90.91 | VERY DOMINANT |
| 183 | 11.5 | 23.14 | NOT DOMINANT |
| 222 | 7.0 | 46.12 | NOT DOMINANT |

# CHAPTER 9

# DISCUSSION AND CONCLUSIONS

## 9.1 INTRODUCTION

In the preceding chapters of this thesis, we described the different aspects pertaining to the construction and implementation of a system for quantifying and characterizing the changes in location, shape, and structure of a moving cell. The discussion in the thesis started in Chapter 2 with the review and analysis of the previous experience in related areas of study. It ended in Chapter 8 by describing higher level processes that are concerned with the integration of the aspects pertaining to locomotion, shape, and structure, in order to understand and characterize the dynamic behaviour of the cell.

This chapter presents a summary and general discussion of the present research. First, in Section 9.2, the different aspects described in this thesis will be summarized, focussing on the main contributions of the thesis. In Section 9.3 we present examples of experimental results and their comparison to those obtained by a physiologist. Suggestions for modifying and expanding the system are discussed in Section 9.4, as well as

possibilities for further experiments and different applications. Finally, Section 9.5 is a conclusion.

## 9.2 SUMMARY AND CONTRIBUTIONS

The primary function of the cell surface is to transform information from the environment to the cell. It has become increasingly evident that the cell surface plays a pivotal role in the life, development, and regulation of cells. The mechanisms that regulate this social behaviour are not well understood, but recent experiments have indicated that the cell membrane plays a vital role. However, there is no existing method for quantifying the observable changes in membrane shape that occur in locomotion. To achieve this objective using automatic techniques of digital image processing, this thesis presents an image interpretation system capable of analyzing the structural changes in the morphology of a non-rigid moving object from a sequence of pictures.

A model for a general dynamic scene analysis system has been constructed. It consists of three basic entities: dynamic data, static data, and a collection of analysis processors. The different types of data which may be manipulated by the system have been classified into: a sequence of images, a group of objects and subobjects, a set of object features, symbolic descriptors, global behaviour characteristics. The latter are functions of groups of features and descriptors used to describe specific

behavioural patterns. A set of rules, which may be classified as representational and control, is also employed.

Based on this model, we have implemented a rule-based image interpretation system for moving cells. The system consists of different cooperating computational processes. Conceptually, two different memories are used, a Short Term Memory (STM) and a Long Term Memory (LTM). Both are implemented as a relational database. The STM is designed to work as a communication channel for all of the processes. It contains a dynamic record of the instantaneous cell motion, shape, and structural changes, as well as the current description of the cell behaviour. The LTM data are static, and are implemented as rules. These describe the general model of the morphology of the cells under analysis, as well as control information pertinent to the computational processes. The latter are activated by the control rules throughout the three hierarchical analysis stages: static, incremental, and global. They interact through the STM using the information stored in the LTM, until a complete description of the dynamic cell motion and morphology is obtained.

Comparing the structure of an image sequence analysis system described in this thesis to others, we may claim two original contributions. First, the construction of our model of cell motion as a rule-based system (knowledge representation and control strategy). Within this

structure, the dynamic behaviour of the moving cell is described using generic knowledge (constraints) and rules. Consequently, the system has much wider application, especially for those sequences containing moving objects whose motion patterns are not known a priori, or which exhibit random motion. Second, in our work we analyze, quantify, and symbolically describe the structural changes of non-rigid moving objects, a subject hitherto neglected. In particular, the quantification of the pseudopod dynamics has not appeared in the literature.

Constructing and implementing an understanding system capable of analyzing and describing the dynamic behaviour of a moving cell represents a merging of four different disciplines in computer vision and image processing. They are: (a) Automatic Processing of Microscopic Images, (b) Image Sequence Analysis, (c) Shape Analysis and Description, and (d) Knowledge-Based Systems. An analysis and brief review of the significant work done in each of these areas, as well as the contribution of our work in each field, was summarized and presented in Chapter 2.

With regard to the general problem of processing dynamic images, two main issues have been ignored by most of the past research: (a) shape and structural changes of a non-rigid moving object, and (b) motion understanding and description. These issues are among the aspects addressed in this thesis. We analyzed the dynamics of a moving cell, which changes its shape and structure randomly due to its

physical properties. Thus, we have considered three kinds of changes with time: locomotion, shape, and structure. All alter randomly from frame to frame and interact with each other.

To achieve the objectives of motion understanding and description, it is not enough to merely determine the incremental movements or changes that occur between consecutive images. What is required is a system which abstracts a description of the global motion characteristics from the static and incremental data. Development of such a system represents the approach taken in our research.

Shape analysis and description is a central issue to most computer vision and pattern recognition systems. This problem was discussed in the thesis. Besides the general difficulties of describing an arbitrary shape in a specific image, we have studied the following problems: (a) Estimating the incremental change in the shape and structure of a non-rigid moving object such as a cell. (b) Detecting and characterizing the structural changes in its morphology over a period of time from a sequence of pictures. (c) Presenting all of the above descriptions in a meaningful terminology to the user.

We have developed a procedure which produces a meaningful symbolic description of the shape and its changes. We have also presented an expression for measuring the complexity of an arbitrary shape. This expression is

based on a group of selected shape properties which are independent of translation, rotation, and size. Another shape property was introduced to measure the degree of curvature regularity (angle and side regularity) of the shape of an object. This property is shown experimentally to play a considerable role in shape discrimination and is used to describe membrane shape.

