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DIFFUSION CHANNEL SYSTEM FOR CONTROLLED ATMOSPHERE STORAGE OF SPINACH

by

Annie F.A. Chimphango
(Nee Jelenje)

November, 1996

A Thesis Submitted to the
Faculty of Graduate Studies and Research
in Partial Fulfilment of the Requirements of the Degree of
Master of Science

Department of Agricultural and Biosystem Engineering
McGill University, Macdonald Campus
QUEBEC, CANADA

DEDICATION

To my dear children, Madalitso, Anderson and Tawina
for depriving them of my care throughout the study period.
God Bless You!

ABSTRACT

Advanced research in Controlled/ Modified Atmosphere storage systems has shown that open channels of different lengths and cross sectional areas, connected to an air tight storage chamber, are able to maintain variable stable gas concentrations which could be near optimal concentrations for CA/MA storage of various commodities.

This study was geared towards assessing the suitability of the diffusion channels in maintaining a desired gas concentration for CA storage of spinach. Initially, the respiratory behaviour of spinach was studied in gas sealed chambers (replicated four times) stored at four different temperatures, 2°C, 8°C, 15°C and 23°C. The respiration rate of spinach was 20 $mgCO_2/kg.h$, 66 $mgCO_2/kg.h$, 163 $mgCO_2/kg.h$ and 271 $mgCO_2/kg.h$ for 2°C, 8°C, 15°C and 23°C, respectively. A model was developed based on principles of enzymatic kinetics which could reliably predict the respiration rate of spinach at any given storage temperature.

Two other sets of experiments were carried in a cold room set at 2°C. In one of the sets, spinach was stored for 30 days in 18 air tight chambers installed with diffusion channels of 0.006 m, 0.03 m, 0.07 m, 0.12 m, 0.18 m and 0.25 m in length and $4*10^{-6}$ m^2, $1.8*10^{-5}$ m^2 and $1.15*10^{-4}$ m^2 in cross sectional area. CO_2 produced by the respiring spinach was completely scrubbed from all the chambers using 250 g of lime ($Ca(OH)_2$). The O_2 steady state levels decreased with an increase in length and became more distinct as the cross sectional area of the channels increased. There was a great variation in the O_2 steady state levels with changes in the cross sectional area when the length of the diffusion channel was between 0.03m and 0.07 m. Spinach maintained colour and freshness in chambers where O_2 was in the range of 1 to 5 %. A diffusion channel of 0.18 m in length and $4*10^{-6}$ m^2 in cross sectional area maintained the best conditions for CA/ MA storage of spinach. A model was developed based on Fick's first law of molecular diffusion for predicting length of a diffusion channel which could maintain desired gas concentration for CA/MA storage of spinach.

In the second set, spinach was stored for 49 days in 18 air tight chambers which were split into 9 pairs. The chambers were installed with diffusion channels of 0.043 m, 0.11 m and 0.19 m in length and $9*10^{-6}$ m^2, $4*10^{-5}$ m^2 and $1.7*10^{-4}$ m^2 in cross sectional area. CO_2

was scrubbed using 250 g of lime from each chamber of one of the pairs. The O_2 steady state levels were high in CO_2 unscrubbed chambers. The CO_2 steady state levels increased with an increase in the length of the diffusion channels and became more distinct as the cross sectional area increased. The channels were able to maintain CO_2 and O_2 in variable ratios. A diffusion channel of 0.19 m in length and a cross sectional area of $9*10^{-6}$ m^2 maintained a 1:1 ratio of CO_2 to O_2 which was best in maintaining the quality of the spinach.

The diffusion channels have proved to be effective in maintaining desired gas concentration for CA storage of spinach. The shelf life of spinach could be extended by over 100% in diffusion channel CA/MA storage.

RÉSUMÉ

Des recherches poussées dans le domaine de l'entreposage sous atmosphère modifié ou contrôlé (AM/AC), ont démontré que des canaux de diffusion, de longueurs et de surfaces d'orifices différentes, rattachés à des chambres hermétiques, sont capables de maintenir stables les concentrations gazeuses pouvant se rapprocher des concentrations optimales recommandées pour l'entreposage AM/AC de différents produits frais périssables.

L'étude présentée ici, a été entreprise avec pour but de déterminer la capacité des canaux de diffusion de maintenir la concentration gazeuse désirée lors de l'entreposage AC d'épinards. Pour commencer, le comportement respiratoire des épinards a été étudié dans de petits contenants hermétiques (quatre réplicats) entreposés à quatre niveaux de température, 2°C, 8°C, 15°C et 23°C. Le taux de respiration des épinards était de 20 mg CO_2/kg·h, 66 mg CO_2/kg·h, 163 mg CO_2/kg·h, et 271 mg CO_2/kg·h à 2°C, 8°C, 15°C et 23°C respectivement. Un modèle a été développé, basé sur les principes des réactions enzymatiques qui peuvent prédire, de façon efficace, le taux de respiration des épinards à n'importe quelle température donnée.

Deux autres séries d'expériences ont été effectuées à une température d'entreposage de 2°C. Dans la première série, les épinards ont été entreposés pendant 30 jours dans 18 chambres AC hermétiques munies de canaux de diffusion de 0.006 m, 0.03 m, 0.07 m, 0.12 m, 0.18 m, et 0.25 m de longueur avec 4×10^{-6} m^2, 1.8×10^{-5} m^2, et 1.15×10^{-4} m^2 de surface des orifices. Le CO_2 produit durant la respiration des épinards a été entièrement lessivé de chacune des chambres avec l'utilisation de 250 g de chaux ($Ca(OH)_2$). Le niveau stationnaire de l'O_2 a diminué avec l'augmentation de la longueur des canaux et s'est démarqué avec une augmentation de la surface de l'orifice des canaux de diffusion. Le niveau stationnaire de l'O_2 a connu de grandes variations suivant le changement de la surface de l'orifice lorsque la longueur des canaux de diffusion était entre 0.03 m et 0.07 m. Les épinards ont conservé leur couleur et leur fraîcheur dans les chambres où l'O_2 a été maintenu entre 1 et 5%. Un canal de diffusion de 0.18 m de longueur ayant une surface d'orifice de 4×10^{-6} m^2 a maintenu les meilleurs conditions pour l'entreposage AC/AM des épinards. Un modèle a été développé, basé sur la première loi de Fick sur la diffusion moléculaire, pour la prédiction de la longueur nécessaire à un canal de diffusion afin

de maintenir les concentrations gazeuses voulues pour l'entreposage AC/AM des épinards.

Dans la deuxième série d'expériences, les épinards ont été entreposés pendant 49 jours dans 18 chambres AC hermétiques groupées en 9 paires. Les chambres ont été munies de canaux de diffusion de 0.043m, 0.11m et 0.19 m de longueur et 9×10^{-6} m^2, 4×10^{-5} m^2, et 1.7×10^{-4} m^2 de surface d'orifice. Le CO_2 a été lessivé d'une des deux chambres de chacune des paires avec l'utilisation de 250 g de chaux. Les niveaux stationnaires de l'O_2 se sont avérés élevés dans les chambres dont le CO_2 n'a pas été lessivé. Les niveaux stationnaires du CO_2 ont augmenté avec une augmentation de la longueur des canaux de diffusion et se sont démarqués avec une augmentation de la surface de l'orifice des canaux de diffusion. Les canaux ont réussi à maintenir les concentrations du CO_2 et de l'O_2 à différents ratios. Un canal de diffusion de 0.19m de longueur avec une surface d'orifice de 9×10^{-6} m^2, a maintenu un ratio de 1:1 du CO_2 par rapport à l'O_2 ce qui est considéré comme optimal pour la conservation de la qualité des épinards.

Les canaux de diffusion ont démontré être aptes à maintenir les concentrations gazières désirées pour l'entreposage AC des épinards. La durée de conservation des épinards pourrait être augmentée de 100% avec l'utilisation des canaux de diffusion en entreposage AC/AM.

ACKNOWLEDGEMENTS

The author wishes to express her profound gratitude to Dr. G.S.V. Raghavan, thesis supervisor and chair of the Agricultural and Biosystem Engineering Department, for his guidance and the support rendered throughout the study period. His care and encouragement were exceptional. The author is very grateful to the sponsors of Thomas and Willa Ferguson Smythe Fellowship and Natural Science and Engineering Research Council of Canada (NSERC) for the financial support which has enabled her to pursue higher studies in Postharvest Engineering of crops, a field which will have a greater impact on establishing a sustainable agricultural production system in Malawi. She is also very thankful to Prof. B.B. Chimphamba, Vice Chancellor of the University of Malawi, Dr. Z. Kasomekera, Principal of Bunda College of Agriculture, Malawi and Dr. D.G. Simango, Head of Agricultural Engineering Department, Bunda College of Agriculture, Malawi, for their support and encouragement.

The author is indebted to the following: Dr. C. Ratti, Mr Y. Gariépy and Mr M. Ramachandra for their technical and moral support throughout the study period. She also sincerely thanks Mr P. Alvo for the wonderful job done in editing the thesis and Ms. V. Orsat for the excellent translation of the abstract into French. The author acknowledges and appreciates the assistance rendered by Dr. S. Barrington, Ms. S. Gregus, S. Nagy and M. Gabriel, all of the Agricultural and Biosystem Engineering Department and the many friends and colleagues whose names do not appear on the list.

The author is also very grateful to her loving husband Mr. S.B. Chimphango, for his patience, care and encouragement during this hardest period. The going was tough but he persevered, God bless him! She also thank her parents and relatives for their support and prayers throughout the study period.

LIST OF SYMBOLS

a	Colour characteristic (hue)
A	Area (m^2)
A	Integration constant as in Equation (3.7)
a_1, a_2, a_3	Constants of an Equation
A_1, A_2	Integration constants in Arrhenius Equation
a,b,c,d,e,f	Letters assigned by Duncan Multiple Range Test
A_c	Cross sectional area (m^2)
A_m	Silicone membrane area (m^2)
b	Colour characteristic (chroma)
b_1, b_2, b_3	Constants of an Equation
c	Total gas concentration (mg/l or mol/m^3)
$C_2 H_4$	Ethylene molecule
C_{co2}	Concentration of Carbon dioxide (mg/l)
C_i	Inside gas concentration (mg/l)
C_o	Outside gas concentration (mg/l)
C_{o2}	Concentration of Oxygen (mg/l)
CO_2	Carbon dioxide molecule
$[CO_2]$	Carbon dioxide concentration (mg/l)
Cp	Specific heat capacity (J/kg.°C)
D	Diffusion coefficient (m^2/min)
$D_{O2\text{-}N2}$	Diffusivity of Oxygen in Nitrogen (m^2/s)
E_a	Activation energy (kJ)
H	Hydrogen atom
ΔH	Heat of respiration (J/mg CO_2)
ΔH^o	Standard Enthalpy of reaction (J/mol)

h_g	Heat transfer Coefficient ($W/m^2.°C$)
J	Mass flux ($g/m^2.h$)
K_1, K_2, K_3	Kinetic constants
K_i	Inhibition constant
K_m	Michaeli's Menten constant
L	Length of diffusion channel (m^2)
L	Colour characteristic (Value)
M	Mass of product (kg)
M_{CO2}	Mass of Carbon dioxide (mg)
M_{CO2ev}	Mass of Carbon dioxide evolved (mg)
m_{CO2} (t+Δt)	Mass of Carbon dioxide (mg) at time "(t+Δt)"
m_{CO2} (t)	Mass of Carbon dioxide (mg) at time "t"
M_{O2}	Mass of Oxygen (mg)
M_{O2cons}	Mass of Oxygen consumed (mg)
m_{O2} (t+Δt)	Mass of Oxygen (mg) at time "(t+Δt)"
m_{O2} (t)	Mass of Oxygen (mg) at time "t"
m_s	Mass of stored product (kg)
N_2	Nitrogen molecule
n_{O2}	Oxygen mass flux ($g/m^2.h$)
$[O_2]$	Oxygen concentration (mg/l)
O_2	Oxygen molecule
P_{CO2}	Permeability of membrane to CO_2 ($l/d.m^2.atm$)
Phy.	Physiological mass loss (%)
Q_{10}	Temperature gradient
r_{CO2}	Respiration rate ($mgCO_2/kg.h$)
r_{O2}	Respiration rate ($mgO_2/kg.h$)
r	Respiration rate ($mgCO_2/kg.h$ or $mgO_2/kg.h$)

R	Universal gas constant
RQ	Respiratory Quotient
RR	Respiration rate ($mgCO_2/kg.h$ or $mgO_2/kg.h$)
s	Substrate concentration (g/l)
S_1	Rate of product quality loss (unit/Day) at temperature t_1
S_2	Rate of product quality loss (unit/Day) at temperature t_2
T	Temperature (°C or °K)
Tr.	Trimming mass loss (%)
v	Rate of reaction
V_{CO2} ($\Delta t + t$)	Volume of Carbon dioxide (l) at time "$\Delta t+t$"
V_{CO2} (t)	Volume of Carbon dioxide (l) at time "t"
V_m	Maximum respiration rate (mg or ml/kg.h)
V	Maximum velocity
V_{O2} (t)	Volume of Oxygen at (l) time "t"
V_{O2} ($\Delta t + t$)	Volume of Oxygen at (l) time "$t+\Delta t$"
y_{O2}	Oxygen mole fraction
z	Spatial coordinate (m)

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ABBREVIATIONS

BNS	Brown Stain disease
CA	Controlled Atmosphere
ch #	Chamber number
Cham	Chamber
Conc	Concentration
CSA	Cross Sectional Area
DMRT	Duncan's Multiple Range Test
LPS	Low Pressure System
MA	Modified Atmosphere
RA	Regular Atmosphere
RH	Relative Humidity
PSA	Pressure Swing Adsorption System
SMS	Silicone Membrane System

CHAPTER I

INTRODUCTION

Fruits and vegetables are important nutritionally as well as economically world wide. However, their production is in most cases localised and seasonal. Their market price during the off seasons can increase two or threefold. The price increase, in addition to the cost of storage and handling, is greatly attributed to postharvest losses. This factor constitutes the major constraint in many fruit and vegetable industries. The physiological characteristics, mainly respiration and transpiration, make these commodities highly perishable and difficult to handle. The postharvest losses can be estimated up to 100%, particulary, in developing countries where handling facilities are very limited.

A lot of resources have been invested in developing improved varieties, installing irrigation facilities so as to increase the availability of fresh fruits and vegetables. However, the availability of these commodities can be tremendously improved by simply minimising the extent of postharvest losses. Strategically, the losses can be prevented at production, market and household levels. Drying, freezing and canning have been the common methods of preventing postharvest losses in fruits and vegetables. On the other hand, there has been an increasing demand for these commodities in their fresh state. The advantages of eating fresh fruits and vegetables are so numerous.

Cooling and refrigeration in regular gas atmospheres (RA), 0.03% CO_2, 21% O_2 and 78% N_2 are the most common techniques used in extending shelf life of many fresh produce. However, longer transit times, high energy costs and need to increase food supply has necessitated some technological improvements.

Advanced research in postharvest technology of fresh fruits and vegetables has shown that proper control of temperature, relative humidity and manipulation of storage gas composition in Modified/Controlled atmosphere (CA/MA) storage systems, in addition to harvesting at optimal maturity stage, minimization of injury

during handling and reduction in storage microbial infection, can successfully extend the shelf life and minimize the extent of postharvest losses (Raghavan and Gariépy, 1985; Kader, 1992a). The most commercially used technique for altering gas composition, so far, is the membrane system. This consists of a membrane of differential permeability to gas as part of the enclosure walls of an air tight storage chamber. Although the performance of the membrane systems is so outstanding, in addition to commodity diversity in response to CA/MA, the system has been associated with some technological and economical problems.

Following the idea of selective permeability of the membrane system is the concept of the diffusion channels. Basically, these are open channels which provide a path for diffusion of gases in and out of an air tight storage chamber. The amount of the gas diffusing into the chamber is related to the respiration characteristics of the stored product and the dimensions of the channel. This concept could lead to a much simpler, cheaper and flexible way of modifying the gas composition in the atmosphere surrounding a commodity. Currently, information on the use of the diffusion channels for CA/MA storage is scanty (Ratti et al., 1994). To achieve an adequate design a thorough understanding of the physiological and biological characteristics of the stored commodity and the functioning of the system under particular conditions is essential.

It is hypothesized that changing the dimensions of a diffusion channel can vary the gas composition in the storage chamber. Although precise control of the individual gas concentrations is envisaged as being difficult to achieve, it could be possible to reach a quasi-equilibrium at near optimal concentrations for CA storage of various commodities.

Objectives

The overall objective of carrying out the study was to assess the suitability

of the diffusion channel system in maintaining desired gas concentration for CA storage of spinach. The specific objectives were:

(a) To asses the effect of temperature on the respiration rate of spinach.

(b) To develop a model for predicting respiration rate of a given mass of spinach at any given storage temperature.

(c) To asses the effect of changing the length and cross sectional area of a diffusion channel on gas concentration, under steady state conditions in CA storage of spinach.

(d) To develop a model for predicting length of a diffusion channel which can maintain a desired O_2 concentration for storage of a known mass of spinach.

(e) To asses the effect of scrubbing CO_2 in the diffusion channel system on the steady state O_2 concentration.

(f) To asses quality of spinach after CA storage by the diffusion channel system.

CHAPTER II

LITERATURE REVIEW

2.1 Respiration Process

Fruits and vegetables go through three developmental stages: maturation, ripening and senescence (Burton, 1982). These developmental stages involve some chemical and physical changes which may lead to loss of optimal eating qualities if the crop is detached from the parent crop (Phan et al., 1975). A basic important fact in postharvest handling of crops is that, upon harvest, the crops are still living structures (Kays, 1991 and Wills et al., 1989) hence, the metabolic activities which were performed when attached to the parent crop continue. The major physiological activity in the postharvest life of a produce is respiration (Burgheimer et al., 1967; Kays, 1991). Respiration is described as an oxidative breakdown of starch, sugars and other organic compounds into simple molecules, such as carbon dioxide, water and other intermediates with subsequent production of energy, in the form of heat (Burton, 1982; Ryall and Pentzer, 1979; Wills et al., 1989 and Kader 1992a). Respiration is usually represented as oxidation of hexose sugars as follows (Phan et al., 1975):

$$C_6H_{12}O_6 + 6O_2 \ldots\ldots 6CO_2 + 6H_2O + Energy \qquad (2.1)$$

The rate of respiration is generally expressed in terms of O_2 consumed or CO_2 evolved per unit weight of produce per unit time (Bidwell, 1979). In some cases, it is expressed as heat liberated and substrate loss, as indicated by mass loss (Wills et al., 1981). However, it is rather difficult to accurately measure the heat liberated due to the many interrelated metabolic processes that occur in the produce at the same time, some of which utilise the respiratory energy. Similarly, expressing respiration rate in terms of substrate loss might not give reliable results

because, in some cases, relatively large rates of respiration over a short measurement period may result in just a small change in the total substrate (Kays, 1991). The other factor is that respiration involves metabolism of different substrates which differ in their degree of oxidation. The ratio of the moles of CO_2 evolved to moles of O_2 consumed is called the Respiratory Quotient (RQ) (Equation 2.2). The RQ provides a general indication of what type of substrate is being used in the respiration process. The value of the RQ is reported to vary with varying degrees of substrate oxidation (Phan et al., 1975; Bidwell, 1979; Wills et al., 1981).

$$RQ = \frac{M_{CO_{2_{ev}}}}{M_{O_{2_{cons}}}} \qquad (2.2)$$

Where:

RQ = respiratory quotient

M_{CO2ev} = mass of carbon dioxide evolved (mg)

M_{O2cons} = mass of oxygen consumed (mg)

Respiration is necessary to maintain vigour of the plant tissue and provide defence against spoilage in the post harvest life of the produce. The rate of respiration is an index of the metabolic turnover in the produce (Phan et al. 1975; Bidwell, 1979 and Kays, 1991) and is believed to be proportional to the rate of deterioration (Burton, 1982; Kader, 1992a). The higher the rate, the faster the deterioration rate because of the increase in the breakdown of the organic compounds stored in the harvested product. This process subsequently leads to accelerated senescence, loss of food value, reduced flavour and respiratory losses of carbon that represent loss of saleable weight (Wills et al., 1981). High respiration rates are therefore associated with short storage lives. To extend produce shelf life, the storage conditions should minimize the respiration rate.

Respiration is mostly influenced by temperature, relative humidity and atmospheric gas composition (Kays, 1991). Any changes in these factors may lead to subsequent changes in the internal environment (cells) of the stored produce (Phan et al., 1975). The changes occur because of alterations in the cell metabolic pathways. Ryall and Pentzer (1979), Plasse and Raghavan (1983), Raghavan and Gariépy (1985) and Shewfelt (1986) indicated that there could be a reduction in the respiration rate with subsequent reduction in the rate of product deterioration if the above mentioned factors are controlled to optimum levels.

2.1.1 **Temperature**

Temperature is probably the most important factor in the respiration process (Kays, 1991). Its effect on the respiration rate can be related to that on chemical reactions where the rate of reaction increases exponentially with an increase in temperature (Wills et al., 1981 and Burton, 1982). Any 10°C increase may result in a two or three fold increase in the rate of the reaction, mathematically expressed as Q_{10} (Bidwell, 1979 and Wills et al., 1981).

$$Q_{10} = \left(\frac{R_2}{R_1}\right)^{10/(t_2 - t_1)} \qquad (2.3)$$

R_1 and R_2 are rates of the reaction at temperature t_1 and t_2 respectively. It can also be expressed in terms of quality loss as follows

$$Q_{10} = \left(\frac{S_2}{S_1}\right)^{10/(t_2 - t_1)} \qquad (2.4)$$

where S_1 and S_2 are rates of quality loss at temperature t_1 and t_2 respectively. Q_{10} values for fresh fruits and vegetables are normally given within a specified range of temperatures and the concept may not be valid at low temperatures for chilling sensitive commodities (Shewfelt, 1986). A large Q_{10} indicates that the intensity of the process is high (Wills et al., 1981). Knowledge of the Q_{10} can therefore assist

in deducing the shelf life of the product (Wills et al., 1981).