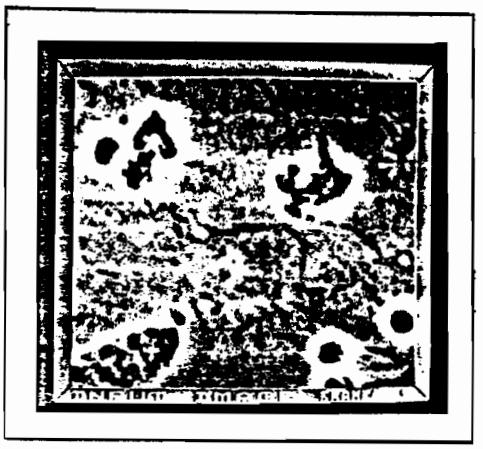
It is of interest to describe the dynamic activity of a cell using symbolic terminology which is meaningful to the individuals working in cell biology. With the aid of this system, one of the cell's primary behavioural characteristics is described, namely, the chemotaxis. This refers to the directional locomotion of the cell influenced by an external factor. Thus, the effectiveness of the latter on modifying the cell locomotion is quantified. The global changes in the cell structure are also analyzed. Hence, a subpart of the cell is classified as being either "pseudopod" or "cell body". A pseudopod is described as "growing", "contracting", or "stationary". Furthermore, other aspects of the global behaviour of the cell are characterized and described. For example, the "domination" of a pseudopod in contributing to the locomotion of the cell. Samples of the generated characterization are given in Descriptions 4.1, 5.1, 6.1, 7.1, 8.1, and 9.1.

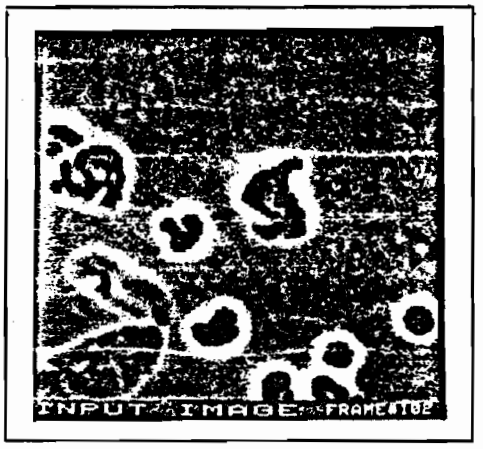
# 9.3 EXPERIMENTAL RESULTS

## 9.3.1 Neutral And Chemotactic Conditions

The system has been tested by quantifying and characterizing the dynamic behaviour of a polymorphonuclear leucocyte (PMN), as well as, its pseudopod kinetics in both chemotactic and neutral conditions. In the chemotactic case, the cell motion was recorded under the influence of a small sample of 'E. coli' (bacteria). Figure (9.1) shows two frames of the same sequence (9 and 102). In this figure, the bacteria are shown in the South-West corner of the image. We can see that the cell in the North-West corner in frame 9 has moved towards the bacteria in frame 102. This indicates positive chemotaxis. A sequence of 450 frames (225 seconds) was analyzed by the system. The complete description of the results of the different stages of the analysis was presented throughout the thesis. For example, the global locomotion of the cell is described by the system as "THE CELL HAS AN AVERAGE POSITIVE CHEMOTAXIS" (see Description 6.1 for complete details). In addition, the pseudopod dynamics that contributed to this chemotactic locomotion are quantified and characterized, a complete description of which is presented in Descriptions 8.3 and 8.4 (also see Figures 8.10 to 8.12 inclusive).

With respect to the neutral condition, the cell motion was recorded without the presence of external chemotactic factors. The same experiments conducted for chemotactic



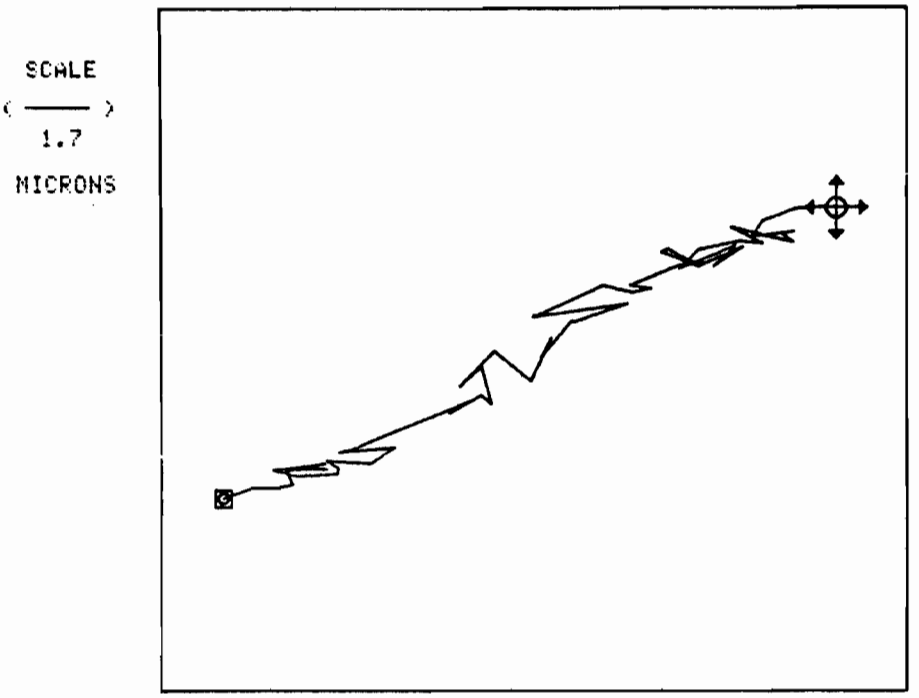



**Figure(9.1)** *Typical examples of input images at different times in a recorded sequence of the dynamic movement of a neutrophil cell.*

*The cell in NE corner in frame 9 has moved in a South-Westerly direction in frame 102.*