Comprehensive tables of respiration rates of most fruits and vegetables at different temperatures have been compiled (Ryall and Lipton, 1979; Burton, 1982; Ryall and Pentzer, 1979 and Hardenburg et al., 1990).

Generally fruits and vegetables store longer at low temperatures (Wills et al 1981, Zagory and Kader, 1988). However, commodities do not respond in the same way to such temperatures. Most fruits and vegetables of the tropical and sub tropical origin suffer freezing or chilling injury if stored at low temperatures (Brown, 1979; Shewfelt, 1986 and Kader, 1992b) which eventually aggravates the respiration process, with subsequent deterioration and loss of value of the product. Freezing points of some commodities have been compiled (Kays, 1991).

Temperature also influences the water vapour deficit between the commodity and its microenvironment (Van de Berg and Lentz, 1978; Grierson and Wardowski, 1978 and Peleg, 1985). Depending on the direction of the deficit, water may diffuse out of the product (transpiration) or condense on the product surface (sweating). The former leads to desiccation, shrivelling, loss of saleable weight and limpness, therefore, reducing the market value of the product. The latter may result in splitting of the plant tissues which makes the product more susceptible to microbial infection. As a consequence, senescence is facilitated (Zagory and Kader, 1988 and Kays, 1991).

Proper management of temperature in storage of fresh produce would be the most important part of good postharvest handling. The effects of temperature can be minimized by pre-cooling the products prior to storage (Shewfelt, 1986 and Mitchell, 1992). For highly respiring commodities, pre-cooling may be the single most important step in extending postharvest life (Wills et al., 1981 and Shewfelt, 1986). Effective cooling requires knowledge of thermal properties of the product such as specific heat capacity and thermal heat conductivity. Mohsenin (1980) and

Kays (1991) indicated that heat transfer in the harvested product normally involves three modes of heat transfer: convection, conduction and radiation. Each of these is a function of the characteristics of the product in terms of shape, size, nature of the surface and the internal chemical composition, in addition to the environmental factors (Mohsenin, 1980). Most products of high surface to volume ratio have greater potential to transfer heat because they have thin boundary layers (Mohsenin, 1980 and Kays, 1991). When, how and where to precool also depends on the characteristics of the product and its shelf life requirements. Such methods as forced air-cooling, hydro-cooling, hydroair-cooling (fine mist spray combined with forced air cooling), and vacuum cooling are normally used. Detailed discussions on precooling are available (Ryall and Pentzer, 1979; Kays, 1991 and Kasmire and Thompson, 1992).

The impact of temperature could also be minimized by varying the gas composition of the environment. Hardenburg et al. (1990) indicated that reduced chilling injury in some crops has been associated with elevated CO_2.

2.1.2 **Relative Humidity**

Relative humidity (RH) is another important environmental factor that affects the postharvest life of many fruits and vegetables (Grierson and Wardowski, 1978 and Shewfelt, 1986). It plays an important role in the quality of produce by influencing moisture loss. In some harvested produce, minor water losses may result in poor texture of the product. The loss of only 5-10% of the product weight as moisture is reported to render a wide range of products unsaleable. Maximum acceptable loss of water for some fruits and vegetables are available (Table A-1) (Kays, 1991).

RH and temperature are inextricably linked. Moisture loss from produce can be related to evaporation process and is proportional to the partial water vapour pressure gradient between the saturated internal atmosphere of the produce and the

less saturated surrounding atmosphere. The partial water vapour pressure gradient is normally influenced by temperature (Kays, 1991). The RH therefore indicates the amount of water vapour in the air as a percentage of the maximum amount of water air can hold at a specified temperature (Peleg, 1985 and Kays, 1991). Increasing the temperature at constant RH, results in a greater water vapour gradient which forces water to diffuse out of the product (Grierson and Wardowski, 1978). However, the impact of high temperature is cushioned by saturated conditions of the storage room (Pantastico et al., 1975).

The effect of RH on harvested produce also varies depending on the characteristics of the produce and environmental factors other than temperature. Commodities with high surface to volume ratio tend to have higher transpiration rates while those with thick cuticles transpire less (Kays, 1991). Low RH accompanied by high air velocity also facilitates moisture loss from produce (Shewfelt, 1986).

Ideally, humidity close to saturation results in zero moisture loss. Gariépy and Raghavan (1983) and Kader (1992b) reported that those saturated conditions can be achieved by humidifying systems. High humidity storage systems such as Jacketed, Filacell and plastic liners have been developed in North America (Van den Berg and Lentz, 1978; Ryall and Pentzer, 1979; Raghavan et al., 1980 and Raghavan and Gariépy, 1985). Experimental results have shown that the storage life of produce can be extended by 50% at RH of 90-100%. Carrots, parsnips, and rutabagas stored well at RH of 98-100% (Van den Berg and Lentz 1978; Raghavan et al., 1984). As noted by Shewfelt (1986), cherries and lemons had their shelf life extended by a week and a month, respectively, when stored in near-saturated storage. Some physiological disorders such as red blotch in lemons, stem-end rind breakdown in oranges and puffiness in tangerines may also be minimised by storing the products in plastic bags in which air is nearly saturated (Pantastico et al., 1975).

Although the advantages of the high relative humidity storage are so numerous, such systems are expensive and difficult to manage (Raghavan and Gariépy, 1985). Furthermore, high RH sometimes induces pathogenic infection rather than extending the storage life (Gariépy and Raghavan, 1983). High relative humidity may also result in splitting of the tissues in the produce (Kays, 1991). In view of these technological shortfalls, the performance of high RH storage systems could be improved by using it in combination with other systems.

2.1.3 **Atmospheric Gas Composition**

Based on the fact that respiration is a partly reversible chemical reaction, it is also influenced by concentration of reactants (O_2) and accumulation of the products (CO_2) (Burton, 1974 and Cornish-Bowden, 1979). Equation 2.1 suggests that if oxygen concentration is reduced and/or carbon dioxide augmented, the respiration rate should decrease, thereby extending postharvest life of the produce (Platenius, 1943; Pflug and Dewey, 1957; Burgheimer et al., 1967; Burton, 1974 and Raghavan and Gariépy, 1993). This is the basis for the establishment and maintenance of optimal gas composition in Modified Atmosphere storage systems. In these technologies, the gas concentrations may be altered passively by the combined effects of the respiring produce and selective gas diffusion through a storage enclosure or by an active system where gas is pulled out of the storage room and replaced with gas of desired concentration which is constantly monitored (Shewfelt, 1986 and Zagory and Kader, 1988).

2.1.3.1 **Controlled Atmosphere**

Precise control of the respiratory gases in MA leads to Controlled Atmosphere (CA). These technologies, though not in a sophisticated manner, have been in use for some decades (Wills et al., 1981 and Kays, 1991). The systems became the most important innovation since the introduction of mechanical refrigeration (Do and Salunkhe, 1975). Though research on CA started as early as

1819 by Berard followed by Nyce in 1860, cited by Kays (1991), little interest was generated among the fruit and vegetable growers. It was the classic work of Kidd and West in 1926 that stimulated commercial use of CA conditions for storage of fruits (Smock, 1979 and Wills et al., 1981). In Canada, research on CA storage is recorded as early as 1933 (Ryall and Pentzer, 1979 and Thorne, 1983). Since then, CA storage has been used for commercial storage of fruits and some vegetables such as: apples, pears and cabbage (Meheriuk, 1985; Richardson, 1985; Smock, 1979; Gariépy et al., 1986). The adoption of this technology was enhanced by the seasonality in the production of the commodities and the requirement for produce to be transported longer distances from place of production to market places (Gariépy et al., 1986; Eskin, 1989; Barth, 1993). The characteristic advantages of the systems have resulted in extended marketing season for fruits and vegetables which was once limited to a few months (Gariépy et al., 1984b). Such systems have favoured high economic returns and wide distribution of the produce (Van den Burg and Lentz, 1966; Pendergrass et al., 1976; Dilley, 1983 and Beaudry and Gran, 1993). Vegetables such as cabbage, can now be stored for over 8 months (Isenberg and Sayles, 1969; Bohling and Hansen,1977 and Raghavan et al., 1984). Fruits such as apples are also reported to store over 10 months (Ginsburg et al., 1982). Minimally processed mixed vegetables, whose shelf life is deemed to be too short, can also be stored for longer periods than in RA (Cantwell, 1992 and Lee et al., 1996).

It has been recognised by many researchers that the ability of the CA to maintain quality of produce for longer periods is because of its influence on some metabolic processes. Respiration has been found to be retarded by CA storage (Kader, 1992b). Platenius (1943) found that lowering O_2 concentration to about 1.2% reduced the respiration rate of asparagus by 55%. Pflug and Dewey (1957) and Hardenburg et al. (1990) also indicated that respiratory activity of apples was

slowed down in CA which resulted in a shelf life twice that of the RA. Respiration retardation by CA for other commodities has also been reported (Kasmire et al., 1974 and Do and Salunkhe, 1975).

CA also delays the onset of the respiratory peak in climacteric fruits (Ulrich, 1975). Consequently, there has been extension of storage life for such fruits as bartlett pears and bananas. Wills et al. (1981) reported a twelvefold increase in the storage life of bananas when stored at 5% CO_2, 3%O_2 and 92%N_2. The increase is even significant with O_2 level of approximately 8% and/or CO_2 above 1% and is attributed to the reduction in natural production and sensitivity to ethylene by the bananas (Do and Salunkhe, 1975; Kader, 1992b). Sub-atmospheric pressure conditions in CA storage have been reported to increase the shelf life of bananas. For instance, a regular atmosphere of 760 mmHg keeps bananas in good quality for only 30 days whereas with an atmosphere of 100 mmHg, bananas can stay on shelf for 120 days (Raghavan and Orsat, 1995).

Reduction in pectin hydrolysis for CA stored commodities has also been reported. Apples and pears stored in CA had less pectin breakdown than those in RA (Do and Salunkhe, 1975; Wills et al., 1981 and Peleg, 1985), so much so that firm texture is retained for longer periods. Improved retention of green colour has also been observed in CA stored commodities. Cabbage stored in CA retained green colour than in RA (Gariépy et al., 1984a). Such results are also true for broccoli (Lebermann et al., 1968; Lipton and Harris, 1974; Makhalouf et al., 1989 and Ramachandra, 1995) and lettuce (Singh et al., 1972). A contrasting effect however, has been observed in potatoes where greening due to exposure to light is prevented by storing tubers in an atmosphere of 15% CO_2 (Wills et al., 1981). Jonathan spot, a disorder in apples, can completely be prevented by CA storage where as little as 2% of CO_2 is available (Wills et al., 1981).

Importance of CA storage is also realised in minimizing loss of some

organic acids and vitamins. There was retention of total organic acids in carrots stored in 10% CO_2 (Do and Salunkhe, 1975). However, the effect of CA conditions on acid loss is not consistent for all the commodities. Acid loss is reported to have been accelerated in CA stored asparagus (Wills et al., 1981).

The CA conditions are beneficial not only because of direct changes in the metabolism of the produce per se, but also because of the imposition of stress injury or death of pathogenic organisms and insects that could otherwise cause or aggravate infection with subsequent deterioration of the produce. Kader (1992b) reported that elevated CO_2 (10-15%), significantly inhibits development of Botrytis rot on strawberries and cherries. Decay, pink rib, butt discolouration and russet spotting in CA stored lettuce were minimal (Singh et al., 1972 and Wills et al., 1981). Molding and rotting were reported to be less in CA stored cabbage than in RA (Gariépy et al., 1984a). Grey speck disease in cabbage is reported to have been suppressed by CA conditions (Berard, 1985). Leaf rollers insects in apples are also reported to have been completely eliminated under standard CA, 3% O_2 and 3% CO_2 (Klag, 1985). However, standard CA conditions are not adequate enough to get rid of the most problematic insects in fresh fruits and vegetables, such as fruit fly (Mitchell and Kader, 1992). Mitchell and Kader (1992) also indicated that such insects could be controlled by long exposure to limited O_2 of about 0.5% and CO_2 of more than 70%. Use of CA in eradicating insect infestations could be more appropriate for such crops as strawberries, pears, apples and nectarines which are able to withstand such high levels of CO_2 for relatively longer periods. Furthermore, it could be a way of eradicating Western flower thrips which attack strawberries (Klag, 1985 and Mitchell and Kader 1992). In actual fact, some investigators have reported that 100% mortality was observed in control of Western flower thrips when exposed to CO_2 levels as high as 40-100% and oxygen levels as low as 0.25% for a period range of 48 hours (Klag, 1985).

Such high carbon dioxide levels could be used for quarantine treatment of fruits and vegetables and possibly minimize use of chemicals (Mitchell and Kader, 1992). However, some logistical problems are expected especially when fruits or vegetables are handled in mixtures. Longer carbon dioxide exposures and concentrations may not be favoured by the other commodities (Ulrich, 1975 and Wills et al., 1981). More importantly, it might be a healthy hazard to humans working in such an environment. The success in control of pathogenic micro-organisms and insects by CA conditions, basically lies on the response of the produce and also the sensitivity of the pathogen/insect to the CA conditions generated (El-Goorani and Sommer, 1981).

CA has proven to be effective in minimising moisture loss, thus maintaining a firm and succulent texture of the commodities (Singh et al., 1972). Relative humidity of close to saturation has been reported in CA (Gariépy et al., 1984b). Claims have also been made of the possibility of using CA in reducing the extent of chilling injury in some produce (Wills et al., 1981 and Kader, 1992b). Not much has been done to confirm such results. Pantastico et al. (1975) indicated that an atmosphere of 7% O_2 is optimal in preventing chilling injury in most tropical and subtropical fruits and vegetables.

Important to note is that the striking performance of the CA storage systems rely on good temperature management (Gariépy et al., 1988). This is probably the reason why CA storage systems are rarely independent of refrigeration systems (Do and Salunkhe, 1975).

However, the response of commodities to CA, varies. Some adverse effects and toxic substances have been associated with the CA conditions. Particulary notable are: undesirable changes in flavour/aroma and offensive odours (Kays, 1991). Such odours have been reported when opening CA storage containers of broccoli (Kasmire et al., 1974). A deleterious effect on flavour has also been

reported for raspberries and strawberries stored in O_2 levels of 3% (Burton, 1982). Spinach was found to have an unacceptable flavour after one week of storage in 13% CO_2 (McGill et al., 1966). Tissue browning (core-flush) and breakdown in apples are also some of the disorders attributed to CA storage (Ulrich, 1975). Some cultivars of lettuce suffer brown stain (BNS) in CA storage (Isenberg, 1979). Worthy to note is also that achrolophyllous tissues have been found to be more susceptible to CA conditions than chrollophylous tissues (Isenberg, 1979). Kader (1992b) classified fruits and vegetables according to their tolerance to CA gas levels, O_2 and CO_2 as shown in Table 2.1 and 2.2 respectively.

There are several techniques of achieving CA conditions. Currently documented are: oxygen control, carbon dioxide control, hypobaric and membrane systems (Dilley, 1983; Gariépy and Raghavan, 1985; Kader, 1992b; Gariépy et al., 1984a; and Wills et al., 1989) and to some extent, Diffusion Channels (Baugerod, 1980; Ratti et al., 1994 and Ramachandra et al., 1995).

2.1.3.1a **Oxygen Control System**

CA establishment by respiration (Passive CA) is fairly slow (Thompson, 1992). Therefore, for product quality to be maintained for longer periods, oxygen has to be reduced to the desired levels at a faster rate. Several ways have been devised to quickly and reliably reduce the atmospheric oxygen level of 21% to a desired storage range of 1-3% (Bartsch, 1992). One such method is by purging the storage chambers with an inert gas or nitrogen (Smock. 1979 and Kader, 1992b). Special catalytic burners such as the Smit Oxydrain machine, are available, which convert gaseous fuels such as Ammonia into N_2 and H_2O and use the N_2 to flush out O_2 from the storage room, through Oxy-reduction process (Smock, 1979; Bartsch, 1992 and Thompson, 1992). Desired O_2 levels can also be achieved by recirculating system where air is recirculated from CA room into a generator and back into the store room with low oxygen (Wills et al., 1989 and Bartsch, 1992).

Table 2.1 Fruits and Vegetables Classified According to Low O_2 Tolerance

Minimum O_2 Tolerated (%)	Commodities
0.5	Tree nuts, dried fruits and vegetables
1.0	Some cultivars of apples and pears, broccoli, mushroom, garlic, onion, most cut or sliced (minimally processed) fruits and vegetables
2.0	Most cultivars of apples and pears, kiwifruit, apricot, cherry, nectarine, peach, plum, straw berry, papaya, pineapple, olive, cantaloupe, sweet corn, green bean, celery, lettuce, cabbage, cauliflower, brussels sprouts
3.0	Avocado, persimmon, tomato, pepper, cucumber, artichoke
5.0	Citrus fruits, green pea, asparagus, potato, sweet potato

Source: Kader (1992b)

Other methods include molecular sieves (Pressure Swing Adsorption Systems or PSA) and purging with exhaust of a fossil fuel burn (Thompson, 1992).

2.1.3.1b **Carbon Dioxide Control System**

Respiration causes carbon dioxide levels to rise well above the required levels. Methods available to scrub off the excess CO_2 include water and chemical scrubbers such as Sodium Hydroxide, Potassium Carbonate, Ethanolamine and

Table 2.2 Fruits and Vegetables Classified According to Elevated CO_2 Tolerance

Minimum CO_2 Tolerated (%)	Commodities
2	Apple (Golden Delicious), Asian Pear, European pear, apricot, grape olive, tomato, pepper (sweet), lettuce, endive, Chinese cabbage, celery, artichoke, sweet potato
5	Apple (most cultivars), peach, nectarine, plum, orange, avocado,banana, mango, papaya kiwifruit, cranberry, pea, pepper (chili), eggplant, cauliflower, cabbage, Brussels sprouts, radish, carrots
10	Grape fruit, lemon, lime, persimmon, pineapple cucumber, summer squash, snap bean, okra, asparagus, broccoli, parsley, leek, green onion, dry onion, garlic, potato
15	Strawberry, raspberry, blackberry, blueberry cherry, fig, cantaloupe, sweetcorn, mushroom, spinach, kale, swiss chard

Source: Kader (1992b)

hydrated lime ($Ca(OH)_2$) (Wills et al., 1981 and Kader, 1992b). However, most of these methods are expensive. The cheapest and the most effective is use of hydrated lime (Smock, 1979; Raghavan and Gariépy, 1985 and Bartsch, 1992). Thompson (1992) indicated that 1 to 3 kg of lime per 100 kg of produce is enough to maintain required levels of CO_2. Similar to oxygen control systems, molecular sieves are sometimes used to bring down carbon dioxide levels to desired levels (Bartsch, 1992 and Kader, 1992b). Widely used are also activated carbon dioxide adsorption systems such as use of activated charcoal (Thompson, 1992 and Wills et al., 1981) and to some extent brine water (Thompson, 1992). In cases where CO_2 is below the minimum required level, dry ice is used as a source of carbon dioxide (Kader, 1992b), simultaneously, providing a cooling effect.

2.1.3.1c **Hypobaric System**

This is a form of storage in which CA conditions are achieved by storing the produce under low pressure (Wills et al., 1981). Sometimes it is referred to as Low Pressure System (LPS) (Burg, 1973). It consists of a vented vacuum chamber which is continuously flushed with saturated air by a humidifier from the exterior and expelled by a vacuum pump, thus creating a slowly moving stream of air at low pressure (Burg and Kosson, 1983; El-Goorani and Sommer, 1981). Figures 2.1a & 2.1b are schematic diagrams of the hypobaric system and its associated water system, respectively.

Reducing the atmospheric pressure in the ambient environment results in a proportional decrease of the individual gas partial pressures as well as water vapour pressure (Brecht, 1980 and Kader, 1992b). Reducing atmospheric pressure from 100 kPa to 10 kPa, reduces the partial pressure of O_2 to about 2% (Wills et al., 1981; Burton, 1982 and Burg, 1973). It is therefore evident that reducing the pressure automatically creates desired gas levels in the storage room, regardless of the metabolic activity of the stored produce (Burg, 1973). Low

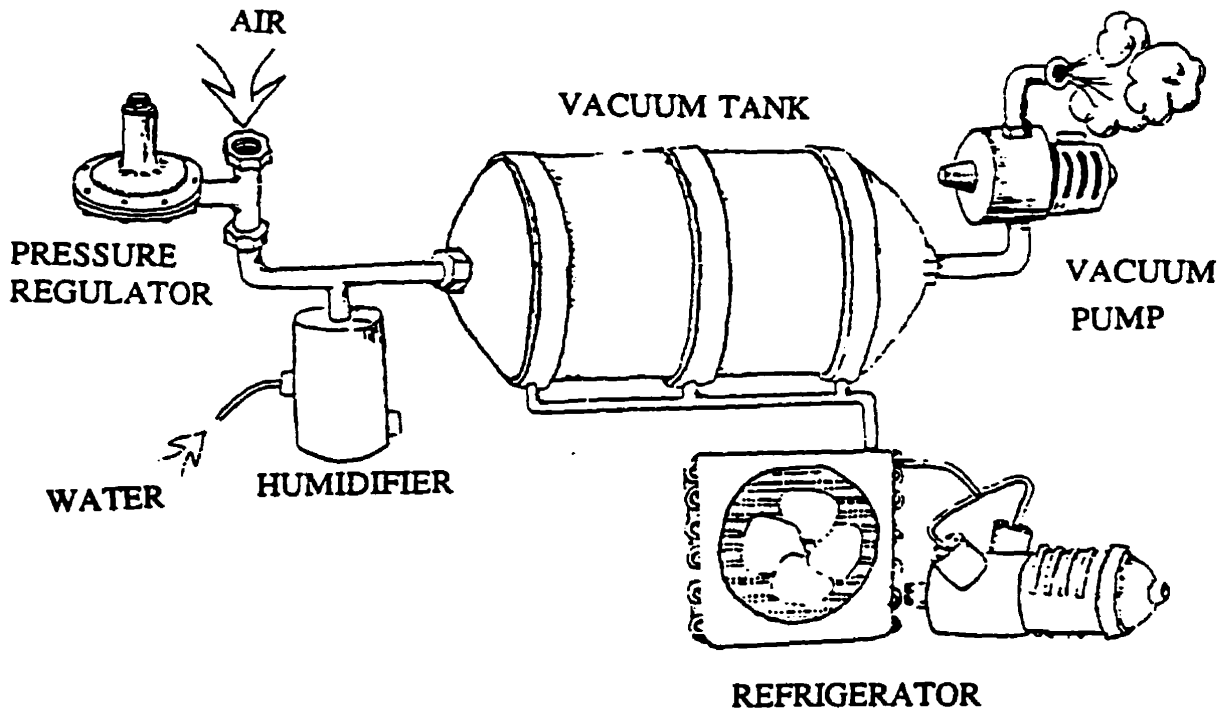

Figure 2.1a Schematic Diagram of a Hypobaric System

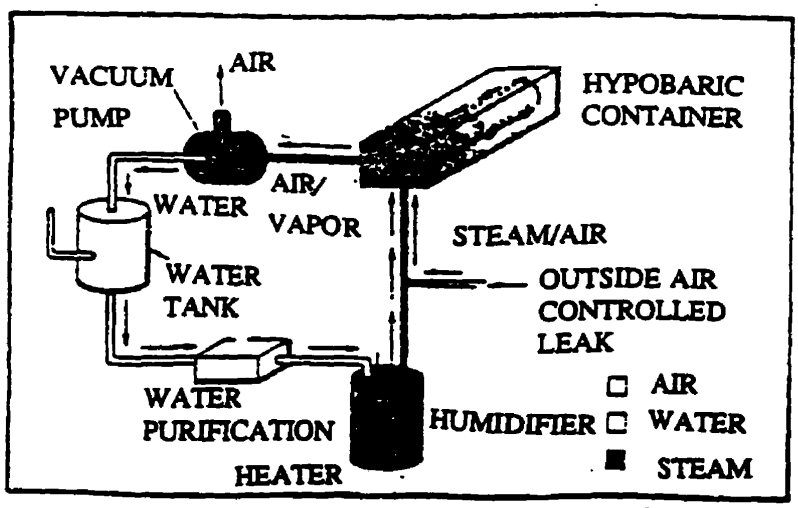

Figure 2.1b Vacuum/Water Subsystem

atmospheric pressure also reduces the hydrostatic pressure, thereby reducing cellular water activity (Burg and Kosson, 1983).

Since the diffusion coefficient of a gas is inversely proportional to pressure (Brecht, 1980 and Gebhart, 1993), beside lowering the concentration of gases, reducing pressure of the storage room subsequently increases the outward diffusion of gases to equilibrate the inside and outside pressure (Burg, 1973; Brecht, 1980; Jamieson, 1980; Burton, 1982 and Kays, 1991). Such gases as ethylene and the volatiles which would otherwise accumulate in the storage room can constantly diffuse out at a faster rate (Burg and Kosson, 1983). In this type of system, non-compatible products (for example, ethylene sensitive and non ethylene sensitive), could be handled together without much complication (Shewfelt, 1986). The low O_2 levels as well as the reduced pressure, in addition to altering the metabolic activities of the produce also alters metabolic activities of the pathogenic micro-organisms and insects. Burg (1973) and Lougheed et al. (1978) indicated the possibility of using LPS in suppressing postharvest pathogens.

Characteristic advantages of the hypobaric system, in addition to removal of pollutants and suppression of pathogens are:

(i) prevention of moisture loss from the product because of continuous pumping of saturated air into the storage room (Burg, 1973). Ideally, excessive water loss is expected because the low pressure lowers its boiling point (Kays, 1991). However, such an effect can enhance cooling of the produce through evaporation (Burg and Kosson, 1983).

(ii) The hypobaric system can be used as a vacuum cooler (Burg, 1973 and Raghavan and Gariépy, 1985).

(iii) The inside gas atmosphere is very stable, such that the storage chambers can be opened as many times as possible without altering the gas composition (Burg, 1973).

Though the hypobaric system seem to have so many advantages, its use is limited due to the cost of constructing a reinforced air tight refrigerated room (Raghavan and Gariépy, 1985 and Hardenburg et al., 1990). Furthermore, some commodities lose flavour, aroma and experience unsatisfactory ripening after storage (Burg, 1973). In some instances, functional levels of CO_2 are very difficult to achieve (Raghavan and Gariépy, 1985 and Kader, 1992b).

2.1.3.1d **Membrane System**

In view of the limitations in the use of the high relative humidity storage system, the hypobaric system and the other gas control systems mentioned previously in this Chapter, Marcellin and Leteinturier, (1966 and 1967) cited by (Smock, 1979), suggested the use of semi-permeable membranes to establish desired gas composition for CA storage of fresh fruits and vegetables. Further investigations have proven the effectiveness of such membranes (Plasse, 1987; Shewfelt, 1986 and Gariépy et al., 1988). Raghavan et al. (1984), Gariépy et al. (1984a) and Plasse (1987) reported of excellent results in quality of cabbage, celery and leeks, stored in air tight chambers with silicone membrane windows. The systems were capable of maintaining a gas composition of 5-6% CO_2 and 2-3% O_2 which stored cabbage well over 7 months at 1-1.5°C. Figure 2.2 illustrates the mechanism involved.

The systems are mainly characterised by the membrane's differential permeability to the respiratory gases (Gariépy et al., 1988). However, the membrane's functionality, in addition to the overall storage design characteristics also depends on the respiration rate of the stored commodity, the chemical and the physical characteristics of the respiratory gases (Plasse, 1987 and Gariépy et al., 1986). In most cases the membranes are designed to maintain specific ratios of the respiratory gases. For instance, at an atmospheric pressure of 100 kPa, the membrane's permeability to gases is normally 5.5 and 2.5 for CO_2:O_2 and

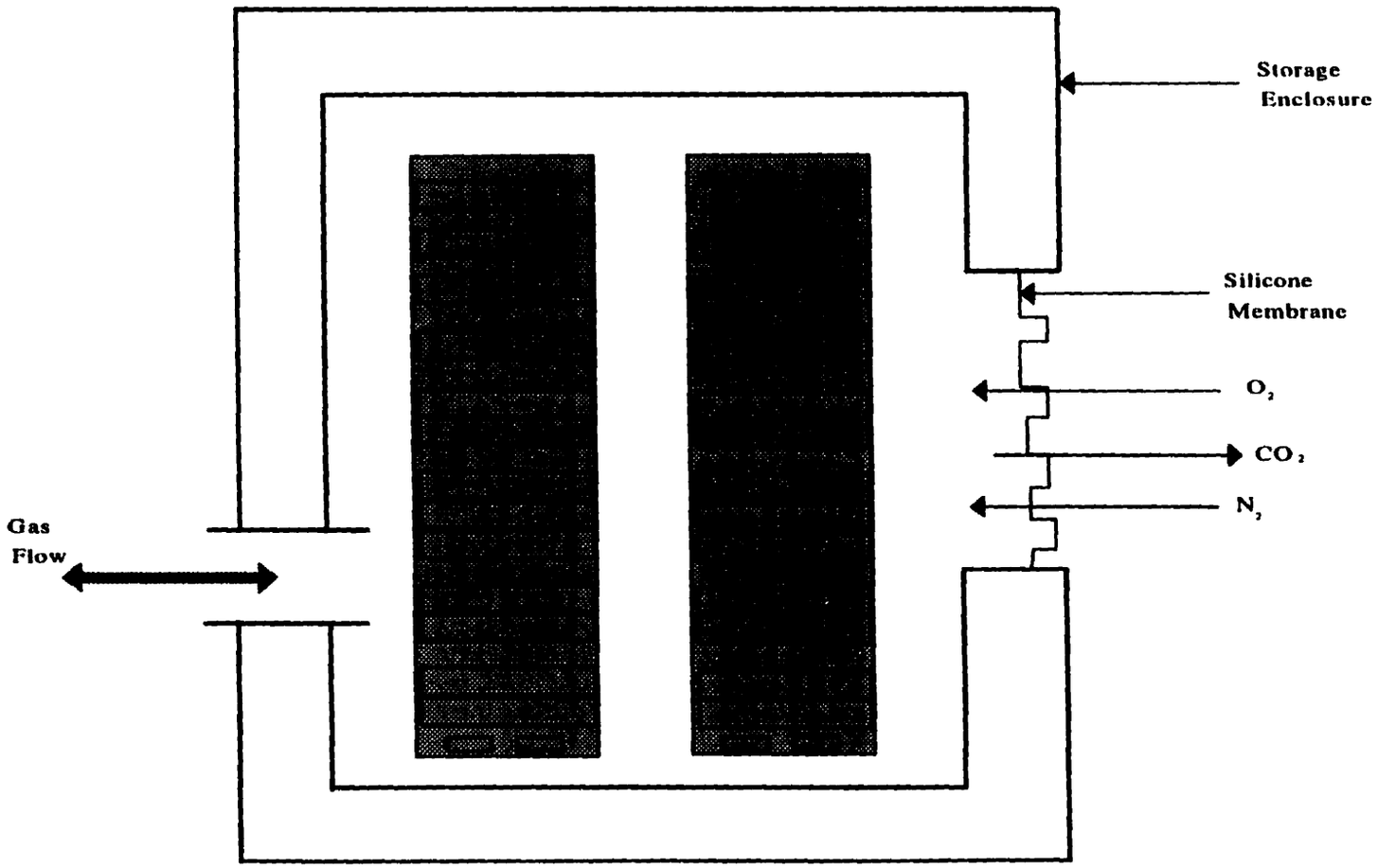

Figure 2.2 Schematic Diagram of a Membrane CA Storage System.

CO_2:C_2H_4 ratios, respectively (Gariépy et al., 1984b). The inflexibility in the ratios of the respiratory gases maintained in the storage chambers by the membranes makes the membrane system to be confined to storage of specific commodities. The membrane's permeability is also influenced by temperature (Smock, 1979 and Gariépy et al., 1988). As a consequence, large temperature fluctuations result in changes in gas composition and a detrimental effect on the quality of the stored product. There is also a tendency for water to condense on the inside surface of the membranes such that the membrane permeability is altered (Edmond and Chau, 1990).

Membrane systems that are available on the market include: the Pallet Package, Marcellin and the Atmolysair. Descriptions of the individual membrane systems, their pro and cons are reviewed as follows:

(i) **Pallet Package System**

The Pallet Package system was initially developed for controlling O_2 levels but coincidentally, it also regulated CO_2 levels (Smock, 1979). It consists of a pallet box wrapped in a heavy polyethylene bag with a twist tied end which ensures gas seal. A silicone rubber elastomer window is installed on one side to regulate gas exchange (Smock, 1979, and Gariépy et al., 1988). Figure 2.3 is a schematic diagram of the pallet package system.

The Pallet package system is relatively easy but time consuming in manipulating the gas composition (Smock, 1979). The design characteristics make it possible to market in individual pallets without interfering with the atmosphere of the remaining pallet boxes (Gariépy et al., 1988). The system however, involves some time consuming procedures. The system requires frequent analysis of the gas composition the pallets so as to ensure sufficient gas concentrations in the boxes (Smock, 1979). The process of wrapping and unwrapping can be tedious (Gariépy

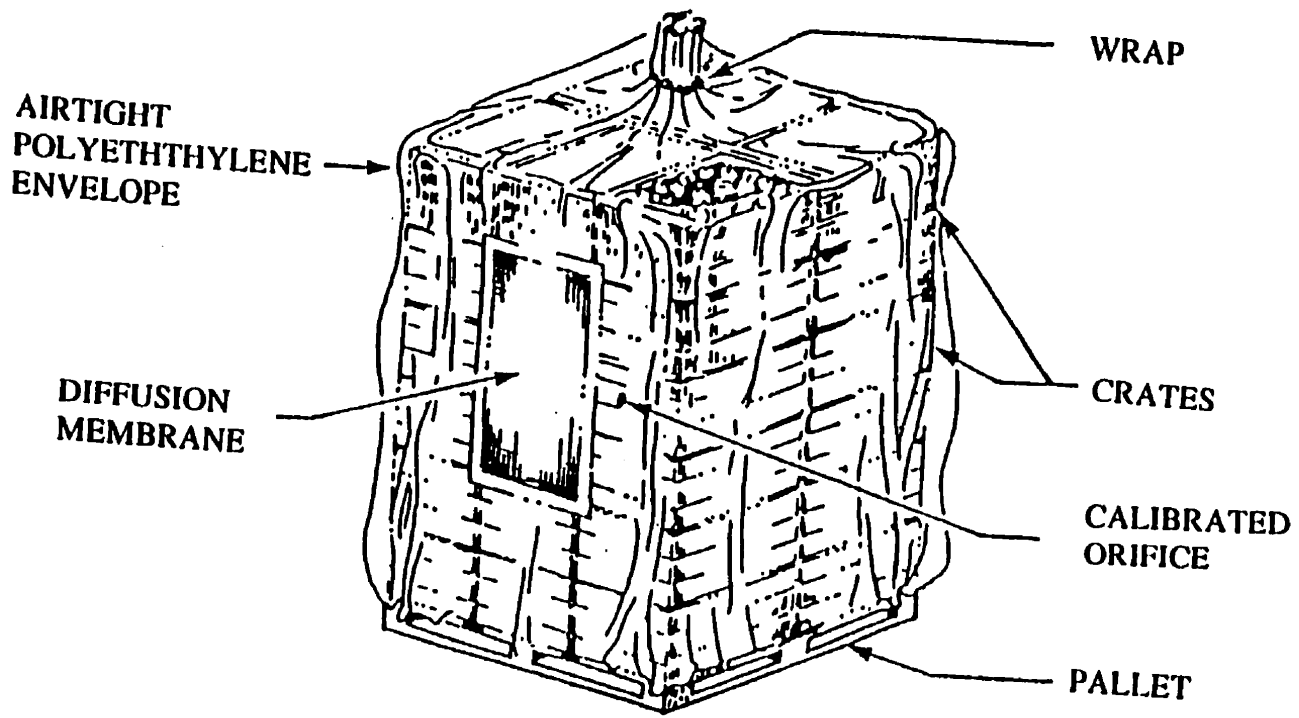

Figure 2.3 Diagram of The Pallet Package Membrane System.

et al., 1988). To obtain full advantage of the system, the pallets have to be well spaced and that reduces storage capacity of the room. The way the system is designed, requires it to be handled with special care (Gariépy, et al., 1988).

(ii) **The Marcellin System**

This is a membrane system in which a series of silicone rubber bags are connected to the storage room to regulate the inside atmospheric conditions (Figure 2.4). The unit can be hanged in an open chamber, outside the CA room (Smock, 1979) or can be installed inside the CA, cited by Gariépy et al. (1988). The required number of silicone bags is determined by the size of the room, storage temperature and type of the stored product (Smock, 1979, Gariépy et al., 1988). Frequent analysis of the inside atmosphere detects whether the bags should shut off or more bags should be added (Gariépy et al., 1988). The gas composition inside is also influenced by the membrane area. Gariépy et al. (1988) indicated that 100 Mt of produce with a bulky density in the range of 200 to 250 kg/m^3 will require a silicone membrane of 50 m^2 in area to maintain the standard CA gas composition of 3% CO_2 and 3% O_2.

(iii) **The Atmolysair System**

The Atmolysair system developed by Atmolysair LTD in Canada, is a modified version of the Marcellin System. The system as described by Raghavan et al. (1984) and Gariépy et al. (1988) consists of a square frame gas diffusion panel in which silicone membranes are fixed, and enclosed in an air tight metallic container which allow two separate air flow paths (figure 2.5). The gas diffusion channels are banked side by side so that there is no mixture of air from the storage room and the outside. Air circulation is achieved by use of centrifugal blowers regulated by a timer. Raghavan et al. (1982) presented a formula for calculating the required membrane area to maintain a specific desired gas composition as follows:

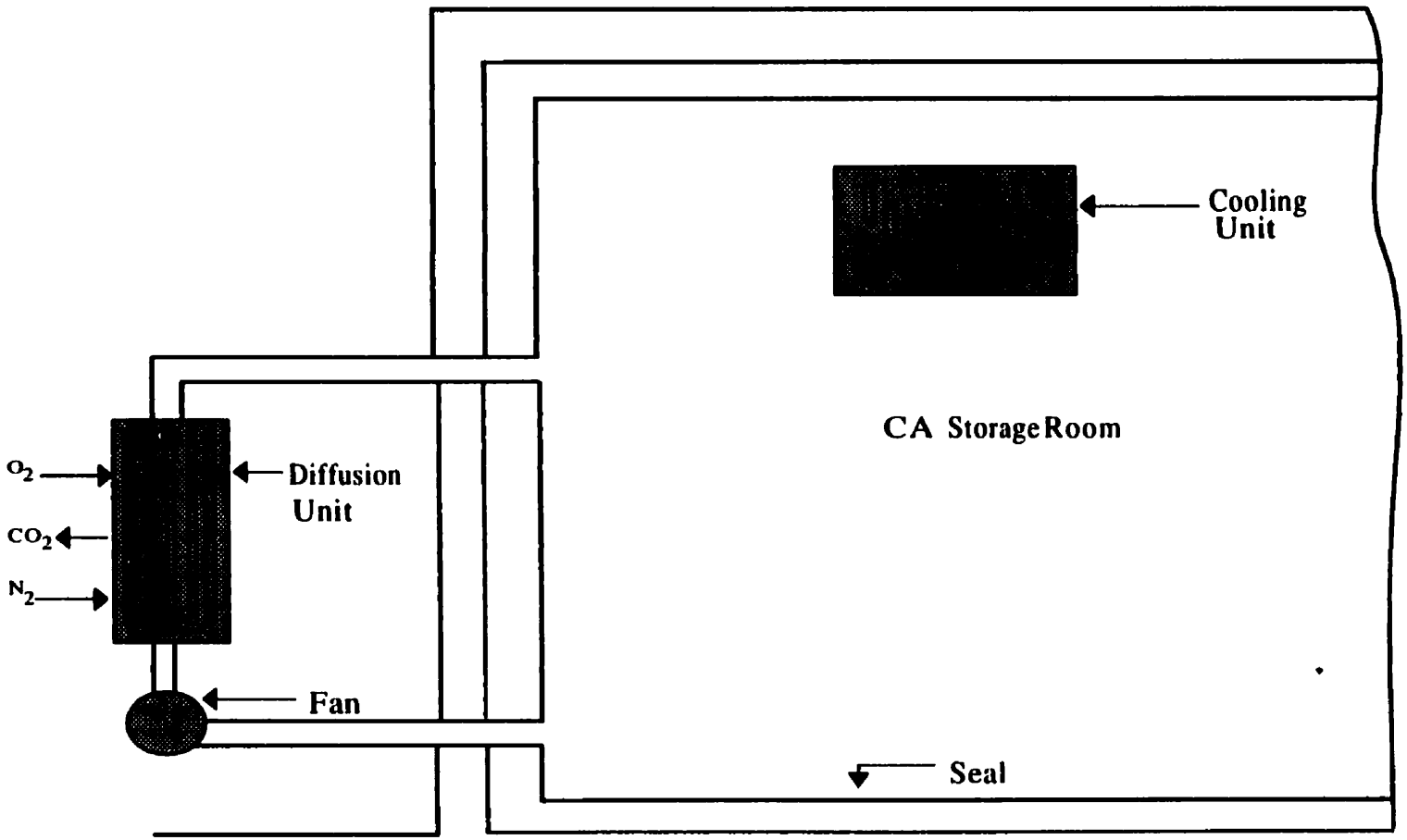

Figure 2.4 Schematic Diagram of the Marcellin Membrane System Installed Outside CA Room (Raghavan and Gariépy, 1985)

$$A_m = \frac{r * M}{P_{CO_2} * CO_2} \quad (2.5)$$

Where

A_m = Silicone membrane area (m^2)

P_{co2} = permeability of the silicone membrane to CO_2 (1750L/d.m_2.atm)

r = respiration rate of produce (L CO_2/ kg.d)

CO_2 = desired carbon dioxide partial pressure

M = mass of produce (kg)

When respiration rate of produce stored under CA is not known, "r" can be estimated between 60 and 70% of the ambient respiration rate (Gariépy et al., 1988). The set up has an advantage over the other two membrane systems because of its ease in managing and can easily be automated (Smock, 1979 and Gariépy et al., 1988). However, it is expensive.