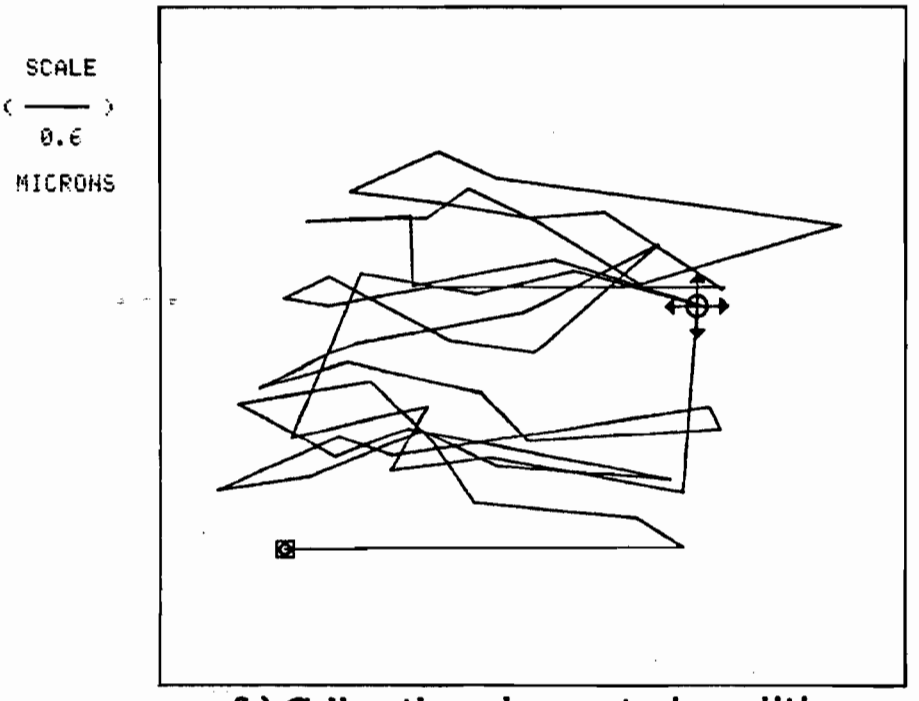
locomotion were used to analyze the cell dynamic behaviour under the neutral conditions. Examples of the results of these experiments are shown in Figures (9.2) to (9.5). In order to understand the difference between the cell dynamics under chemotactic and neutral conditions, we present in these figures the cell path using the same sampling parameters for both chemotactic and neutral conditions. The same number of frames (60) is shown in each case. From these figures, we can observe the following: (a) The cell path under chemotactic conditions is straighter and more directed than the one under the neutral conditions (see Figures 9.2 and 9.3). (b) The vector representing the total locomotion of the cell in the different directions of the plane is ALMOST SIMILAR for the neutral conditions. This situation is described by the system as RANDOM LOCOMOTION. In the chemotactic case, the vector is LONGER in the direction of the bacteria (see Figures 9.4 and 9.5). The vector sum of the cell locomotion is almost zero in the neutral case, whereas in the chemotactic condition it has a value in the direction of the bacteria (see Figure 9.5). This is the situation even though a relatively short path is examined in the example shown in these figures. The amplitude and direction of the motion vector are utilized to quantify and describe the chemotaxis behaviour (see Section 6.4).

**Frame number: First=1, Last=60, Sampling=1 frame (.5 seconds)**




*(a) Cell path under chemotactic condition.*

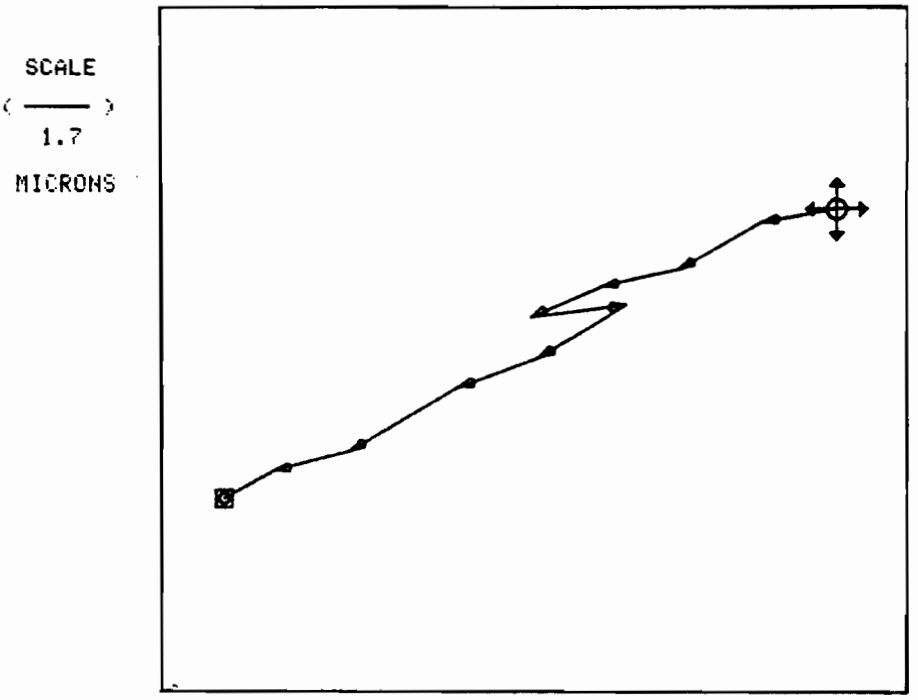
**Frame number: First=1, Last=60, Sampling=1 frame (.5 seconds)**




*(b) Cell path under neutral condition.*

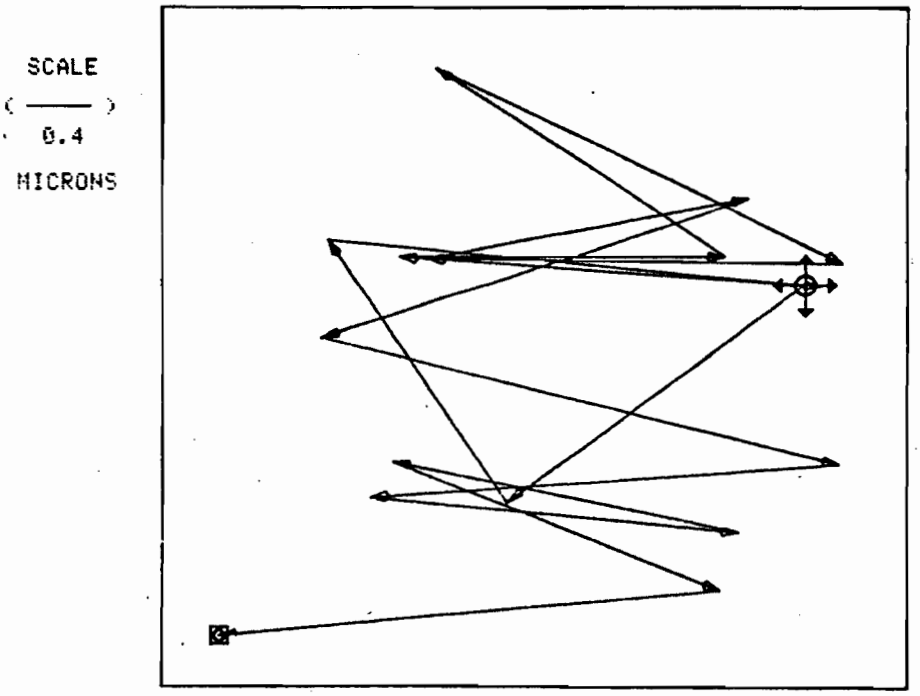
*Figure (9.2) Time sampling of the cell path under neutral and chemotactic conditions. (a) Chemotactic (b)Neutral.*