2.1.3.1e **Diffusion Channel System**

Studies by Baugerod (1980), Ratti et al. (1993) and Ramachandra (1995) have shown that some of the technological and economical limitations associated with the membrane system could be solved by use of diffusion channels. The diffusion channels are simple to design and can be made from any impermeable materials which are readily available (Ratti et al., 1993 and Ramachandra, 1995). Figure 2.6 is a schematic diagram of the diffusion channel system showing how the channels are installed on the storage chamber. Baugerod (1980) described the principle behind the diffusion channels in controlling gas levels in the CA storage. In his study, gas diffusion into the CA storage through the channels was numerically related to bulk gas flow into the chambers by ventilation. Ratti et al. (1993) assessed the ability of different sized diffusion channels in maintaining desired gas composition for CA storage of fresh cauliflower. Ramachandra (1995)

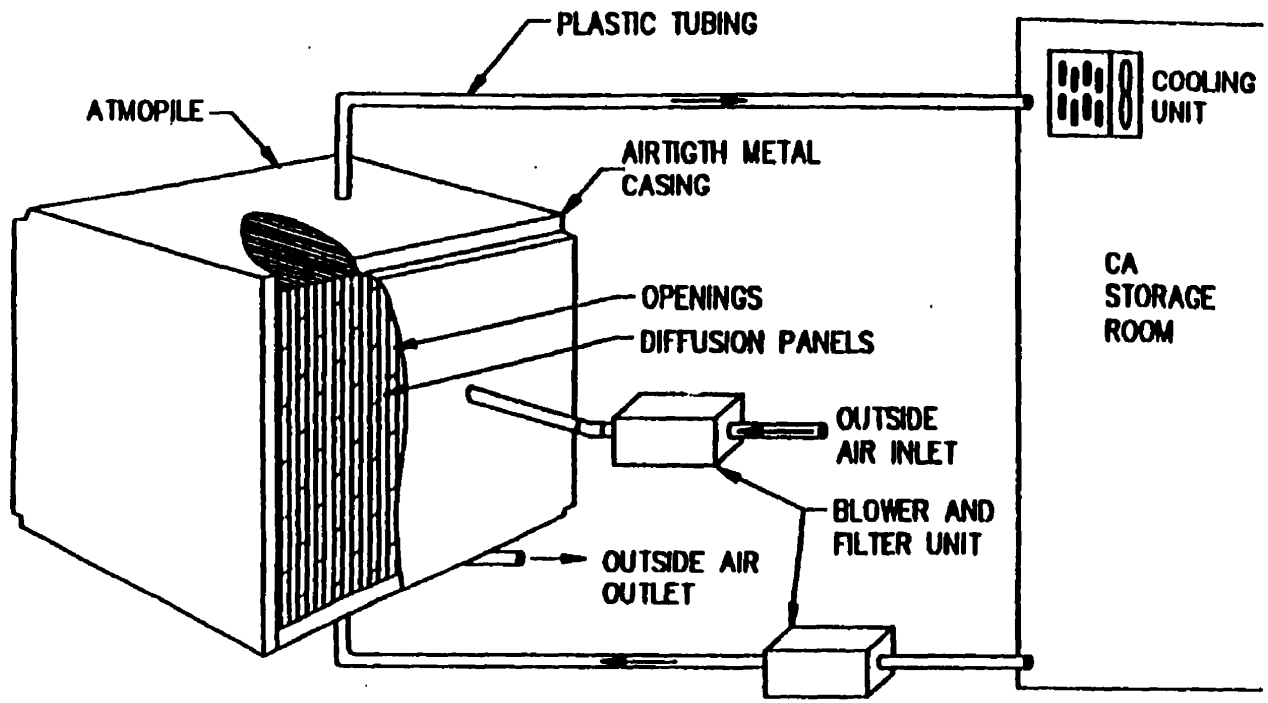

Figure 2.5 Schematic Diagram of The Atmolysair Membrane System (Raghavan and Gariépy, 1982)

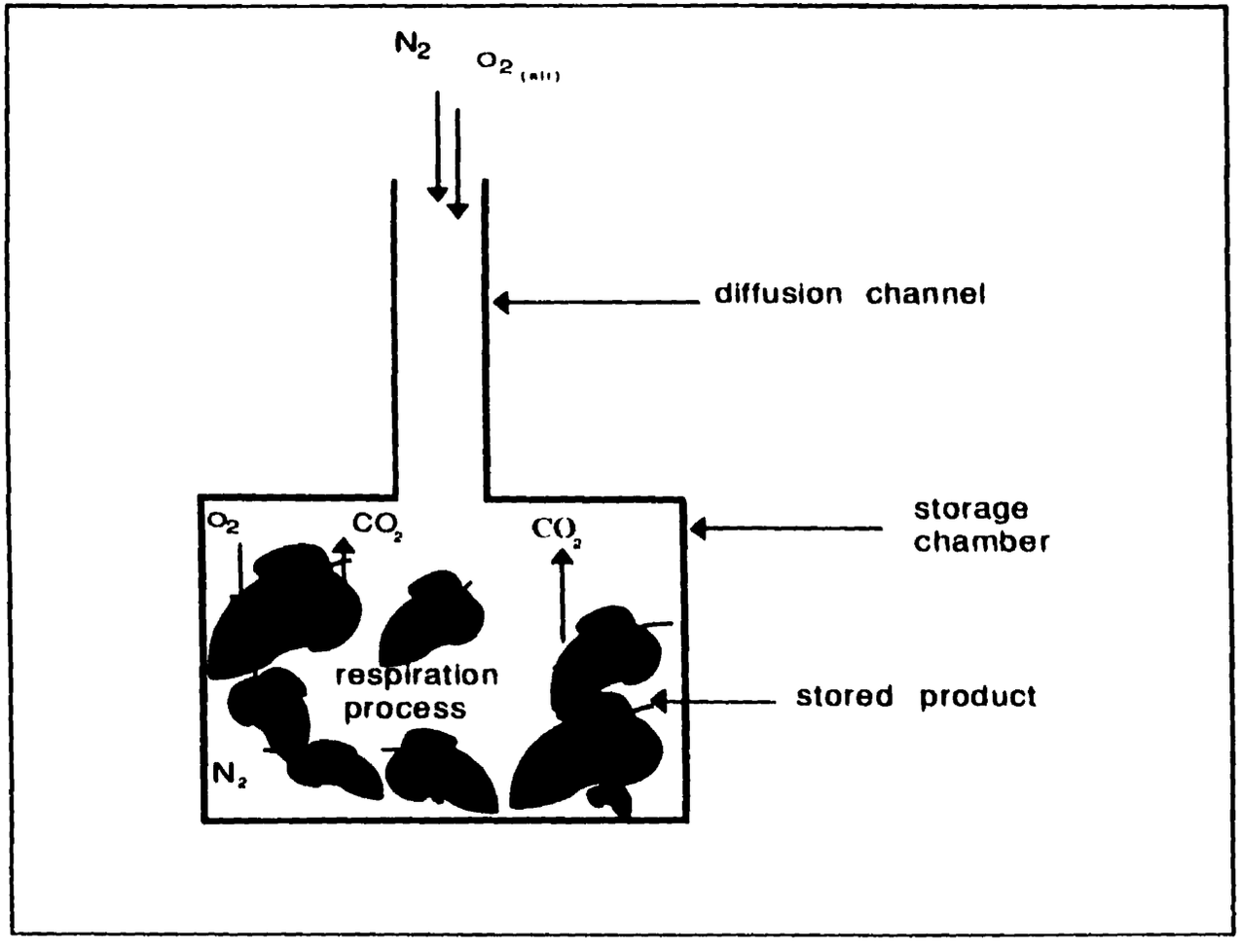

Figure 2.6 Schematic Diagram of The Diffusion Channel System

carried out a similar study on broccoli. In these studies the diffusion channel system was found to be capable of maintaining desired O_2 levels for longer periods and that it was relatively insensitive to changes in barometric pressure and temperature. Ratti et al. (1993) and Ramachandra (1995) found the performance of this new technology to be quite satisfactory in maintaining quality of the produce and also observed that the gas composition in the storage containers could easily be altered by varying the dimensions of the channels. It is also possible to attain gas levels higher than in the membrane system (Ratti et al., 1993). The relationship of the dimensions of the diffusion channels with the physiological characteristics of the stored product is not fully established. Ratti et al. (1993) and Ramachandra (1995) presented models which incorporate such a relationship. Theories behind those models are reviewed in the next Chapter.

2.2 CA Storage of Spinach

A lot of studies have been done to determine optimum CA storage conditions for many crops. However, emphasis has been mainly on fruits such as apples and pears, very little is available on vegetables (Kader, 1982; Thorne, 1983 and Wills et al., 1989). Cabbage, lettuce, tomato and cauliflower are the vegetables which to some extent have been subjected to CA research studies (Singh et al., 1972 and Isenberg, 1979).

Requirements and recommendations for CA storage are in most cases commodity specific and may vary within species (Hatton et al., 1975; Wills et al., 1981). The major variation comes about because of the diversity in the morphology and composition of the plant part that forms the vegetable. Vegetables can be leaves, tubers, roots, stems , flowers, sprouts and buds, which differ in their physiological characteristics (Wills et al., 1981). Optimal CA conditions for storage of fruits and vegetables have been compiled by several researchers (Wills et al., 1981; Meheriuk, 1985; Richardson, 1985; Saltveit, 1985; Hardenburg et al.,

1990 and Kader, 1992b). Table 2.3 and 2.4 are summaries of the recommended CA conditions for some fruits and vegetables respectively.

Application of CA conditions for some of the commodities has been limited because the extra cost of establishing a CA storage may not be economically justifiable (Wills et al., 1981). Priority, is therefore, given to commodities with high economic value.

Spinach, *Spinacia oleracea* L. is one of the most important leafy vegetables widely cultivated and used in many dishes (Isenberg, 1979). It is mostly consumed because of its vitamins and minerals (Ryall and Lipton, 1972). Nutritional composition of spinach is given in Table 2.5. Spinach is, however, one of the highly perishable commodities, as such, it has a relatively short shelf life (Hardenburg et al., 1990 and Kader, 1992a). Such a characteristic is attributed to the high water content (which is almost 90%) and the large surface to volume ratio, that permits rapid loss of water and also facilitates gaseous exchange (Burton, 1982 and Peirce, 1987) with subsequent deterioration of the product.

Spinach is normally stored in RA at temperatures between -1°C and 4°C (Wills et al., 1981) and is in most cases prepacked in perforated plastic bags in order to reduce moisture loss and physical injuries. Sometimes crushed ice is added for rapid field heat removal (Hardenburg et al., 1990). Nevertheless, its storage life is limited to only 1-2 weeks (Wills et al., 1981 and Hardenburg et al., 1990).

There has been attempts to use CA conditions in maintaining quality of spinach for extended periods. Potential benefits have been reported. Platenius (1943) observed a pronounced reduction in respiration rate of spinach when stored in limited oxygen. Similar results have been reported by McGill et al. (1966) and Burgheimer et al. (1967). Spinach stored in CA was also found to retain the green colour which is as an indication that degradation of chlorophyll is retarded (McGill

et al., 1966). Platenius (1943), in his study suggested the use of CA in retarding formation of oxalic acid which make calcium unavailable. Ascorbic acid loss in spinach, to some extent, is also reported to be reduced or maintained by CA storage (McGill et al., 1966). Similar results were found by Murata and Ueda cited by Isenberg (1979). It is therefore, evident that CA conditions can indeed alleviate the current problems in storing spinach (Isenberg, 1979). However, there is need for more research to establish the optimum gas levels for its good storability. Furthermore, to determine an economical and effective CA storage system which can extend the shelf life of spinach. In this regard, diffusion channel system is assessed for its suitability.

Table 2.3 Summary of Recommended CA/MA Conditions During Transport/Storage of Some Vegetables.

	CA				
Commodity	T (°C)	O_2 (%)	CO_2 (%)	Potential Benefit	Remarks on Usage
Artichokes	0-5	2-3	2-3	good	Commercial use
Asparagus	0-5	air	5-10	excellent	Limited commercial use
Beans, Snap	5-10	2-3	4-7	fair	Potential use by processors
Beets	0-5	N/A	N/A	N/A	98-100% RH is best
Broccoli	0-5	1-2	5-10	excellent	Limited commercial
Brussels sprouts	0-5	1-2	5-7	good	No commercial use
Cabbage	0-5	2-3	3-6	excellent	Commercial use for longterm storage of some cultivars
Cantaloupes	3-7	3-5	10-15	good	Limited commercial use
Carrots	0-5	N/A	N/A	N/A	98-100% RH is best
Cauliflower	0-5	2-3	2-5	fair	No commercial use
Celery	0-5	1-4	0-5	good	Limited commercial use in mixed loads with lettuce.
Corn, sweet	0-5	2-4	5-10	good	No commercial use
Cucumbers	8-12	3-5	0	fair	No commercial use
Honeydews	10-12	3-5	0	fair	No commercial use
Leeks	0-5	1-2	3-5	good	No commercial use
Lettuce	0-5	1-3	0	good	Commercial use with 2-3% CO_2
Mushroom	0-5	air	10-15	fair	Limited commercial use
Okra	8-12	3-5	0	fair	No commercial use; 5-10% CO_2 beneficial at 5-8°C
Onions, dry	0-5	1-2	0-5	good	No commercial use; 75% RH
Onions, green	0-5	1-2	10-20	fair	Limited commercial use
Peppers, bell	8-12	3-5	0	fair	Limited commercial use
Peppers, chili	8-12	3-5	0	fair	No commercial use; 10-15% CO_2 beneficial at 5-8°C
Potatoes	4-12	N/A	N/A	N/A	N/A
Radish	0-5	N/A	N/A	N/A	98% RH is best
Spinach	0-5	air	10-20	good	No commercial use
Tomatoes,					
Mature green	12-20	3-5	0-3	good	Limited commercial use
Partially ripe	8-12	3-5	0-5	good	Limited commercial use

Source: Kader (1992b)

Table 2.4 Summary of Recommended CA/MA Conditions During Transport/Storage of Some Fruits.

Commodity	CA T (°C)	CA O_2 (%)	CA CO_2 (%)	Potential Benefit	Remarks on Usage
Deciduous Trees					
Artichokes	0-5	1-3	1-5	excellent	Commercial use: 50% stored under CA
Apricot	0-5	2-3	2-3	fair	No commercial use
Cherry sweet	0-5	3-10	10-15	good	Some commercial use
Fig	0-5	5-10	15-20	good	Limited commercial use
Grape	0-5	2-5	1-3	fair	Incompatible with SO_2 fumigation
Kiwifruit	0-5	1-2	3-5	excellent	Some commercial use (C_2H_4<20ppb)
Nectarine	0-5	1-2	3-5	good	Limited Commercial use
Peach	0-5	1-2	3-5	good	Limited commercial use
Pear Asian	0-5	2-4	0-1	good	Limited commercial use
Pear European	0-5	1-3	0-3	excellent	Some commercial use
Persimmon	0-5	3-5	5-8	good	Limited commercial use
Plum and Prune	0-5	1-2	0-5	good	Limited commercial use
raspberry and	0-5	5-10	15-20	excellent	Increasing use during transport
Other caneberries	0-5	5-10	15-20	excellent	Increasing use during transport
Strawberry	0-5	5-10	15-20	excellent	Increasing use during transport
Nuts and dried fruits	0-5	0-1	0-100	excellent	Effective insect control
Sub Tropical and Tropical Fruits					
Avocado	5-13	2-5	3-10	good	Limited commercial use
Banana	12-15	2-5	2-5	excellent	Some commercial use during transport
Grape fruit	10-15	3-10	5-10	fair	No commercial use
Lemon	10-15	5-10	0-10	good	No commercial use
Lime	10-15	5-10	0-10	good	No commercial use
Olive	5-10	2-3	0-1	fair	No commercial use
Orange	5-10	5-10	0-5	fair	No commercial use
Mango	10-15	3-5	5-10	fair	Limited commercial use
Papaya	10-15	3-5	5-10	fair	No commercial use
Pine apple	8-13	2-5	5-10	fair	No commercial use

Source: Kader (1992b)

Table 2.5 Nutritional Composition of Spinach (per 100g sample).

Nutrient	Amount
Water (%)	91.0
Energy (cal)	26.0
Protein (g)	3.20
Fat (g)	0.30
Carbohydrates (g)	4.30
Vitamin A (IU)	8000
Vitamin C (mg)	51.0
Thiamine (mg)	0.10
Riboflavin (mg)	0.20
Niacin (mg)	0.60
Ca (mg)	93.0
P (mg)	51.0
Fe (mg)	3.10
Na (mg)	71.0
K (mg)	47.0

Source: Yamaguchi (1983)

CHAPTER III

THEORETICAL ASPECTS

3.1 CA Establishment

In an attempt to establish a Controlled Atmosphere (CA) storage system as a means of extending postharvest life of fresh produce, respiration process of the produce has to be well understood. Pflug and Dewey (1957) indicated that there is a strong relationship between the respiring produce and the gas exchange characteristics of the storage room. Respiration removes oxygen from the environment, which if depleted beyond extinction point, may lead to the Pasteur effect (Kays, 1991). As a consequence, the respiration rate is essential in determining the ventilation requirements of the storage room. It is also a guide in determining a suitable storage system to be used for produce. If CA conditions are to be achieved passively (commodity generated) in a storage room with various barriers and restrictions to gas exchange, respiration rate can be useful in determining the duration for the desired gas composition to be achieved. Knowledge of the respiration rate can also give an estimate of the vital heat being produced by the stored product (Van Den Berg and Lentz, 1972; Kays, 1991 and Kader, 1992b) which is essential in determining how much and mode of cooling the product, stacking style and size of a refrigeration system (Mohsenin, 1980 and Shewfelt, 1986). Kader (1992b) expressed a model on the relationship of a stored commodity and its environment as shown in Figure 3.1.

Oxygen is utilised and carbon dioxide released from the respiration process. Baumann (1977) and Baugerod (1980) indicated that a stable atmospheric composition in the CA storage room can be achieved if carbon dioxide is removed at the same rate as it is being released and oxygen be supplied at the same rate of its consumption.

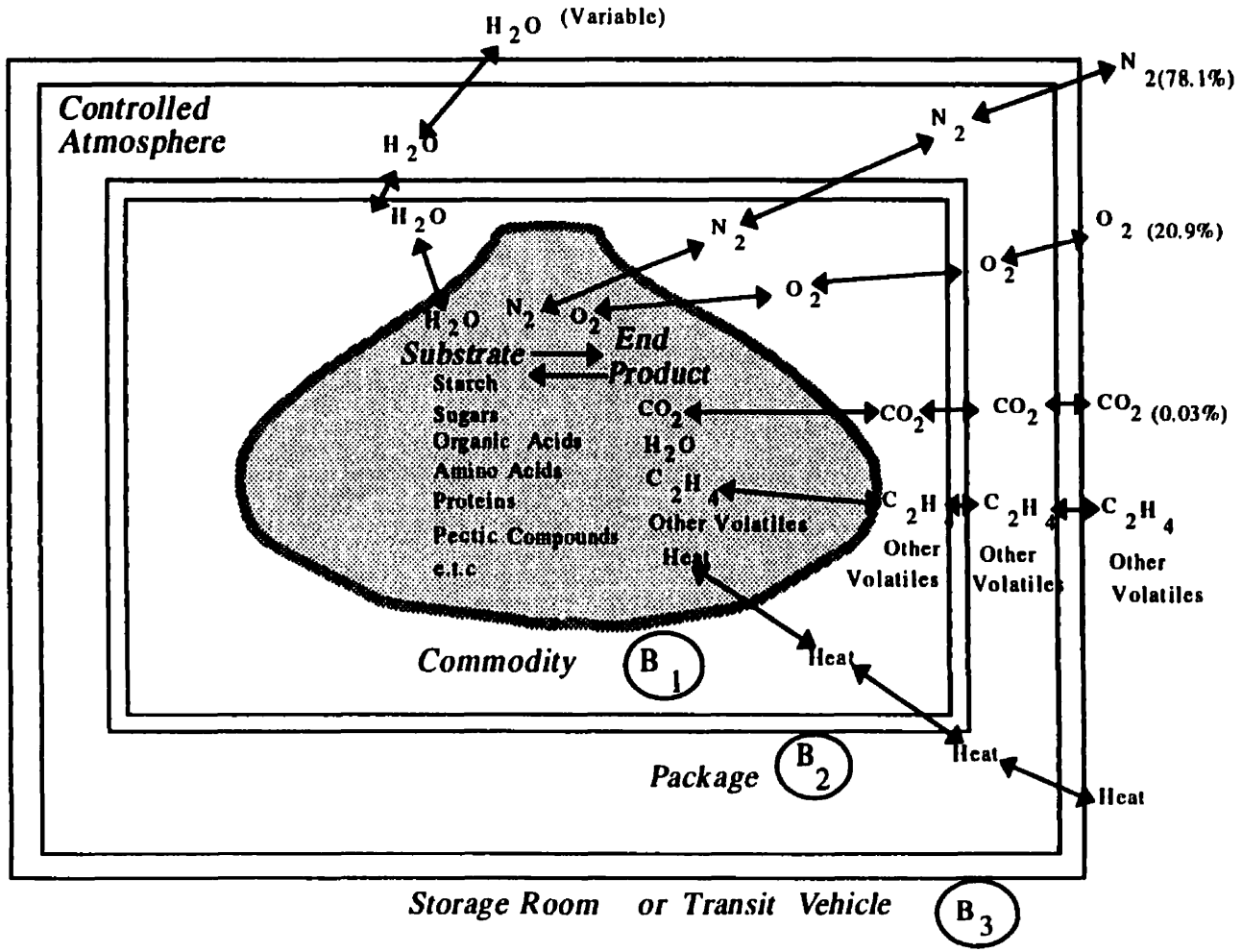

Figure 3.1 The Relationship between a Commodity and its Microenvironment (Model) (Kader, 1992b)

Several mathematical models have been developed for CA/MA storage of fresh fruits and vegetables which recognise the interaction between respiration and permeation of respiratory gases in the storage room (Henig and Gilbert, 1975; Hayakawa et al., 1975; Baumann, 1977; Zagory and Kader, 1988; Edmond and Chau, 1990; Ratti et al., 1993 and Ramachandra, 1995). In their models, it is evident that knowledge of respiration rate of a produce and the factors influencing it, are paramount in designing, establishing and maintaining CA storage systems.