**Frame number: First=1, Last=60, Sample distance=2 microns (2 pixels)**



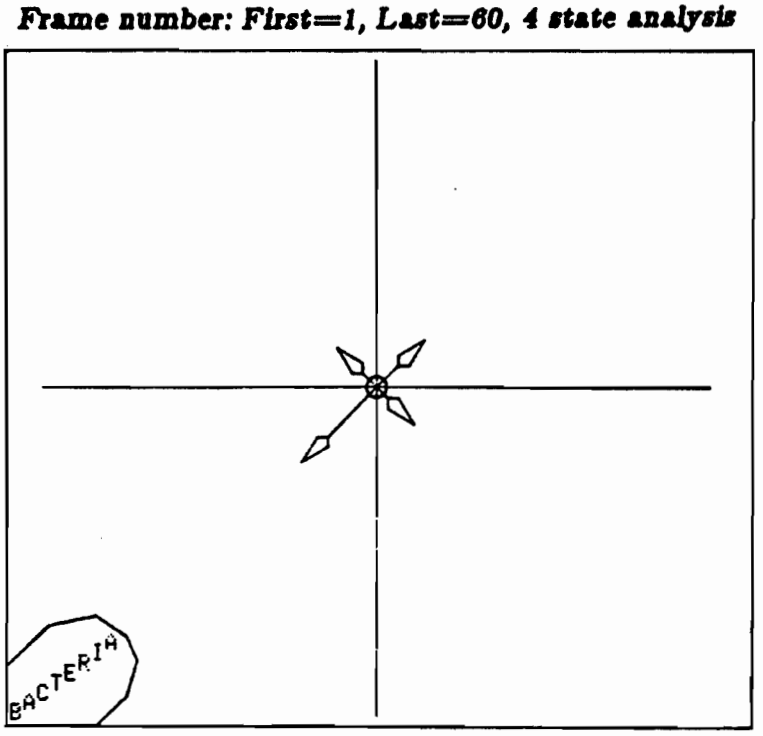

*(a) Cell path under chemotactic condition.*

**Frame number: First=1, Last=60, Sample distance=2 microns (2 pixels)**

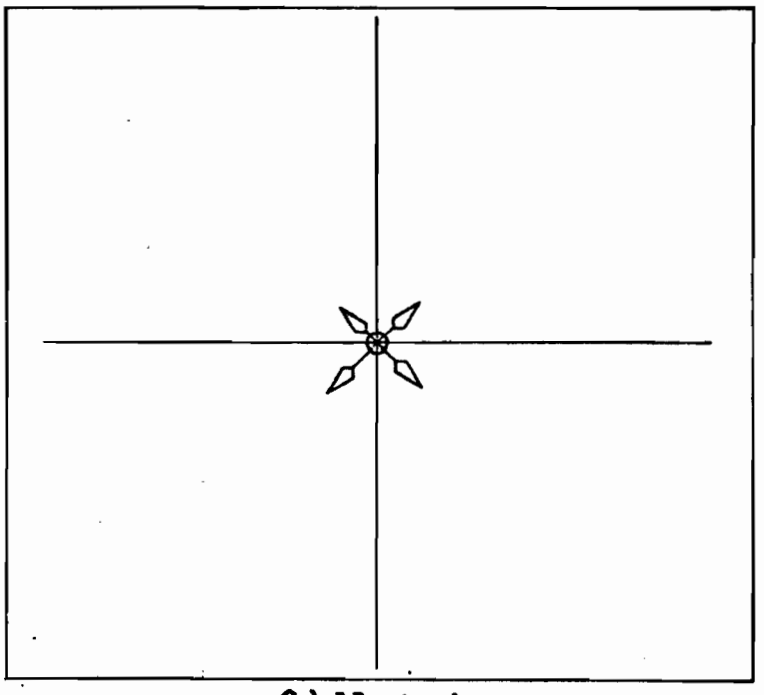



*(b) Cell path under neutral condition.*

*Figure(9.8) Distance sampling of the cell path under chemotactic and neutral conditions. (a) Chemotactic (b) Neutral.*

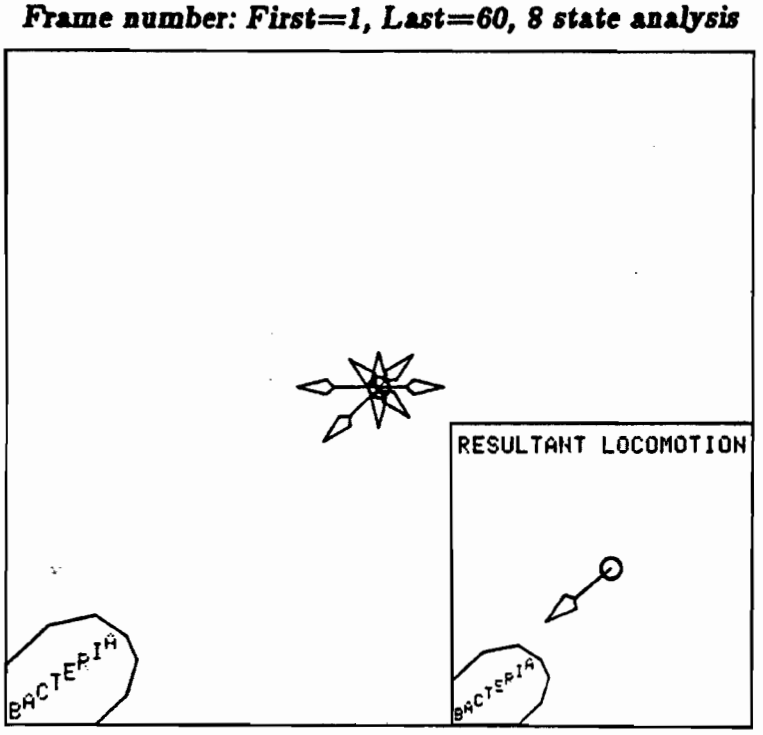



*(a) Chemotactic.*

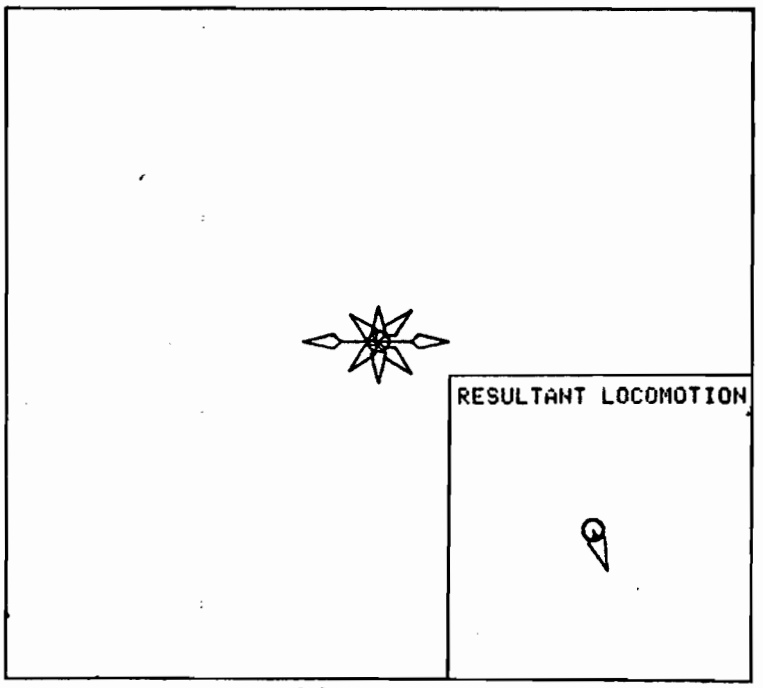



*(b) Neutral.*

*Figure(9.4) Vector sum of cell locomotion in each of four directions under: (a) Chemotactic condition (b) Neutral condition.*




*(a) Chemotactic.*




*(b) Neutral.*

*Figure(9.5) Vector sum of cell locomotion in each of eight directions under: (a) Chemotactic condition (b) Neutral condition.*

The above observations represent an example of the dynamic behaviour that are quantified preciesly by the system and summarized in numeric values and symbolic description that is a meaningful to the biologist.

## 9.3.2 Comparison To Characterization By A Physiologist

The quantification and characterization of the changes in location, shape, and structure of a moving cell that are obtained by this system have not been computed before, either visually (biologist) or automatically (computer). In order to test the accuracy of the resulting data, the ouput information at each stage of analysis was presented to a physiologist (Dr. P. Noble, Faculty of Dentistry, McGill University). Most of the computer results agreed with his a priori knowledge about cell and pseudopod dynamic behaviour. However, some of these data, especially those pertaining to specific membrane shape and structural changes, cannot be evaluated at this time because they have not been reported before, and are not yet well understood.

The same sequence of frames that was analyzed by the system has been studied by the physiologist. The pseudopods that were characterized automatically are described in Descriptions (8.3) and (8.4), and those inspected visually are reported in Description (9.1). We have compared the two sets of results, and this comparison is shown in Descriptions (9.2) and (9.3). A percentage error is estimated based on the differences in the characterization.

The results indicate an error of 18% in the case where cell locomotion and pseudopod dynamic changes were used as characterization parameters. This error is reduced to 10% by using the pseudopod relative location as the third parameter for characterization.

From this comparison, we claim that the system has successfully characterized the kinetics of the pseudopods that contribute to the cell locomotion. Using the developed technique, it is now possible to study why in random motion one pseudopod becomes dominant and to compare these pseudopod characteristics with pseudopods during chemotactic locomotion. The technique, being applicable to other leucocyte types, i.e. lymphocytes, will enable one to study pseudopod kinetics in positive and negative chemotaxis. This future study is discussed in the following section.

## 9.4 FUTURE WORK

Lymphocytes play an important role in host defense mechanisms. They are a heterogeneous group of cells which can be subclassified on the basis of their functional response. 'B' lymphocytes are bone marrow dependent and produce antibodies in response to antigenic challenge. The thymus dependent 'T' lymphocytes are responsible for cell-mediated immune response as well as the regulation of B cell function [Pritchard et al., 73].

Recently, the T lymphocytes have been subfractionated into three classes based on the surface receptors to the Fc portion of the immunoglobulin IgG and IgM. One class consists of 'T' cells having receptors for IgM; that is the Tm cells enhance the activity of B lymphocytes to produce antibody. The second class of T cells, with receptors for IgG, Tg cells suppress B lymphocyte activity [Ferrarini et al., 75; Moretta et al., 75]. A third class of T lymphocytes, To, has neither of the above receptors and has been implicated in the natural killing of tumour cells [Suksela et al., 79]. These lymphocyte subsets have been shown to respond differently to various lymphocyte cytotaxins [El-Nagar et al., 80]. It has been shown recently that lymphocytes exhibit a negative chemotaxis against certain stages of the natural history of tumour cells [Noble and Lewis, 79].