3.2 Modelling Respiration Rate

Measurement of respiration rate of a produce can be a time consuming process (Forcier et al., 1987). Therefore, it is essential to find a rapid and precise method of predicting the respiration rate of a commodity under any given storage condition, without requiring extensive experimentation. In the recent years, innovation of computers and data acquisition systems has made it possible to measure some factors, such as respiration that affect storability of produce in situ (Bishop, 1981; Forcier et al., 1987; Beaudry and Gran, 1993). However, such automation can be costly and may adversely affect the cost-benefit ratio of the whole system.

Several attempts have been made to model the dynamics of the respiration rate for fresh produce (Lee et al., 1991). However, modelling of such biological processes per se, is challenging because of their complexity. As reviewed in the previous chapter, respiration process involves a series of enzymatic reactions that take place through several metabolic pathways, such as Embeden-Meyerhof-Parnas (EMP), Tricarboxylic Acid (TCA) cycle and the associated electron transport system (Phan et al., 1975; Solomos, 1981; Wills et al., 1981 and Burton, 1982). The chemical reaction given in Equation 2.1, indicates that a six carbon sugar is the metabolic substrate for the respiration process. However, in the plant, there are many other organic compounds which are also involved in such reactions. This

effect is reflected in the Respiratory Quotient (RQ). In most cases RQ value does not remain constant with time. It may vary from 0.7 to 1.3 (Bidwell, 1979; Wills et al., 1981 and Forcier et al., 1987). An RQ of 1 would normally represent oxidation of sugar. The variation in the RQ clearly shows that different substrates with different degrees of oxidation, participate in the reactions (Phan et al., 1975 and Zagory and Kader, 1988).

In view of the complexity of the respiration process, Lee et al. (1991) suggested the use of Michaelis-Menten equation (Equation 3.1) which describes enzymatic reactions in an unpurified state (Bailey and Ollis, 1977 and Cornish-Bowden, 1979).

$$v = \frac{Vs}{(K_m + s)} \qquad (3.1)$$

Where,

v = rate of reaction (at time "t"=0)

V = maximum velocity

s = substrate concentration

K_m = Michaelis constant

Based on Equation 3.1, Lee et al. (1991) developed a model for predicting the respiration rate of fresh produce. In the model, the relationship of the respiration rate and oxygen concentration was expressed as shown in Equation 3.2. The dependency of respiration rate on carbon dioxide concentration was considered to be of uncompetitive inhibition (Equation 3.3). This equation could be valid only under aerobic conditions, where sufficient oxygen is available for the respiration process (Lee et al., 1991).

$$r = \frac{V_m[O_2]}{K_m + [O_2]} \tag{3.2}$$

$$r = \frac{V_m[O_2]}{K_m + (1 + [CO_2]/K_i)[O_2]} \tag{3.3}$$

Where,

r = respiration rate (ml or mg $kg^{-1}. h^{-1}$)

V_m = maximum respiration rate (ml or mg $kg^{-1}.h^{-1}$)

K_m = Michaelis-Menten Constant (% O_2)

$[O_2]$ = Oxygen concentration (%)

$[CO_2]$ = Carbon dioxide concentration (%)

K_i = inhibition constant (% O_2)

It is indicated that uncompetitive inhibition occurs almost exclusively as a type of inhibition that is common in reactions with several substrates and products (Cornish-Bowden, 1979).

Ratti et al. (1993) developed a similar model for predicting respiration rate of fresh cauliflower. The effect of temperature was expressed by the Arrehnius relationship (Equation 3.4). Based on the same principle, Ramachandra (1995) developed a model for predicting respiration rate of fresh broccoli.

$$r = \frac{k_1 C_{O_2}}{K_2 + (1 + K_3 C_{CO_2}) C_{O_2}} \tag{3.4}$$

In the present investigation, Ratti et al.'s (1993) model was adopted and modified slightly for modelling of the respiration rate of spinach.

In Equation 3.4, the effect of temperature was expressed by the Michaelis' constant K_i (where i = 1, 2, 3). Vant-Hoff and Arrhenius described similar relationships in their first theories of dependency of temperature on the rate

constants such that constants K_1, K_2 and K_3 can be expressed by Arrhenius relationship as follows (Cornish-Bowden, 1979).

$$\frac{d\ (lnK)}{dT} = \frac{E_a}{R\ T^2} \qquad (3.5)$$

Where,

R = universal gas constant

T = absolute temperature (°K)

E_a = activation energy

Integrating Equation 3.5 with respect to T, gives

$$\ln K = \ln A - (E_a/RT) \qquad (3.6)$$

Where,

A = integration constant

After taking an exponential and rearranging, Equation 3.6 can be expressed as:

$$K = A \exp(-E_a/RT) \qquad (3.7)$$

The activation energy E_a in Equation 3.7 corresponds to the standard enthalpy change of reaction ΔH° as expressed in Vant-Hoffs equation (Cornish-Bowden, 1979).

$$\frac{d\ (lnK)}{dT} = \frac{\Delta H^\circ}{RT^2} \qquad (3.8)$$

Ratti et al. (1993) indicated also that the effect of temperature of the produce can be expressed by the following energy balance equation.

$$\frac{dT}{dt} = \frac{h_g A}{mC_p}(T_g - T) - \frac{(\Delta H_r)r}{C_p} \qquad (3.9)$$

At equilibrium ($-E_a/R$) is constant and can be denoted by b_i. K will be a function of temperature. Equation 3.7 can then be written as follows:

$$K_i = A_i \exp(b_i/T) \qquad (3.10)$$

Equation 3.4 therefore, becomes

$$r = \frac{A_1 \exp(b_1/T) * C_{O_2}}{(A_2 \exp(b_2/T) + (1 + A_3 \exp(b_3/T) * C_{CO_2}) * C_{O_2}} \qquad (3.11)$$

Constants A_i and b_i (where i = 1, 2 and 3) can be obtained through non linear regression analysis of the experimental data. Values of K_1, K_2 and K_3 can then be calculated numerically.

3.3 Modelling Diffusion Channel Length for CA Storage

Most of the work on MA/CA has been on polymeric film packaging (Henig and Gilbert, 1975; Hayakawa et al., 1975; and Zagory and Kader, 1988). These films, in cases where high CO_2 is desired, are limited to their permeability. Their performance can be improved by use of perforations (Edmond and Chau, 1990). Such perforations are related to diffusion channels with a length of equal to the thickness of the film (Ramachandra, 1995).

Ratti et al. (1994) indicated that manipulating the length and cross sectional area of a diffusion channel, the atmospheric gas composition of the storage chamber can change. Edmond and Chau (1990) developed a model for predicting the gas composition in perforated polymeric film packages. Following the same idea, Ratti et al. (1994) and Ramachandra (1995) developed models for predicting length of the diffusion channel which can maintain desired gas level for CA storage of a given mass of cauliflower and broccoli, respectively. In the present

investigation, Ratti's and Ramachandra's models are adopted and modified to predict length of diffusion channel which will be capable of maintaining a desired level of oxygen for CA storage of spinach.

CA storage involves atmospheric gas mixtures and their diffusion characteristics. Knowledge of some of the laws governing behaviour of gases is important. Some laws have been outlined (Kader, 1992c). Gas diffusion normally obeys **Fick's First Law of Diffusion**, which states that "gas diffuses from region of higher concentration to region of lower concentration". The movement of the gases is due to the random motion of the individual molecules caused by their kinetic energy (Gebhart,1993 and Kays, 1991). The gas flux can be expressed as follows:

$$J = -D * A * 1/L * (C_i - C_o) \qquad (3.12)$$

Where,

J = rate of transfer

D = diffusion coefficient (the negative sign is because the substance is moving in the direction of decreasing concentration)

A = area of barrier to the diffusion

L = thickness of the barrier to diffusion

C_i = inside concentration

C_o = outside concentration

In steady state conditions, the diffusion rate of a gas is proportional to the concentration difference (C_i - C_o) across the material and the area "A" perpendicular to the diffusion. It is however, inversely proportional to the length "L" of the gas diffusion path, between the two concentration levels (C_i and C_o) (Gebhart, 1993).

In this study, modelling is done based on the diffusion of oxygen into the chambers. Oxygen was preferred because it is the gas which has a pronounced effect on the rate at which the respiration process proceeds (Platenius, 1943 and Kays, 1991). Since the atmosphere consists of three major gases (O_2, CO_2 and N_2) scrubbing off carbon dioxide, makes the diffusion process to be binary, with nitrogen and oxygen as diffusing gases. Assuming that there is no concentration gradient for nitrogen between the outside and the inside, oxygen will be diffusing in a stagnant N_2 gas. The diffusion of oxygen into the storage chamber, through the channel is conceptualised in Figure 3.2.

In the binary system, the flux of the diffusing substance (O_2) into a stagnant gas (N_2) still obeys the laws of molecular diffusion (Gebhart, 1993) and can be written as:

$$n_{O_2} = - cD_{O_2 - N_2} * \frac{dy_{O_2}}{dz} + y_{O_2} n_{O_2} \qquad (3.13)$$

Where,

n_{O2} = oxygen mass flux ($g/m^2.h$)

c = total gas concentration (mol/m^3)

D_{O2-N2} = diffusivity coefficient of oxygen in nitrogen (m^2/h)

y_{O2} = oxygen mole fraction

z = spatial coordinate (m)

Following Fick's first law of diffusion, oxygen will diffuse into the chamber. Under unsteady state conditions, the mass balance can be expressed by the following differential equation (Ratti et al., 1994).

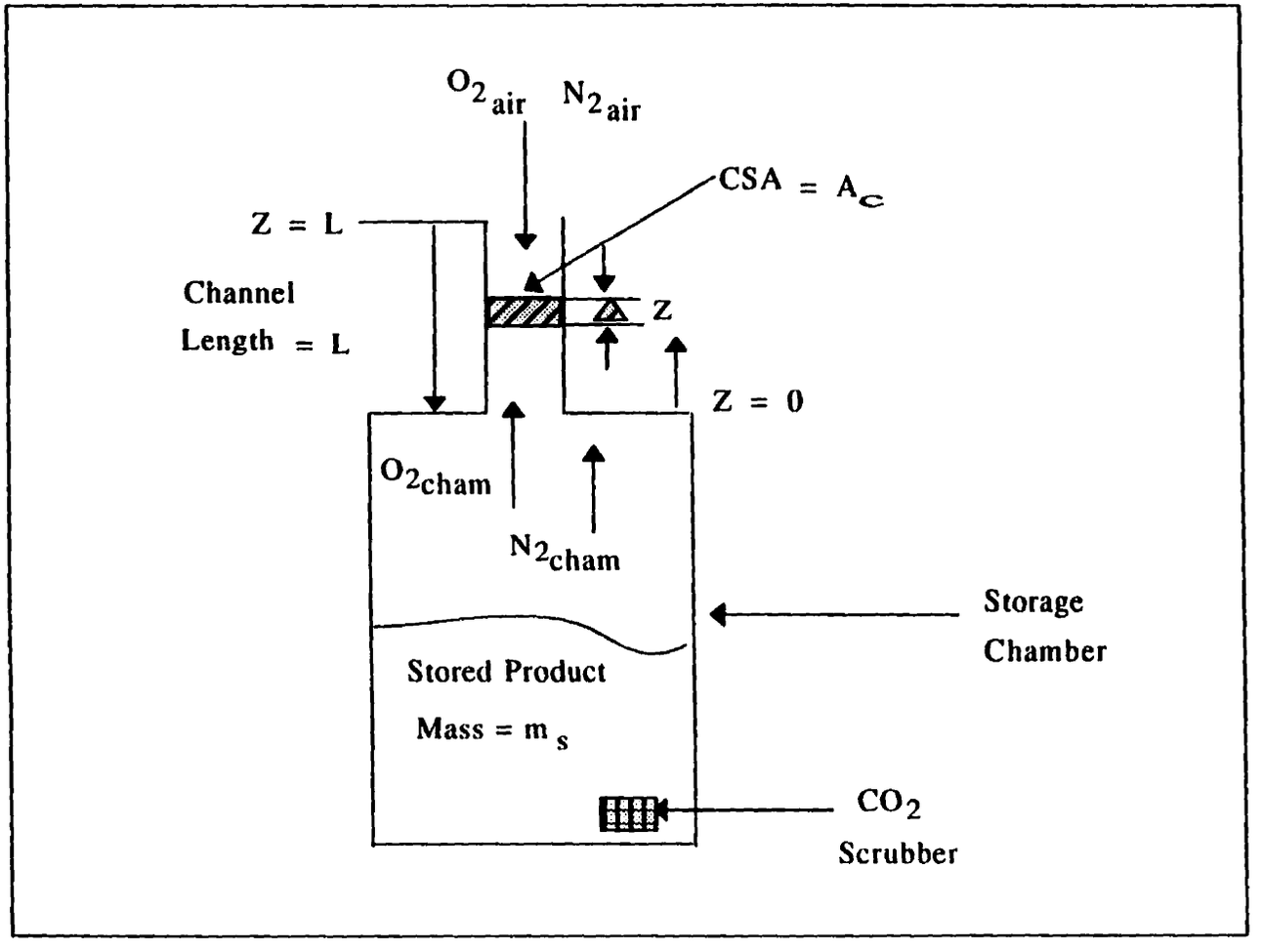

Figure 3.2 Oxygen Diffusion Process into a Storage Chamber Through a Channel

$$\frac{\partial y_{O_2}}{\partial t} = D_{O_2 - N_2} * \frac{\partial^2 y_{O_2}}{\partial z^2} \qquad (3.14)$$

If the oxygen diffusing into the chamber is equal to the amount utilized in the respiration process, i.e under steady state, the time partial component in Equation 3.14 becomes negligible and the flux of oxygen is constant.

With the following initial and boundary conditions

$$t = 0 \rightarrow y_{O_2} = (y_{O_2})_0 \qquad (3.15)$$

$$z = L \rightarrow y_{O_2} = (y_{O_2})_{air} \qquad (3.16)$$

$$z = 0 \rightarrow y_{O_2} = (y_{O_2})_{cham} \qquad (3.17)$$

Integrating between limits and rearranging, Equation 3.13 becomes

$$n_{O_2} = c * \frac{D_{O_2 - N_2}}{L} * \ln\left[\frac{1 - y_{O_{2_{air}}}}{1 - y_{O_{2_{cham}}}}\right] \qquad (3.18)$$

In the two boundaries z = 0 and z = L, it is assumed that there is no absorption, retention nor release of oxygen, hence, the diffusivity coefficient (D_{O2-N2}) is constant and uniform across the region of diffusion. Care has to be taken in choosing the material for making the diffusion channel. Readily oxidised materials can easily alter the diffusivity coefficient (D_{O2-N2}) resulting in lower O_2 levels than desired.

O_2 consumed in the respiration process can be expressed numerically as follows:

$$M_{O_{2cons}} = r_{O_2} * t * m_s \quad (3.19)$$

Where:

M_{O2cons} = Oxygen Consumed (mg)

t = time (h)

m_s = mass of produce (kg)

r_{O2} = respiration rate (mg O_2/kg.h)

and under steady state conditions mass flux can be presented as

$$n_{O_2} = \frac{M_{O_{2cons}}}{t * A_c} \quad (3.20)$$

Where:

M_{O2cons} = M_{O2cham} (g)

A_c = cross sectional area of the diffusion channel (m^2)

M_{O2cham} = oxygen diffused in the chamber (g)

Equation 3.20 can be re written as

$$n_{O_2} = \frac{r_{O_2} * m_s}{Ac} \quad (3.21)$$

Combination of Equations 3.18 and 3.21 can give length of the channel as a function of oxygen concentration, cross sectional area, respiration rate and mass of produce.

$$L = A_c * c * \frac{D_{O_2\text{-}N_2}}{r_{O_2} * m_s} * \ln\left[\frac{(1 - y_{O_{2air}})}{(1 - y_{O_{2cham}})}\right] \quad (3.22)$$

and under steady state conditions, concentration of oxygen can be expressed as

$$C_{O_2} = y_{O_2} * c \tag{3.23}$$

Where C_{o2} is concentration of oxygen ($mol.m^{-3}$)

In this case, respiration rate is taken as a function of oxygen concentration and temperature. Expressing Equation 3.11 in terms of oxygen concentration and temperature, yields:

$$r = \frac{A_1 e^{(b_1/T)} * y_{O_2} * c}{(A_2 e^{(b_2/T)} + (y_{O_2} * c))} \tag{3.24}$$

Equation 3.22 can be written as:

$$L = \frac{A_c}{m_s} * \frac{D_{O_2\text{-}N_2} * [A_2 e^{(b_2/T)} + c * y_{O_2}]}{(A_1 e^{(b_1/T)} * y_{O_2})} * \ln[\frac{(1 - y_{O_{2_{air}}})}{(1 - y_{O_{2_{chamber}}})}] \tag{3.25}$$

In the Equation above:

$$\frac{1}{(A_1 * e^{(b_1/T)})} = a_1 \tag{3.26}$$

$$D_{O_2\text{-}N_2} * c = a_2 \tag{3.27}$$

and

$$D_{O_2\text{-}N_2} * A_2 * e^{(b_2/T)} = a_3 \tag{3.28}$$

Where a_1, a_2 and a_3 are constants and can be obtained by non-linear regression analysis of the experimental data. Equation 3.28 can therefore, be written as

$$L = \frac{A_c}{m_s} * \frac{(a_3 + (a_2 * y_{O_2}) * a_1)}{y_{O_2}} * \ln[\frac{(1 - y_{O_{2_{air}}})}{(1 - y_{O_{2_{chamber}}})} \qquad (3.29)$$

Equation 3.29 is valid only with the following assumptions:

(i) Diffusion of gases in the chamber is one dimensional

(ii) The system is binary thus only oxygen and nitrogen are diffusing

(iii) Concentration of oxygen in the chamber is constant (steady state conditions attained)

(iv) The diffusivity of oxygen in nitrogen is constant

(v) Total pressure in the chamber is 1 atm

(vi) Temperature is constant

(vii) All the carbon dioxide is scrubbed.

CHAPTER IV

MATERIALS AND METHODS

Three series of laboratory experiments were carried out in the Postharvest Technology Laboratory of the Department of Agricultural and Biosystems Engineering at McGill University, Macdonald Campus. The first series was basically done to assess the effect of temperature on the respiration rate of spinach and to use the data in developing a mathematical model that can be used to predict the respiration rate of a known mass of spinach for any given storage temperature. The other two series were geared towards assessing the suitability of the diffusion channel system in creating proper CA conditions for storage of spinach and also develop a mathematical model which could help prospective users of the diffusion channel system in predicting the length of the diffusion channel which can create such desired conditions.

4.1 Effect of Temperature on the Respiration Rate of Spinach

Respiration rate of spinach was studied at four levels of temperature 2, 8 and 15°C and ambient temperature (23°C). The experiment was a complete randomised statistical design, replicated four times. Fresh Quebec grown spinach, purchased in perforated plastic bags from a local wholesaler, was used for the experiment. Prior to starting the experiment, the spinach was pre-emptied from the plastic bags and trimmed accordingly. The spinach was then put in one big container and mixed properly (avoiding mechanical injuries) in order to obtain uniform samples. Samples with an average mass of 200 g were stored in small air tight chambers made from PVC pipe sections of 0.32 m in height and an internal diameter of 0.10 m. The bottom parts of the chambers were tightly sealed with plexi glass, 60 mm thick, whereas, the top parts were covered with transparent lids which could be screwed on and off (Figure 4.1). Two septa were installed on each of the lids as

gas sampling ports.

Prior to storing the spinach, the chambers were tested for air tightness at 5.5 kPa for 20 minutes using a SRI DPI 601 digital pressure indicator with a resolution of 0.05% within the range of 1 to 3500 kPa. Density of the spinach was determined according to Stroshine and Hamann (1994). The volume of the chambers was arithmetically calculated. The two parameters (Density and Volume) were used in determining the amount of spinach to be stored in each chamber. The chambers were cleaned with chlorinated water, 800 p.p.m., so as to minimize incidence of pathogenic infection of the spinach during storage. Before storing the spinach at the specific temperatures, the spinach was precooled in a cold room set at 2°C for 12 hours (before sealing the chambers) in order to ensure uniform starting temperature.

The respiratory gases (O_2 and CO_2) were monitored throughout the entire storage period. Gas samples of 0.5 ml each were drawn from each of the chambers using a syringe at least twice a day for spinach stored at 2 and 8°C and after every 2 hours for spinach stored at 15 and 23°C. The gas samples were analyzed in a Gas Chromatograph (GC), SRI 8610A, using a computer software program called "PEAK SIMPLE INTEGRATION METHOD". The GC was installed with a thermal conductivity detector, set at 45°C and a temperature detector set at 100°C. The experiment was terminated when the oxygen level had gone below 2 %.