It would be of interest to utilize the pseudopod characterization technique discussed in this thesis to study the pseudopod kinetics of T cell subsets undergoing random locomotion and positive and negative chemotaxis. The characterization of lymphocyte locomotory responses would allow us to detect, if any, abnormalities in locomotry response existing in a variety of disease states. If these exist, then the efficiency of lymphocyte host defense mechanism could by severely compromised.

In this section, we discussed the use of the developed technique in future research and experiments pertaining to the understanding of the dynamic behaviour of different types of cells. However, there are other suggestions for modifying and/or expanding the system, in particular for the use in other applications. This will be discussed below.

In this research we have studied the dynamic behaviour of a moving cell from a sequence of two-dimensional images. Most of the false segmentations, decompositions, and artifactual changes are due to the effect of the three dimensionality of the cell motion. Fortunately, in the present application, the three dimensional motion of the cells is restricted to some extent, because these cells (PMN) require a substratum on which they flatten before commencing locomotion. However, for general applications, for example, human, animal, or vehicle motion, it would be necessary to employ more sophisticated processes and rules that consider the three-dimensional changes in motion, shape, and structure of the moving object.

The image sequence analysis discussed includes three main stages: static, incremental, and global. Most of the problems encountered at the incremental and global stages are due to errors committed by the low level processes. The latter are responsible for the segmentation and description of the cell in each frame of the sequence. Improving the performance of the low level processes will solve most of these problems. For example, we are employing simple

segmentation methods based on histogram thresholding techniques and filtering operations that utilize data resulting from the analysis of previous frames and constraint knowledge pertaining to the cell structure. The result of the segmentation may be improved if we employ more sophisticated techniques that utilize general knowledge about lines, regions, areas, and textures, such as for example, the system reported by Levine and Nazif [Levine and Nazif, 82].

## 9.5 CONCLUSION

The structural changes in the cell morphology that occur during locomotion have not been previously reported in the literature. Furthermore, in spite of the importance and great interest in understanding the role that the cell membrane plays in the locomotion, there is no existing method for quantifying and analyzing the observable changes in the membrane shape. In this research, we have developed an image interpretation system capable of quantifying, analyzing, and describing the structural changes in the morphology of a moving cell.

The system has successfully provided all the quantification, description, and characterization information whereby basic questions pertaining to the cell's dynamic behaviour can be answered. This study might provide clues to the nature and distribution of "receptors" on or within the membrane which would be a vital link in the

interaction between the external factors and cell internal processes. Also, it might lead to the understanding of the roles the cell membrane plays in the mechanisms which regulate the social behaviour of the cell.

# DESCRIPTION (9.1)

## PSEUDOPOD CHARACTERIZATION AND DESCRIPTION

Classification of different pseudopods into five categories according to their domination of the cell locomotion. The following characterization is obtained by visual observation.

| PSEUDOPOD NUMBER | CATEGORY NUMBER | DOMINATION DESCRIPTION |
|---|---|---|
| 13 | 2 | SLIGHTLY DOMINANT |
| 10 | 3 | ALMOST DOMINANT |
| 7 | 3 | ALMOST DOMINANT |
| 37 | 4 | DOMINANT |
| 32 | 1 | NOT DOMINANT |
| 51 | 5 | VERY DOMINANT |
| 58 | 1 | NOT DOMINANT |
| 60 | 5 | VERY DOMINANT |
| 59 | 2 | SLIGHTLY DOMINANT |
| 80 | 1 | NOT DOMINANT |
| 91 | 5 | VERY DOMINANT |
| 92 | 5 | VERY DOMINANT |
| 108 | 5 | VERY DOMINANT |
| 150 | - | UNDEFINED |
| 143 | - | UNDEFINED |
| 158 | - | UNDEFINED |
| 177 | - | UNDEFINED |
| 176 | - | UNDEFINED |
| 185 | - | UNDEFINED |
| 183 | - | UNDEFINED |
| 222 | - | UNDEFINED |

# DESCRIPTION (9.2)

## PSEUDOPOD CHARACTERIZATION AND DESCRIPTION

The following is the result of comparing the visual pseudopod characterizations with those generated automatically by the system, using two vectors parameters.

| PSEUDOPOD NUMBER | VISUAL CHARACTERIZATION | AUTOMATIC CHARACTERIZATION | ERROR |
|---|---|---|---|
| 13 | SLIGHTLY DOMINANT | NOT DOMINANT | - 1 |
| 10 | ALMOST DOMINANT | DOMINANT | + 1 |
| 7 | ALMOST DOMINANT | DOMINANT | + 1 |
| 37 | DOMINANT | VERY DOMINANT | + 1 |
| 32 | NOT DOMINANT | NOT DOMINANT | 0 |
| 51 | VERY DOMINANT | VERY DOMINANT | 0 |
| 58 | NOT DOMINANT | ALMOST DOMINANT | + 2 |
| 60 | VERY DOMINANT | VERY DOMINANT | 0 |
| 59 | SLIGHTLY DOMINANT | SLIGHTLY DOMINANT | 0 |
| 80 | SLIGHTLY DOMINANT | DOMINANT | + 2 |
| 91 | NOT DOMINANT | DOMINANT | + 3 |
| 92 | VERY DOMINANT | VERY DOMINANT | 0 |
| 108 | VERY DOMINANT | VERY DOMINANT | 0 |
| 150 | UNDEFINED | VERY DOMINANT | ? |
| 143 | UNDEFINED | NOT DOMINANT | ? |
| 158 | UNDEFINED | VERY DOMINANT | ? |
| 177 | UNDEFINED | NOT DOMINANT | ? |
| 176 | UNDEFINED | ALMOST DOMINANT | ? |
| 185 | UNDEFINED | VERY DOMINANT | ? |
| 183 | UNDEFINED | SLIGHTLY DOMINANT | ? |
| 222 | UNDEFINED | VERY DOMINANT | ? |

TOTAL PERCENTAGE ERROR = 18 %

Note: The UNDEFINED pseudopods are those that could not be characterized visually, and they are not included in the computation of the total percentage error.

# DESCRIPTION (9.3)

## PSEUDOPOD CHARACTERIZATION AND DESCRIPTION

The following is the result of comparing the visual pseudopod characterizations with those generated automatically by the system, using three vector parameters.

| PSEUDOPOD NUMBER | VISUAL CHARACTERIZATION | AUTOMATIC CHARACTERIZATION | ERROR |
|---|---|---|---|
| 13 | SLIGHTLY DOMINANT | NOT DOMINANT | - 1 |
| 10 | ALMOST DOMINANT | DOMINANT | + 1 |
| 7 | ALMOST DOMINANT | DOMINANT | + 1 |
| 37 | DOMINANT | DOMINANT | 0 |
| 32 | NOT DOMINANT | NOT DOMINANT | 0 |
| 51 | VERY DOMINANT | VERY DOMINANT | 0 |
| 58 | NOT DOMINANT | NOT DOMINANT | 0 |
| 60 | VERY DOMINANT | DOMINANT | - 1 |
| 59 | SLIGHTLY DOMINANT | NOT DOMINANT | - 1 |
| 80 | SLIGHTLY DOMINANT | SLIGHTLY DOMINANT | 0 |
| 91 | NOT DOMINANT | NOT DOMINANT | 0 |
| 92 | VERY DOMINANT | VERY DOMINANT | 0 |
| 108 | VERY DOMINANT | DOMINANT | - 1 |
| 150 | UNDEFINED | VERY DOMINANT | ? |
| 143 | UNDEFINED | NOT DOMINANT | ? |
| 158 | UNDEFINED | VERY DOMINANT | ? |
| 177 | UNDEFINED | NOT DOMINANT | ? |
| 176 | UNDEFINED | ALMOST DOMINANT | ? |
| 185 | UNDEFINED | VERY DOMINANT | ? |
| 183 | UNDEFINED | SLIGHTLY DOMINANT | ? |
| 222 | UNDEFINED | VERY DOMINANT | ? |

TOTAL PERCENTAGE ERROR = 10 %

Note: The UNDEFINED pseudopods are those that could not be characterized visually, and they are not included in the computation of the total percentage error.

# APPENDIX (A)
# ANGLE AND SIDE REGULARITY

Regularity is a shape property that humans have always used to describe the shape of different objects. This property has not yet attracted much attenstion as a shape descriptor in the computer vision literature. In order to make use of it, we can determine the irregularity of the polygon approximation of an object by comparing it to a perfectly regular polygon. The measure is based on two criteria, angles and sides. Definitions and mathematical formulas for computing each are given in Section 4.5. The following is a short proof of these formulas, equations (4.26-4.29) inclusive.

## ANGLE REGULARITY

In this measure, the angles of a polygon are used as a criterion for measuring its regularity. Thus, the shape of an object can be described as perfectly regular, if all the angles of its polygon approximation are equal. Examples are squares, rectangles, pentagons, and hexagons. For a polygon with n verticies a1, a2, a3,..., ai,..., an, the sum of the internal angles (An) is:

$$An = \sum_{i=1}^{i=n} ai = (n-2) \cdot 180. \qquad (A.1)$$

In the case of a polygon with regular angles, we have

$$a1 = a2 = a3 = \ldots = ai = \ldots an = A,$$

and $A = (n-2).180 / n.$ (A.2)

The irregularity of a polygon representing a given shape can be defined based on a measurement of the sum of the differences between its angles and a regular one having the same number of vertices. This difference sum AD can be computed as:

$$AD = \left[ \sum_{i=1}^{i=n} (ai - A)^2 \right]^{1/2} \quad \text{(A.3)}$$

Thus, AD = 0 for a perfectly regular polygon, and the higher the value of AD, the more irregular it is. This value can be normalized to range between 0 and 1, where 0 corresponds to the pefectly regular shape, and 1 to the most irregular one. In this case, a normalization factor Ka can be used to obtain a measure for the Angle Regularity (AR), as:

$$AR = AD / Ka, \qquad 0 < AR < 1 \quad \text{(A.4)}$$

To determine the value of Ka, which normalizes AR, Ka should equal the maximum possible value of AD for the different polygons that having the same number of vertices. Thus, set

$$Ka = ADmax , \quad \text{(A.5)}$$

where ADmax is the sum of the differences between the angles of perfectly regular polygon and the corresponding one which has the same number of vertices and the most irregular angles. Figure (A.1a) shows examples of the following

perfectly regular polygons: equilateral triangle, square, and pentagon. Figure (A.1b) shows the corresponding polygons that have the same number of vertices and most irregular angles. From these figures, and by using equation (A.3), the value of ADmax can be determined as function of the number of vertices (n) for polygons with even and odd number of verticies, as follows:

POLYGONS WITH EVEN NUMBER OF VERTICIES

$$ADmax = (n/2 + 1)\,A + (n/2 - 1)\,(360 - A)$$

$$= n/2\,A + A + (n/2 - 1)\,360 - (n/2 - 1)\,A$$

$$= n/2\,A + A + (n - 2)\,180 - n/2\,A + A$$

$$ADmax = 2A + (n - 2)\,180 \qquad (A.6)$$

Substituting from equation (A.2) into (A.6), we obtain

$$ADmax = A(n+2)\,. \qquad (A.7)$$

POLYGONS WITH ODD NUMBER OF VERTICIES

$$ADmax = \left(\frac{n-1}{2} + 1\right) A + \left(\frac{n-1}{2} - 1\right)(360 - A) + (180 - A)$$

$$= \left(\frac{n+1}{2}\right) A + \left(\frac{n-3}{2}\right)(360 - A) + (180 - A)$$