The respiration rate and Respiratory Quotient of spinach were determined by taking into account the following parameters:

(i) Free volume of chamber (V)

(ii) Mass of spinach (m_s) at time t and ($t+\Delta t$)

(iii) Volume of O_2 and CO_2 at time t, ($V_{o_2}(t)$ and $V_{co_2}(t)$

(iv) Volume of O_2 and CO_2 at time ($t+\Delta t$), ($V_{o_2}(t+\Delta t)$ and $V_{co_2}(t+\Delta t)$

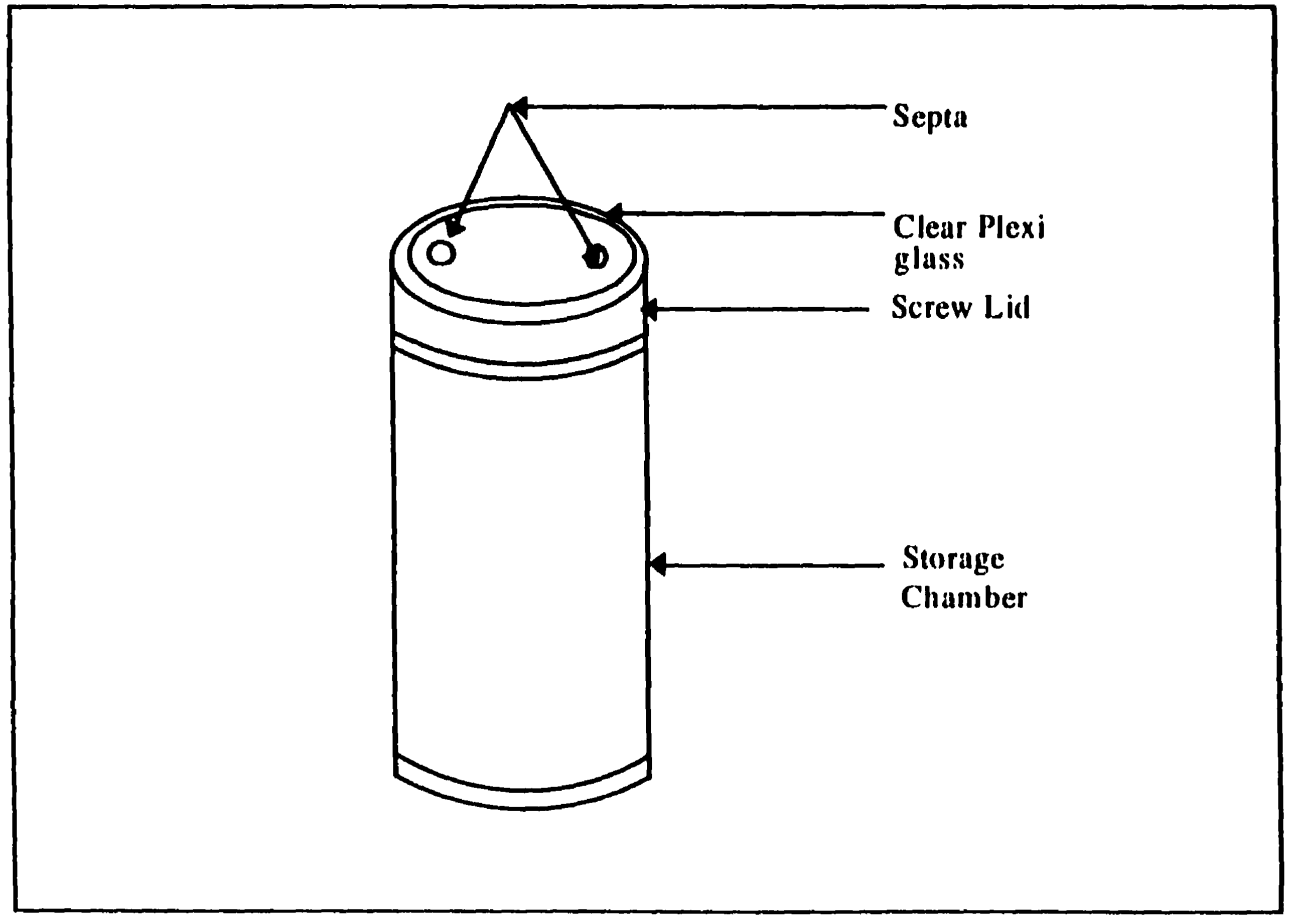

Figure 4.1 Schematic Diagram of the Storage Chamber for Measuring Respiration Rate of Spinach

(v) Partial pressure inside the chamber at time t and t+Δt

The rates mass of O_2 consumed and mass of CO_2 evolved were calculated using the following equations (Ratti et al., 1993):

$$r_{O_2} = -(M_{o_2}(t+\Delta t) - M_{o_2}(t)) \quad (4.1)$$

$$r_{co_2} = (M_{co_2}(t+\Delta t) - M_{co_2}(t)) \quad (4.2)$$

The Respiratory Quotient (RQ) was calculated using the following Equation (Ramachandra, 1995):

$$RQ = \frac{r_{co_{2_{max}}}}{r_{o_{2_{max}}}} \quad (4.3)$$

4.1.1 Modelling of Respiration Rate of Spinach

Modelling of the respiration rate was based on the principles of enzymatic reactions in an unpurified state. A mathematical model for predicting respiration rate of cauliflower developed by Ratti et al. (1993) was adopted and modified for spinach (Equation 3.11). The constants of the equation were obtained by non linear regression analysis of the experimental data using Sigma Plot curve fitting software (Jandel, 1992).

4.2 CA Storage of Spinach Using Diffusion Channel System

Two sets of experiments were carried out consecutively, in a cold room set at 2°C, to asses the suitability of the diffusion channel system in creating proper CA conditions for storage of spinach. Quebec grown spinach was used for the experiments. Preparation of the spinach samples was the same as described in Section 4.1. Both experiments comprised of 18 treatments. The storage chambers were made from PVC pipe sections of 0.5 m in height and an internal diameter of

0.25 m, with both ends sealed with plexi glass, 6 mm thick. The top lids were installed with diffusion channels made either from copper or PVC tubings, of different lengths and cross sectional areas (Figure 4.2). Air tightness of the chambers was tested as described in Section 4.1.

Quality of spinach after both experiments, was evaluated in terms of colour, mass loss, incidence of diseases and presence of off odours. The evaluation where possible was done both by physical measurements as well as by visual observations. Colour was measured by a Minolta chromameter CR-200b on three spinach samples from each of the treatments. The values were interpreted on a tristimulus colour analyzer which describes chromaticity in terms of Value, Hue, and Chroma and are represented by the letters L, a and b respectively. The significance in the differences among the treatment means was determined by Duncan's Multiple Range Test (DMRT). The overall appearance of the spinach from the individual treatments was assessed visually. Mass loss was presented as physiological and trimming mass losses. The former was calculated as the difference between the initial and the final mass of spinach after storage. The latter was the total mass of spinach which was discarded as being in an unsaleable condition. Both the physiological and trimming mass losses were calculated as a percentage of the initial mass of spinach. Incidence of diseases and presence of off odours were noted upon opening the chambers.

Details of the individual experiments were as follows:

Experiment 1

Chambers for this experiment were installed with diffusion channels of six varying lengths , 0.006 m, 0.03 m, 0.07 m, 0.12 m, 0.18 m and 0.25 m and three varying cross sectional areas of $4*10^{-6}$ m^2, $1.8*10^{-5}$ m^2 and $1.15*10^{-4}$ m^2. Two septa were installed on the top lids as gas sampling ports. An average of 1.6 kg of spinach was stored in each of the chambers. To ensure uniform

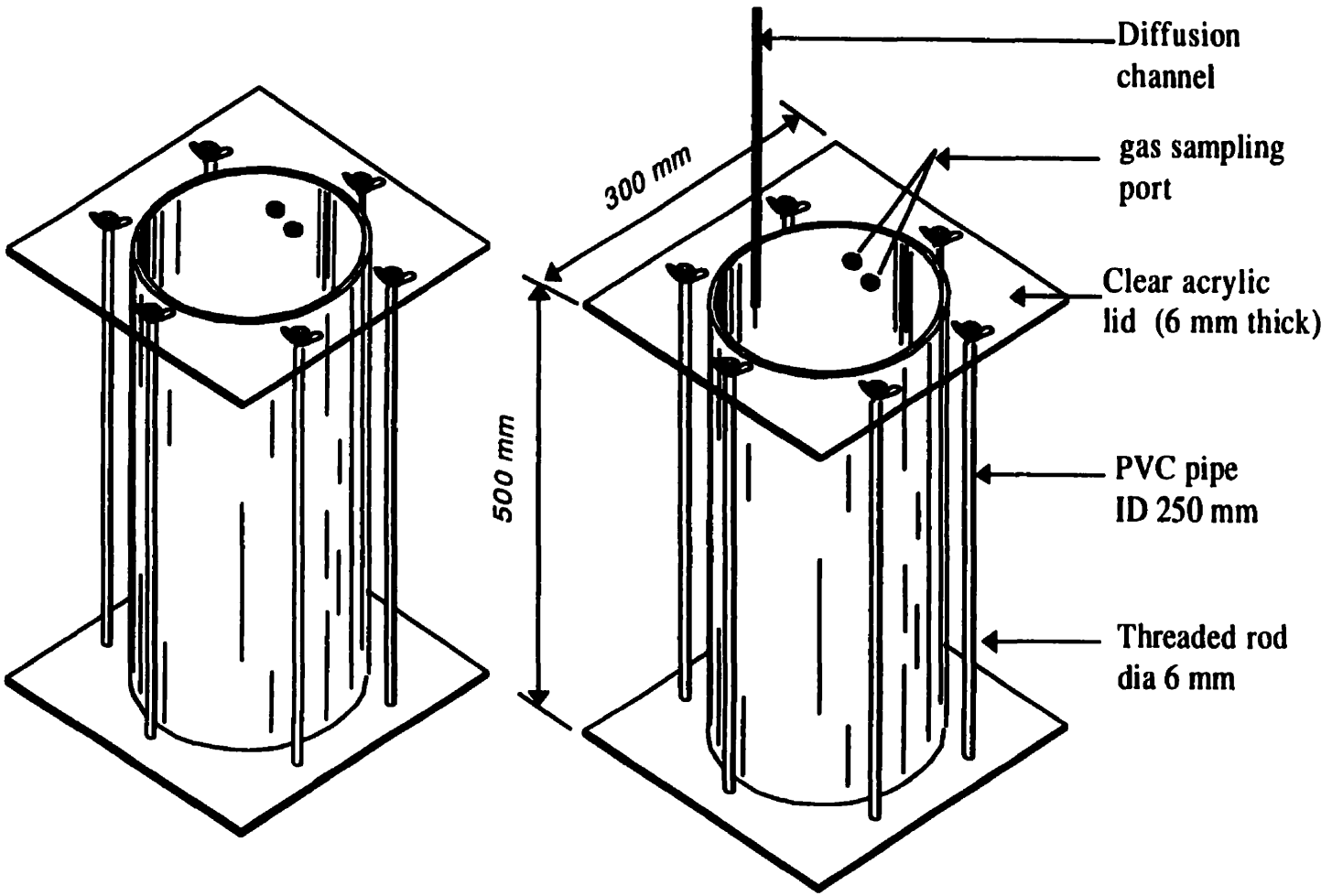

Figure 4.2 Schematic Diagrams of the Experimental Storage Chambers (with and without diffusion channel)

distribution of the air in the chambers, cylindrical wire meshes, 0.5 m in height and 0.1 m in diameter (Figure 4.3) were inserted in the middle of the chambers, prior to packing the spinach. In each of the chambers, 250 g of lime ($Ca(OH)_2$) was placed at the bottom to scrub off CO_2. The experiment ran for 30 days. The performance of the diffusion channel system in storage of spinach was assessed based on the trend of oxygen evolution in the chambers. Oxygen level in the chambers was monitored on a daily basis in the same way as described in Section 4.1. To facilitate establishment of CA steady state conditions, the experimental chambers were flushed with nitrogen. Gas samples were drawn after the flushing for analysis so as to make sure that the oxygen level had decreased from the atmospheric level of 21% to at least close to the CA standard level of 3%.

Experiment 2

Spinach was stored for 49 days in chambers similar to the ones used in Experiment 1. The chambers were divided into nine pairs and installed with diffusion channels of three varying lengths, 0.043 m, 0.11 m and 0.19 m of three different cross sectional areas of $9*10^{-6}$ m^2, $4*10^{-5}$ m^2 and $1.7*10^{-4}$ m^2. Unlike in experiment 1, cylindrical wire meshes were not included in the chambers. CO_2 was scrubbed using 250 g of lime from one of each of the pairs. Oxygen and carbon dioxide were monitored as described in the previous experiments. Comparison was made between the steady state levels of oxygen in CO_2 scrubbed and unscrubbed chambers. Gas was sampled from the top and the bottom side of the chambers installed with diffusion channels of ($9*10^{-6}$ m^2) in CSA so as to find the distribution pattern of the gases in the chambers. The performance of the diffusion channel system was evaluated based on the ability of the diffusion channels in maintaining steady state levels of both carbon dioxide and oxygen.

4.2.1 Modelling Diffusion Channel Length for CA Establishment

Modelling of the length of a diffusion channel for establishing desired CA

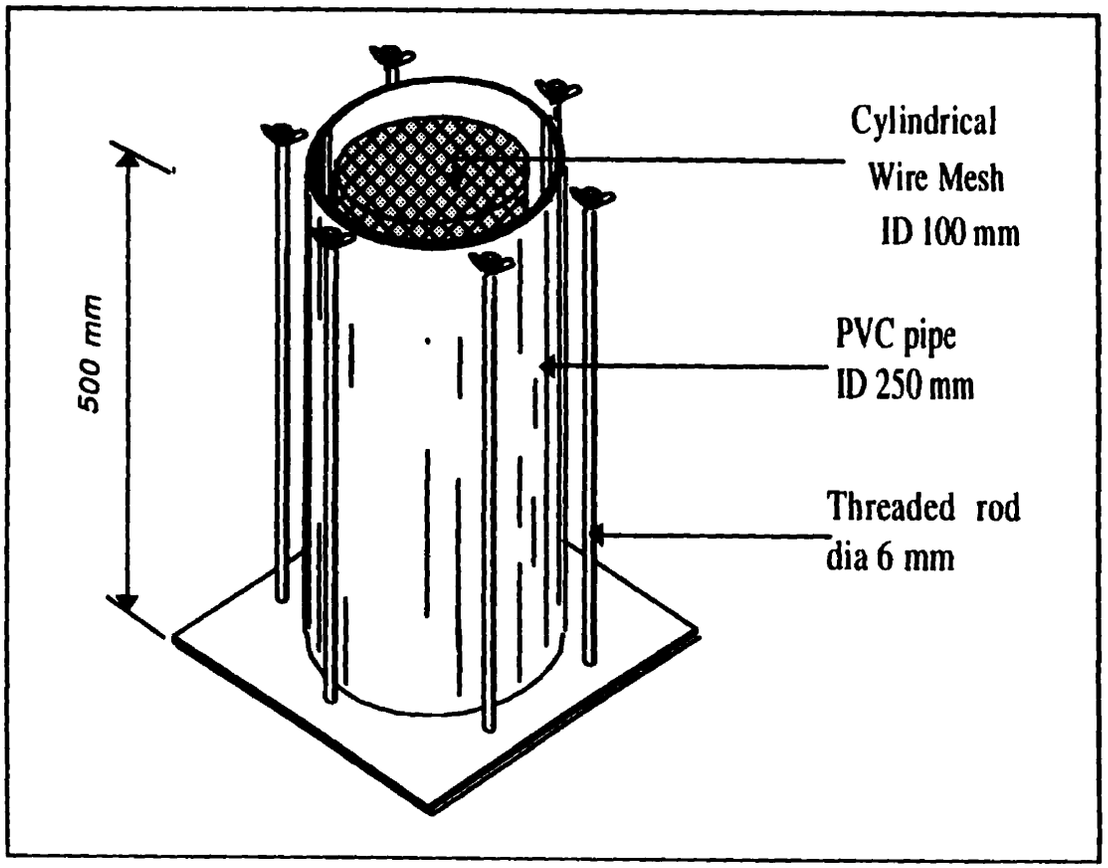

Figure 4.3 Storage Chamber with a Cylindrical Wire Mesh

conditions for storage of spinach, was based on Ficks first law of diffusion which describes a flux of diffusing substance (O_2) in a stagnant gas (N_2). A model developed by Ratti et al. (1994) on diffusion of oxygen through diffusion channel in storage of cauliflower was adopted and slightly modified to predict the length of a diffusion channel which would maintain desired O_2 concentration under steady state conditions, for a given mass of spinach (equation 3.29). The constants in the equation were obtained through curve fitting process by Jandel (1992).

CHAPTER V

RESULTS

5.1 Effect of Temperature on Respiration Rate

The results on the effect of temperature on the respiration rate of spinach and the constants for the respiration model are presented in this section.

Respiration rate of spinach decreased as time progressed. The respiration rates of spinach measured at four levels of storage temperature (2, 8, 15 and 23°C) are presented in Figure 5.1 as a function of time. Oxygen levels in the chambers declined with time whereas carbon dioxide levels increased. The typical trend of the gas levels in the storage chambers is shown in Figure 5.2 as a function of time for a storage temperature of 2°C. The respiration rate increased with an increase in oxygen concentration and subsequently decreased with high carbon dioxide concentration. The progression of the respiration rate as a function of oxygen and carbon dioxide concentrations at 2°C, are shown in Figures 5.3 and 5.4 respectively. Similar progression are compared among the storage temperature levels studied and are shown in Figures 5.5 and 5.6. It is apparent that there was more than a tenfold increase in the respiration rate when temperature was increased from 2°C to 23°C.

The Respiratory Quotient calculated at 2°C as a ratio of the rate of mass of CO_2 evolved to the rate of mass of O_2 consumed in the respiration process, is presented in Figures 5.7, 5.8 and 5.9, as a function of time, O_2 and CO_2 concentrations, respectively. The RQ increased with time. It also increased with subsequent decrease in oxygen concentration and vice versa. However, there was a dramatic increase in the RQ when Oxygen concentration level decreased below 48 mg/litre. An increase in the RQ was also noted with an increase in CO_2 concentration. The RQ increased drastically with CO_2 concentration above 280

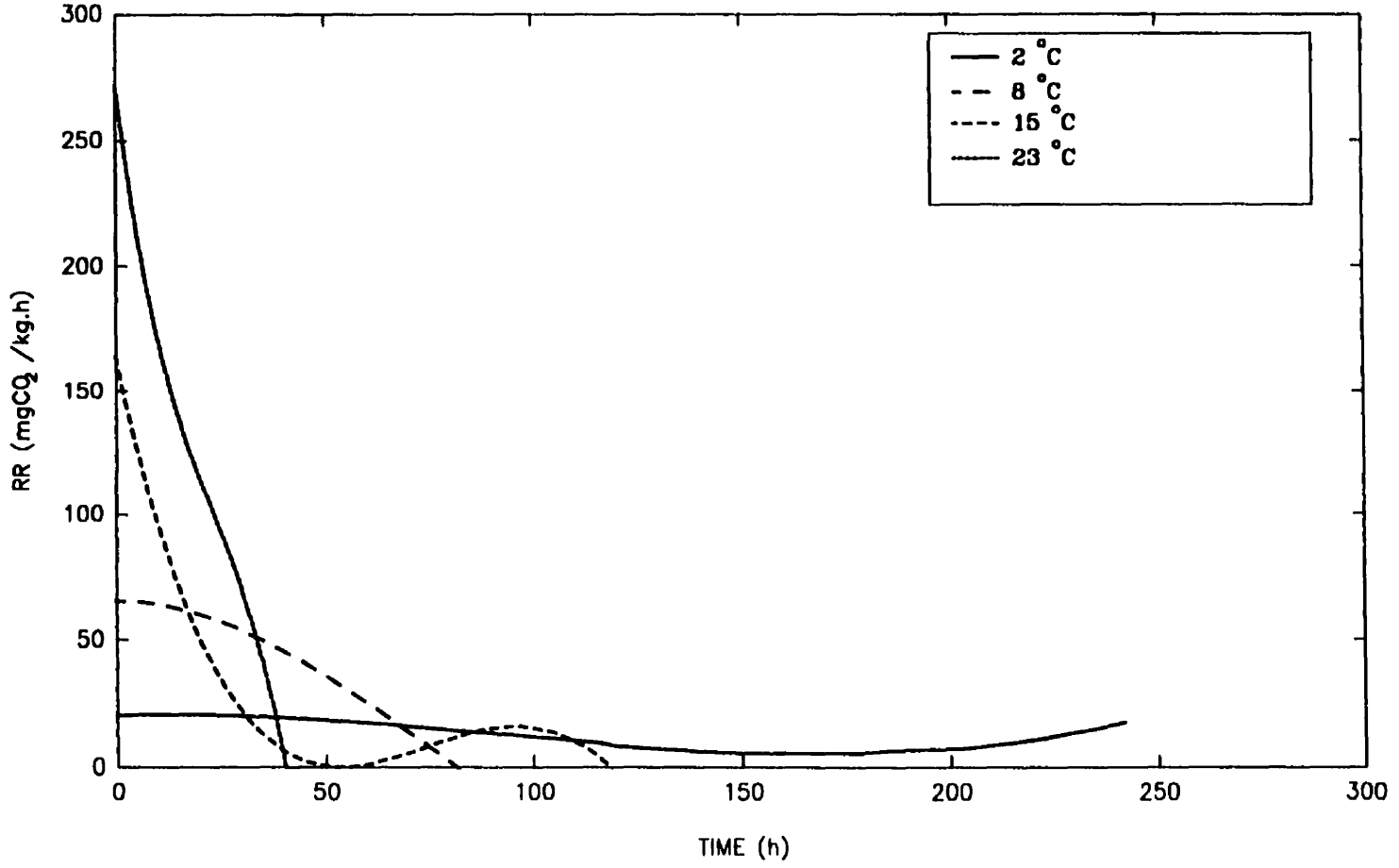

Figure 5.1 Respiration Rate of Spinach at Four Levels of Storage Temperature with Time.