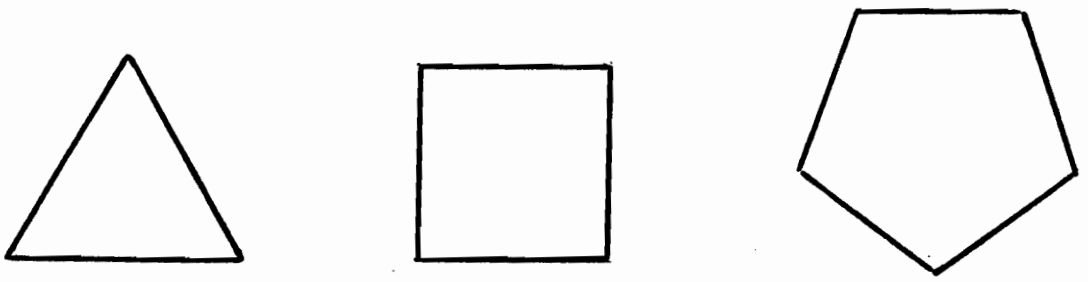
$$= \left(\frac{n+1}{2}\right) A + \left(\frac{n-3}{2}\right)(360) - \left(\frac{n-3}{2}\right)(A) + (180 - A)$$

$$= \left(\frac{n+1}{2} + \frac{n-3}{2}\right)(A) + \left(\frac{n-3}{2}\right)(360) + (180 - A)$$

$$= A + (n - 2)\,180 \qquad (A.8)$$

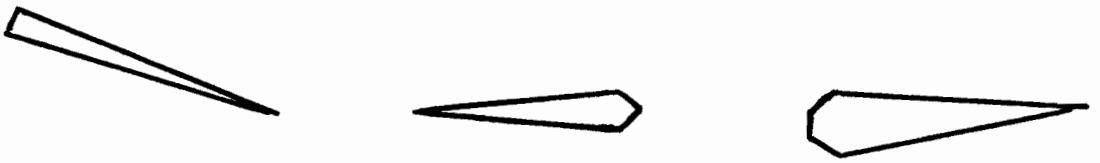
Substituting from equation (A.2) into (A.8), we obtain

$$ADmax = A(n+1)\,. \qquad (A.9)$$



**(a) Examples of perfectly regular polygons.**

**(b) Examples of polygons having the most irregular angles.**



**(c) Examples of polygons having the most irregular sides.**

*Figure(A.1) Perfectly regular polygon approximations with corresponding ones that have the most irregular angles and the sides.*

## SIDE REGULARITY

The lengths of the polygon sides are used in this measure as a criterion for determing irregularity. Thus, a polygon is described as perfectly regular if all the sides have equal length. For example, an equilateral triangle or a square. For a polygon with n sides l1, l2, l3,...., li, ...., ln, the perimeter P is computed as:

$$P = \sum_{i=1}^{i=n} li \,. \qquad \text{(B.1)}$$

In the case of a polygon with regular sides, we have

$$l1 = l2 = l3 = \ldots = li = \ldots ln = L,$$

$$\text{and} \qquad L = P / n \,. \qquad \text{(B.2)}$$

The irregularity of a polygon can be defined based on a comparison with a regular polygon approximation having the same number of sides and perimeter. Thus, the sum of the differences LD can be computed as:

$$LD = \left[ \sum_{i=1}^{i=n} (li - L)^2 \right]^{1/2} \qquad \text{(B.3)}$$

Note that LD = 0 for a polygon with regular sides, and the higher the value of LD, the more irregular the shape. In a similar fashion to that described above, the value of LD can be normalized between 0 and 1 to obtain a measure for the Side Regularity (SR), as:

$$SR = LD / Ks, \qquad 0 < SR < 1 \qquad (B.4)$$

and $$Ks = LDmax \qquad (B.5)$$

where LDmax is the sum of the differences between the lengths of a regular polygon and the corresponding one which has the same number of irregular sides. Figure (A.1c) shows corresponding polygons that have the same number of irregular sides for those shown in Figure (A.1a). From these figures, and by using equation (B.3), the value of LDmax can be determined as function of the number of sides (n) as follows:

$$LDmax = (n-2)\,L + (n\,L - 2\,L)$$

$$= n\,L - 2\,L + n\,L - 2\,L$$

$$LDmax = 2L(n-2) \qquad (B.6)$$

# APPENDIX (B)

# LABORATORY FACILITIES AND METHODS

## Blood Cell Preparation

Polymorphonuclear leucocytes (PMN) were prepared by pricking a finger and placing the drop of blood on a cverslip. After incubating for 25 minutes at 37'C in a moist chamber, the plsma clot was removed and the remaining red cells washed off with Hank's balanced salt solution [Boyarsky and Noble, 77] The adherent PMN were covered with Minimal Essential medium, Harpes buffered pM7.2 containing 20% fetal calf serum and the coversilp was placed on s standard 3" x 1" cover slide and the edges sealed with molten wax. For chemotaxis experiments a small sample of 'E. coli' was heat fixed to the coverslip prior to placing upon it the drop of blood. The locomotory trejectories of the PMN's were followed using a Bolex-Wild time-lapse unit at 2 frames per second. A wild M40 inverted microscope with phase contrast optics and 300x magnification was used.

## COMPUTATIONAL FACILITIES

The experimental work for quantifying and characterizing the dynamic behaviour of a moving cell is presently operating on the computational facilities of the Computer Vision and Graphics Laboratory (CVaGL), McGill University. The main processor of the laboratory is a DEC VAX 11/780 computer with a virtual memory that allows up to two million bytes of address space. The peripheral devices [illegible]

main processor via a UNIBUS interface include a GRINNELL Model GMR 27, 24 plane colour graphic television display and frame grabber. In addition to standard input terminals, a magnetic tape unit, a disc unit, and a line printer.

The GRINNELL also controls a Joystick for interactive communication with the display. A microprocessor interface is presently being added to the facility for control of a film advance unit. It provides complete software capability for advancing 16 mm cine film automatically or on a frame-by-frame basis. The image scanned by A COHU Model 4353 TV camera is captured via the GRINNELL Graphic Digitizer option at a rate of up to 30 frames per second.

The GRINNELL provides a 256X256, 64 gray level image intensity array which is stored in the GRINNELL video plane memory, accessable to the VAX. Under the command of the VAX, a microprocessor interface is used to control the film advance device, while the cine frames are digitized by the GRINNELL and stored on magnetic tape or disc.

The option of real time tracking and analysis is also possible by connecting the TV camera directly to the microscope and viewing live blood cells. This method is described in detail in [Levine and Youssef, 78, Knoll, 79].

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