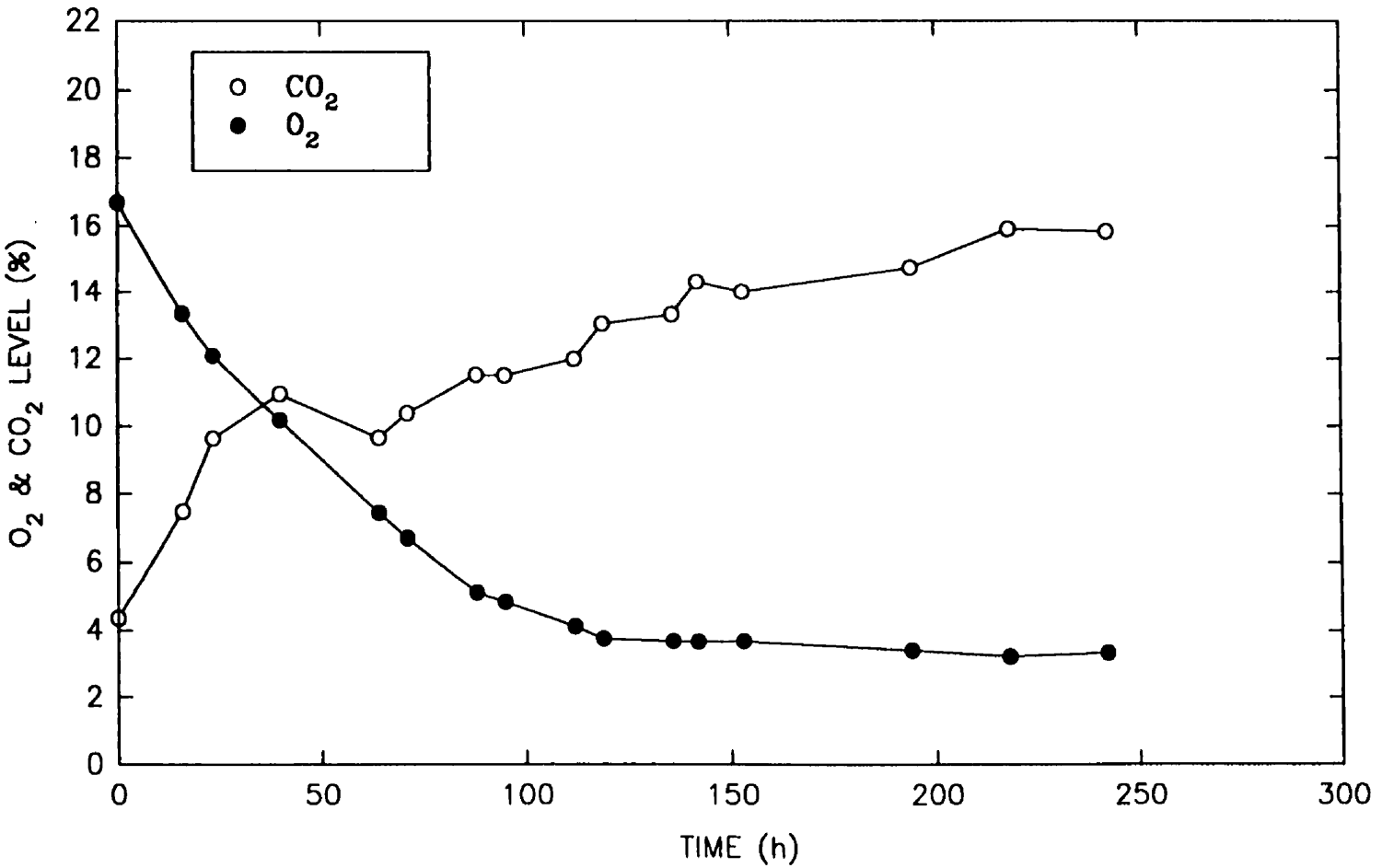

Figure 5.2 Progression of O_2 and CO_2 in Sealed Storage Chambers at 2°C as a Function of Time.

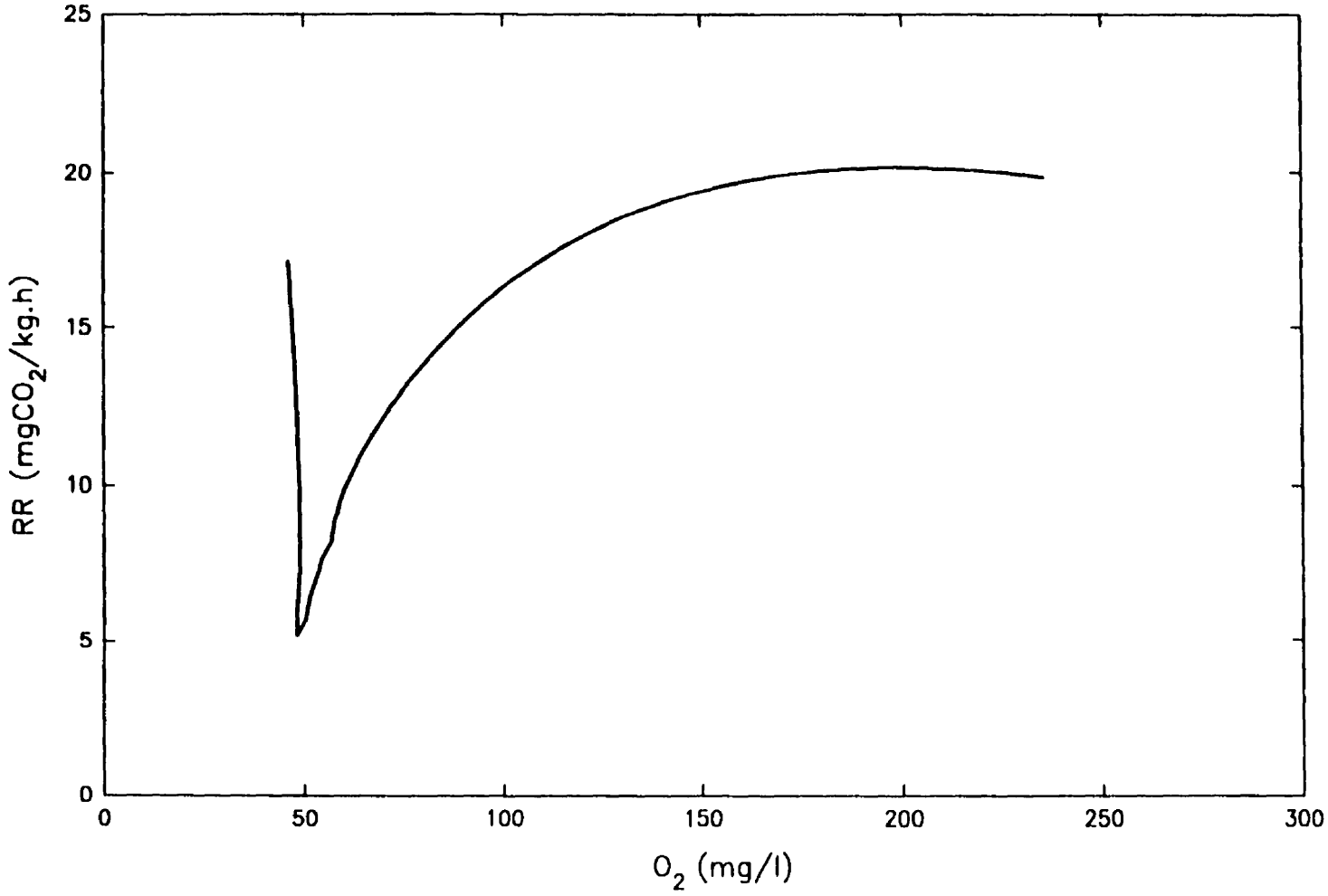

Figure 5.3 Respiration Rate of Spinach at 2°C as a Function of O_2 Concentration.

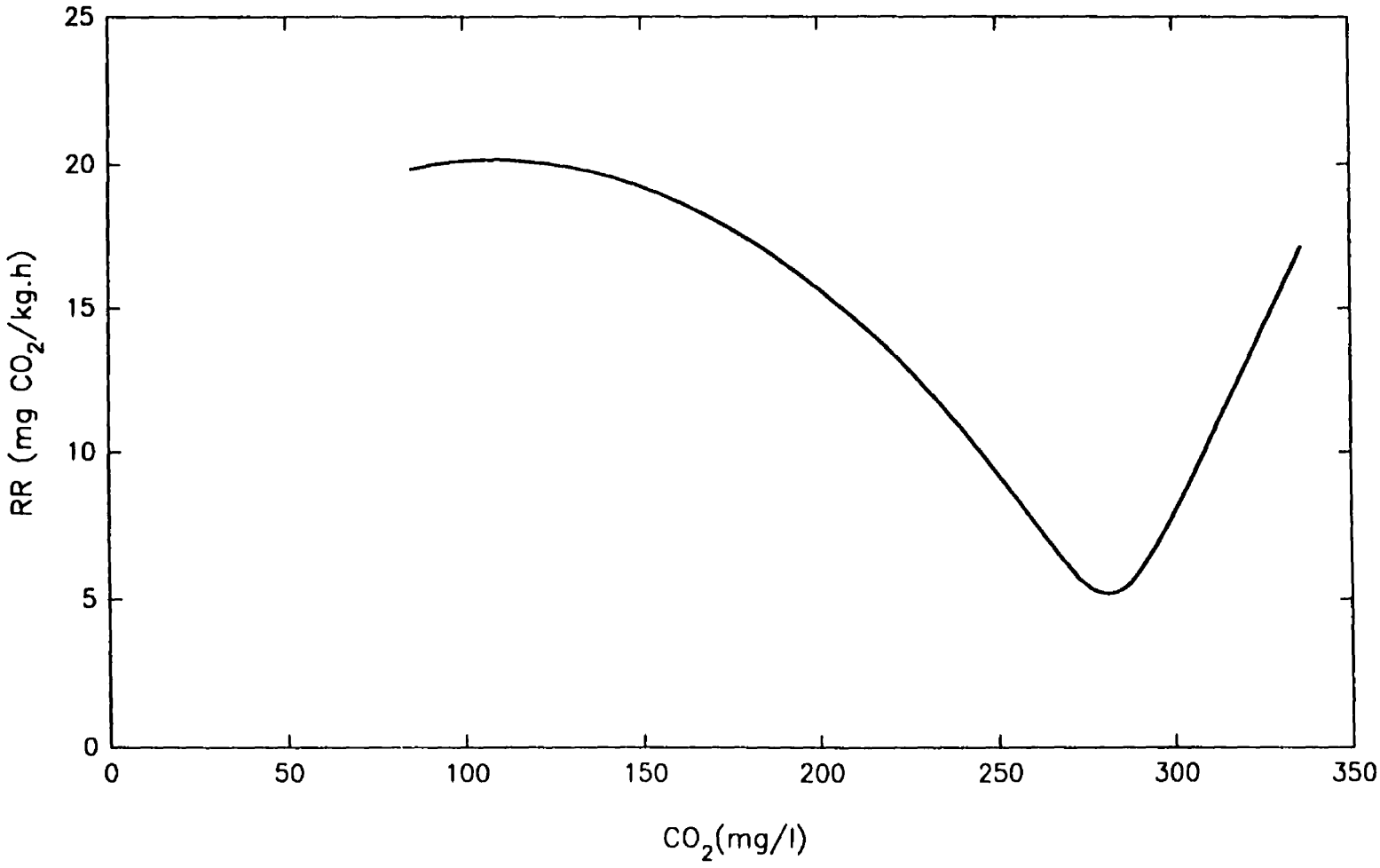

Figure 5.4 Respiration Rate of Spinach at 2°C as a Function of CO_2 Concentration.

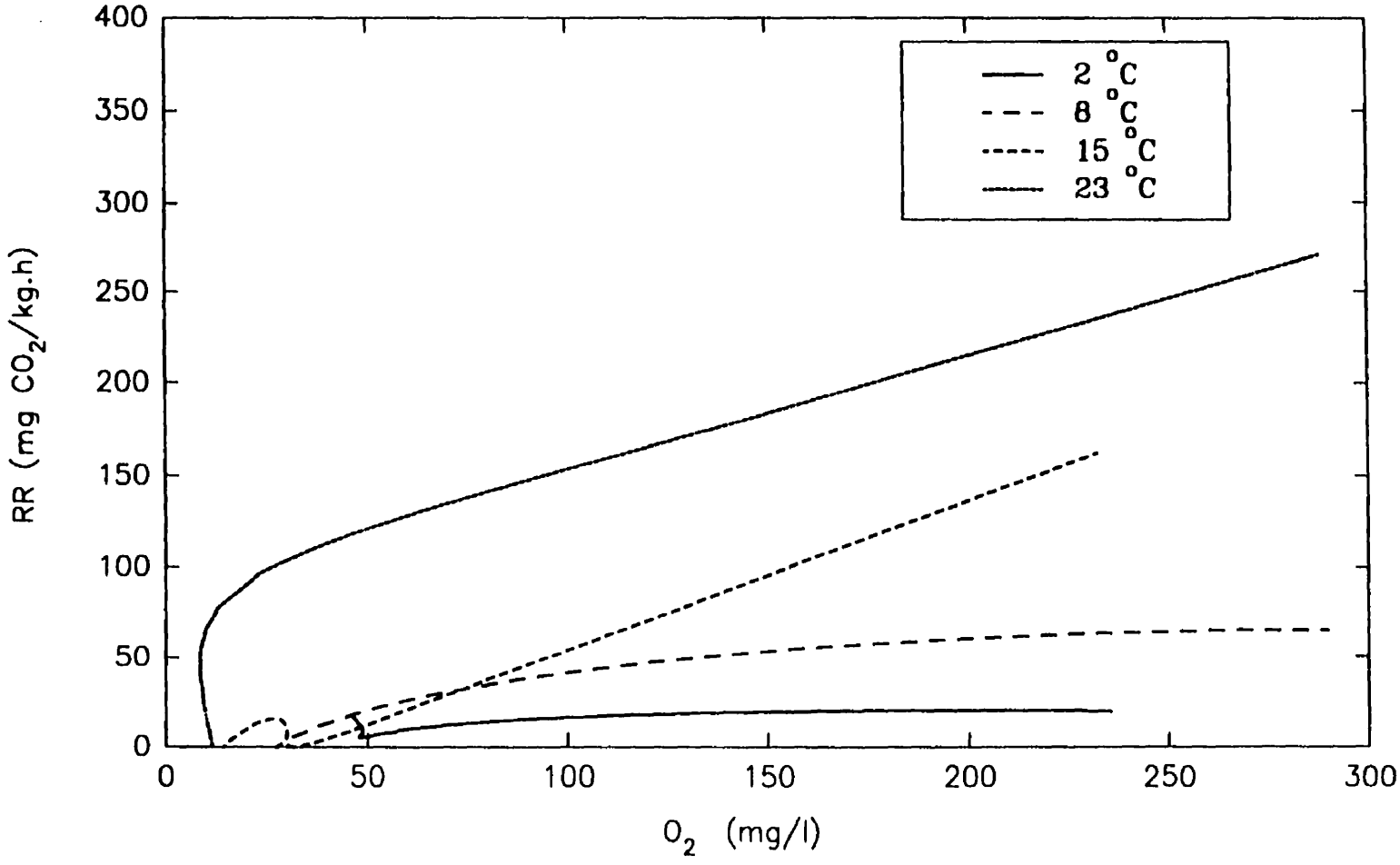

Figure 5.5 Respiration Rate of Spinach at Four Levels of Storage Temperature as a Function of O_2 Concentration.

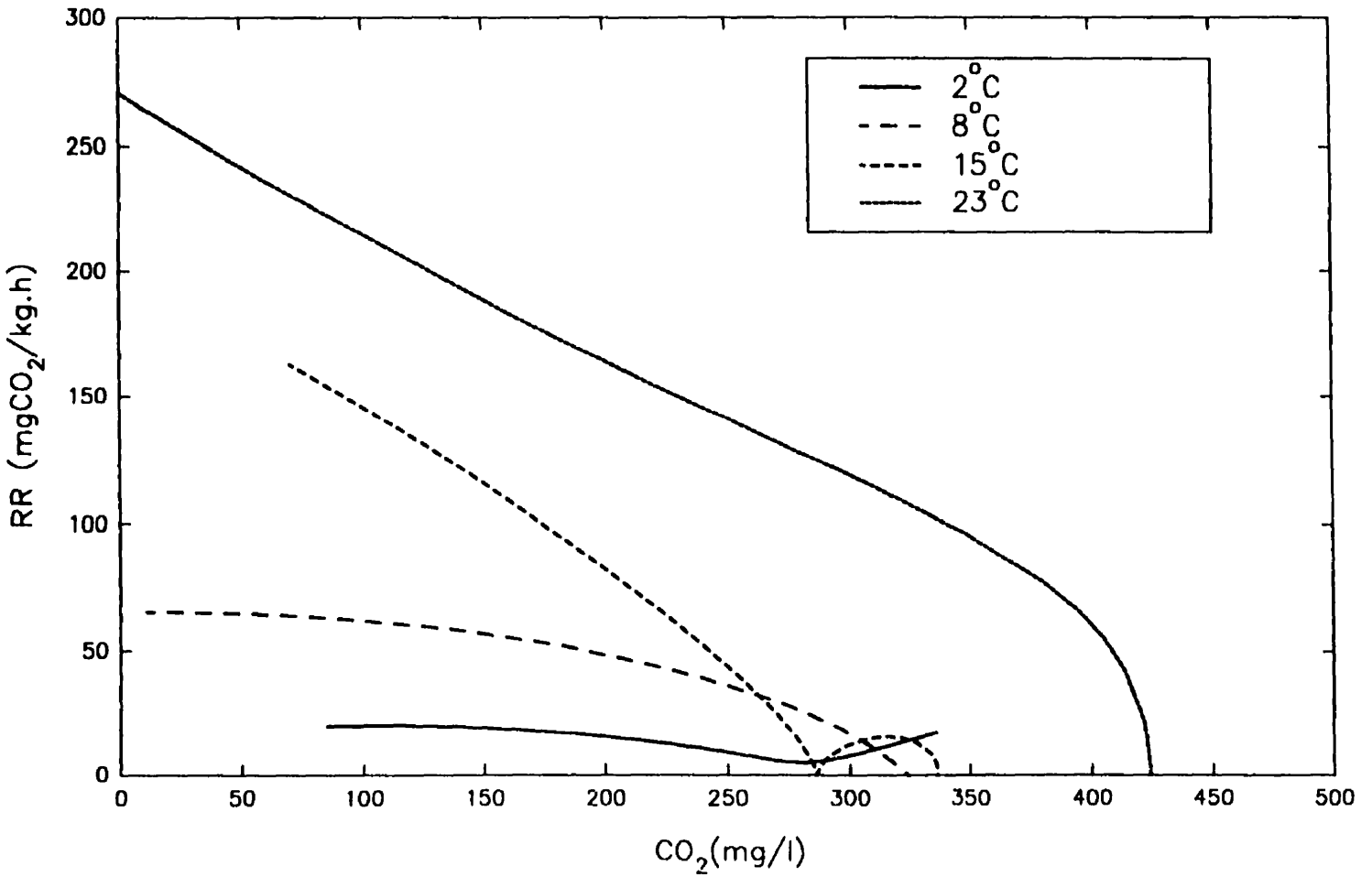

Figure 5.6 Respiration Rate of Spinach at Four Levels of Temperature as a Function of CO_2 Concentration.

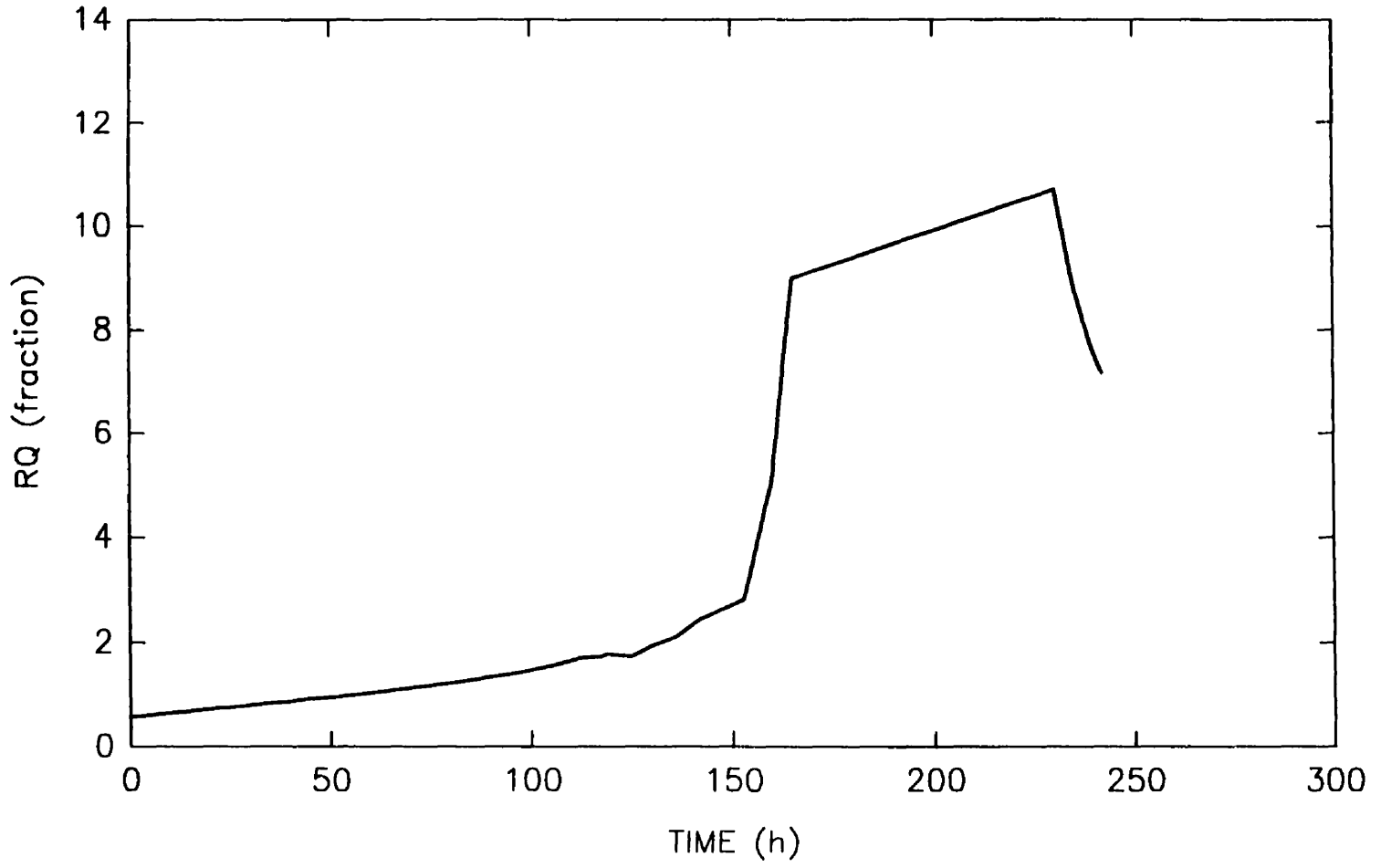

Figure 5.7 Respiratory Quotient of Spinach at 2°C as a Function of Time.

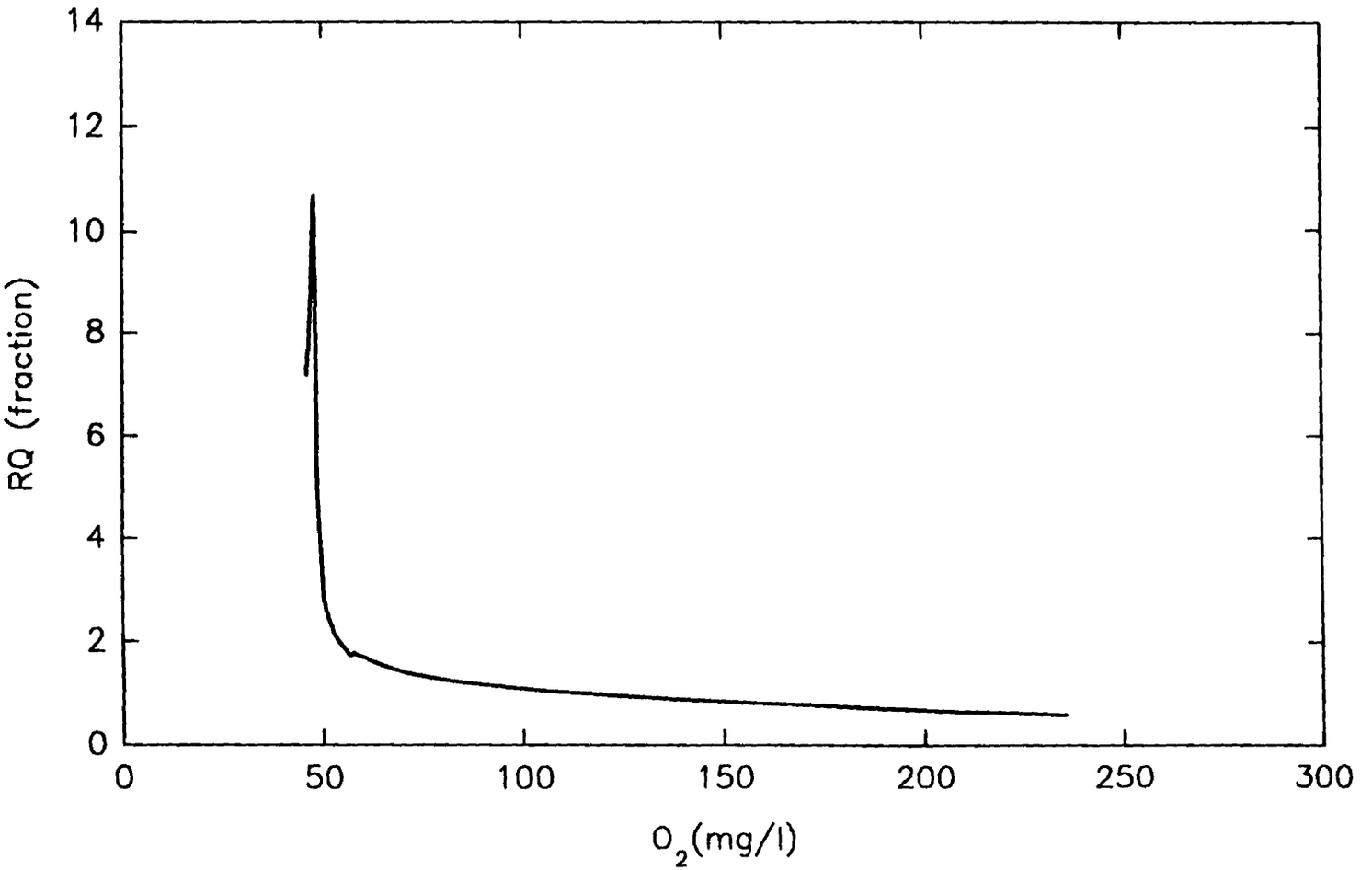

Figure 5.8 Respiratory Quotient of Spinach at 2°C as a Function of O_2 Concentration.

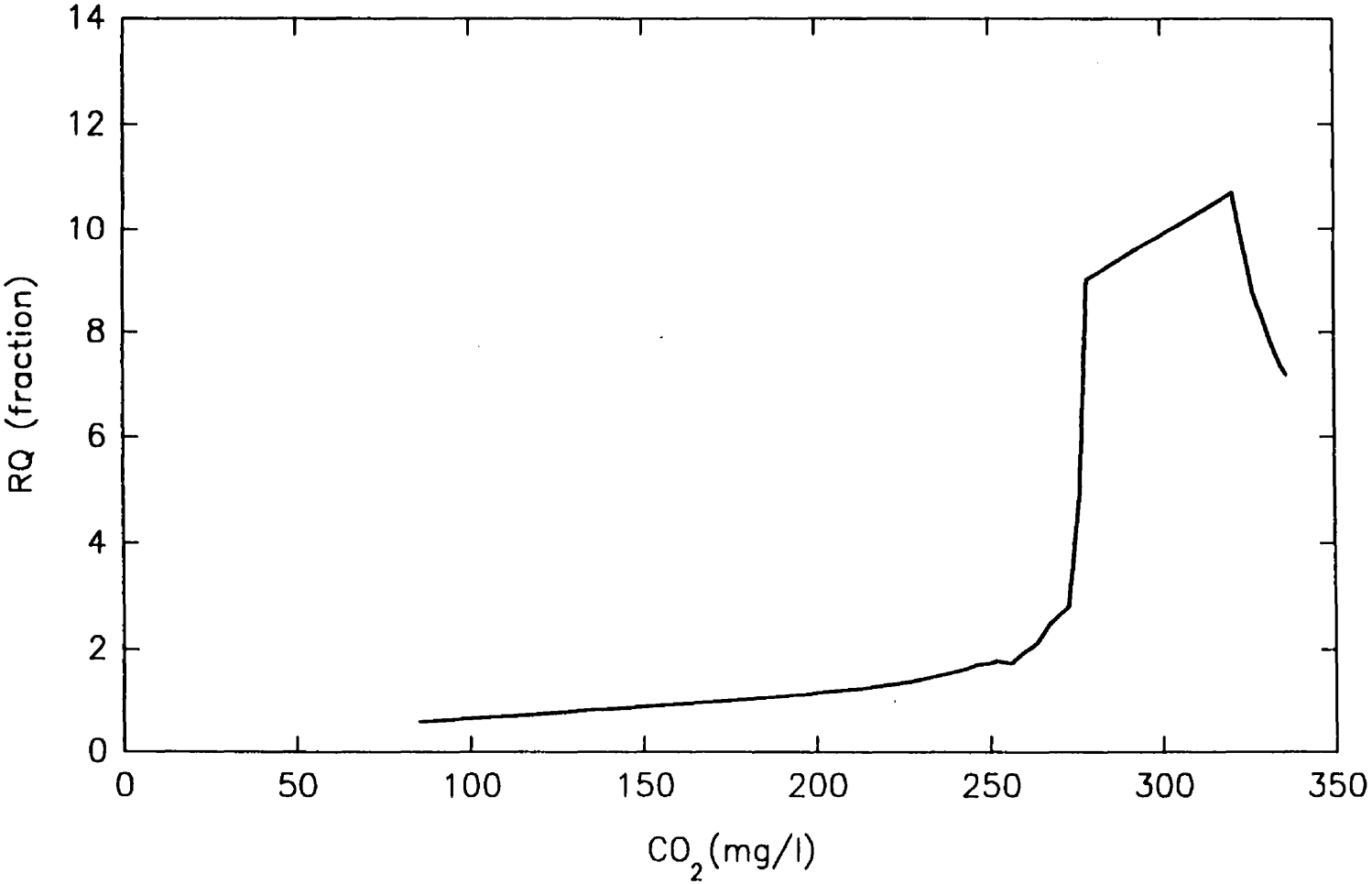

Figure 5.9 Respiratory Quotient of Spinach at 2°C as a Function of CO_2 Concentration.

mg/litre. The changes in the RQ values due to changes in oxygen and carbon dioxide concentrations were the same for the other temperatures.

5.1.1 Model for The Respiration Rate of Spinach

Non linear regression of the experimental data yielded constants A_1, A_2, A_3, b_1, b_2 and b_3 and these were used to calculate inhibition constants K_1, K_2 and K_3 at the four storage temperatures. The values of the Constants are presented in Table 5.1. The calculated K values are shown in Tables 5.2a and b for respiration expressed as the rate of carbon dioxide evolved and oxygen consumed respectively. The best fit curves for the respiration model are shown in figures 5.10 and 5.11 for the respiration rate calculated on the basis of CO_2 evolution rate and O_2 consumption rate respectively. The final model of the respiration rate of spinach for practical purposes is given by:

$$r = \frac{A_1 \exp(b_1/T) * C_{O_2}}{(A_2 \exp(b_2/T) + C_{O_2}} \qquad (5.1)$$

Table 5.1 Constants for The Respiration Model

Constant	R_{CO2}	R_{O2}
A_1	-4.72	18.82
A_2	-53.32	-11.72
A_3	$-1.4*10^7$	-38.47
b_1	3109.68	-3726.71
b_2	17114.74	5198.88
b_3	-41811.3	-272.363

Table 5.2a K Values for The Respiration Rate Calculated Based on the Rate of CO_2 Evolution

T (°C)	K_1	K_2	K_3
2	725.21	7447.20	0
8	569.64	1971.84	0
15	435.30	448.70	0
23	325.12	90.00	0

Table 5.2b K Values for the Respiration Rate Calculated Based on the Rate of O_2 Consumption

T (°C)	K_1	K_2	K_3
2	194.6480	1322.69	$7.3 * 10^{-18}$
8	259.9666	883.3832	$7.4 * 10^{-18}$
15	358.8491	563.4470	$7.6 * 10^{-18}$
23	509.0935	345.9141	$7.8 * 10^{-18}$

5.2 Diffusion Channel for Establishing CA Conditions for Storage of Spinach

5.2.1 Gas levels in the Storage Chambers

(a) Oxygen

Oxygen levels in the chambers installed with diffusion channels of six varying lengths (0.006 m, 0.03 m, 0.07 m, 0.12 m, 0.18 m and 0.25 m) for the first set of the diffusion channel storage experiment, are presented in Figures 5.12, 5.13 and 5.14 for the three CSA, $4*10^{-6}$ m^2, $1.8*10^{-5}$ m^2 and $1.15*10^{-4}$ m^2, respectively.

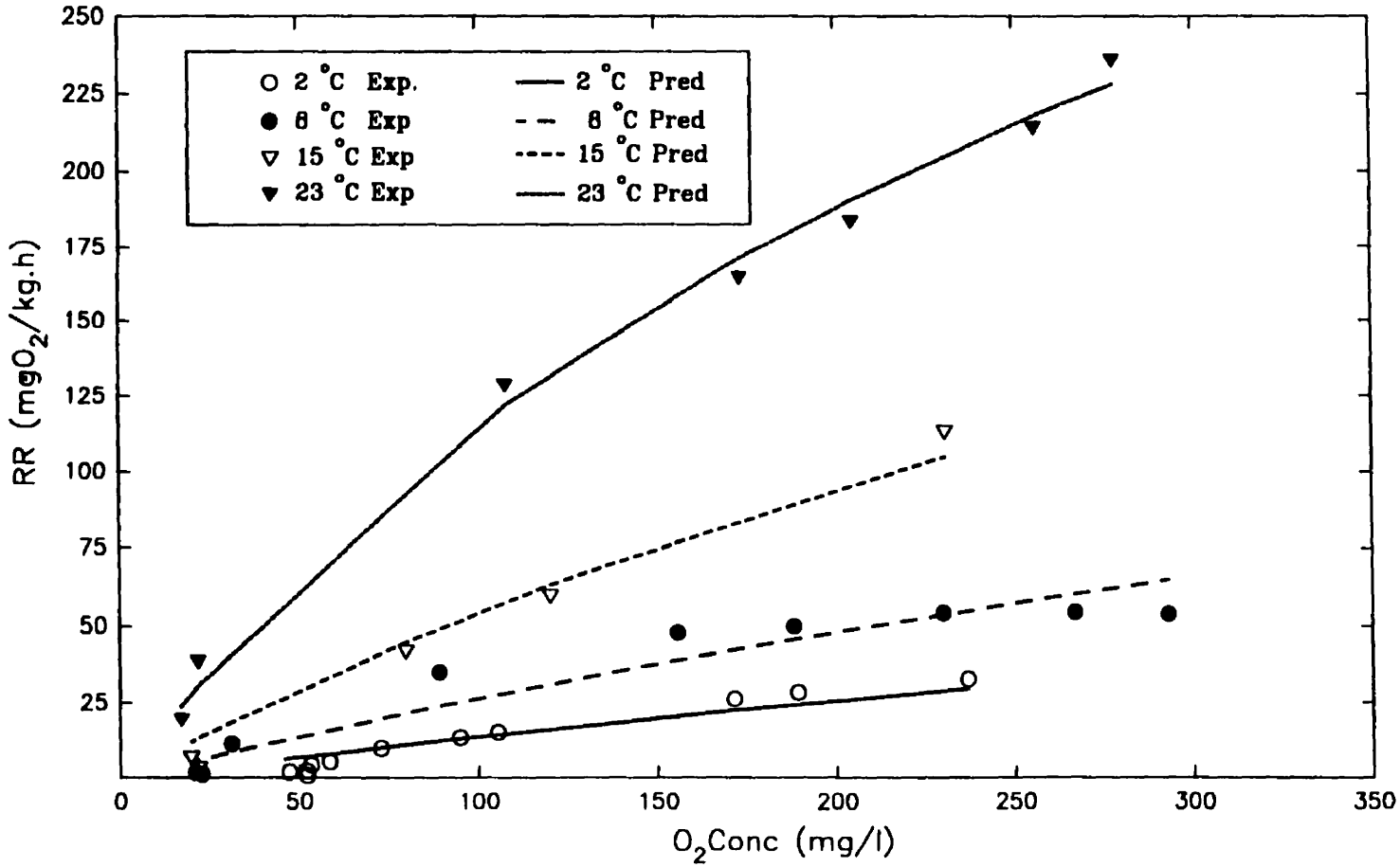

Figure 5.10 Comparison of Predicted and Experimental Respiration Rate of Spinach Calculated on CO_2 Evolution Rate as a Function of O_2 Concentration.

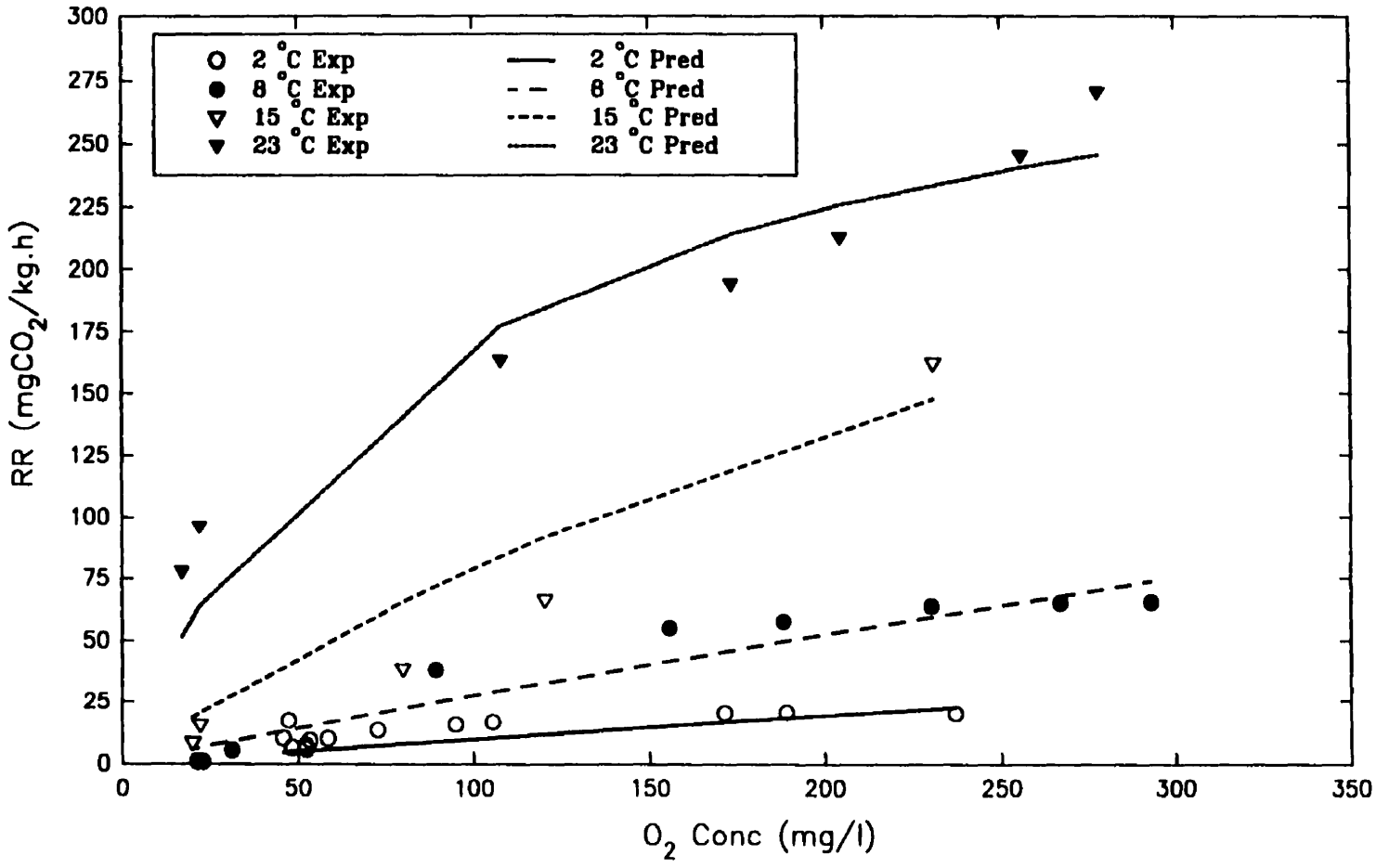

Figure 5.11 Comparison of Predicted and Experimental Respiration Rate of Spinach Calculated on O_2 consumption Rate as a Function of O_2 Concentration.

The steady state oxygen levels were generally high in chambers with diffusion channels of 0.006 m in length. The levels subsequently decreased as the length of the channels increased for all the three cross sectional areas. The levels were quite distinguishable as the cross sectional area of the diffusion channel increased. However, there was no appreciable difference in the O_2 levels when length of the diffusion channel was 0.006 m and 0.25 m despite the changes in the cross sectional area. Figures 5.15 through 5.20 are showing oxygen levels in the chambers installed with channels of the same length but with three varying cross sectional areas. The relationship of cross sectional area and length of the diffusion channel with the steady state oxygen level is shown in Figure 5.21.

It took relatively less days (2 to 4) for O_2 to reach steady state conditions in the chambers with short channels (0.006 m). A similar effect was observed in the chambers with longer channels (0.18 and 0.25 m) of a large cross sectional area ($1.15*10^{-4}$ m^2). The time taken for O_2 to reach steady state is presented in Table 5.3.

Gas levels in the chambers of the second set of the experiment in which sampling ports were installed both on the top and the bottom side, are presented in figures 5.22, 5.23 and 5.24 for the chambers with diffusion channels of $9*10^{6}$ m^2 in CSA and 0.043 m , 0.11 m and 0.19 m in length. There was no noticeable variation in the gas levels when the gas was sampled from top or the bottom side of the chambers.

Comparison of oxygen levels in the CO_2 scrubbed and unscrubbed chambers is presented in Figures 5.25, 5.26 and 5.27 with respect to three cross sectional areas. The O_2 steady state levels were generally high for the CO_2 unscrubbed chambers. The difference was in the range of 1 to 1.5%. Generally, the chambers with channels of shorter lengths of larger cross sectional areas exhibited high O_2 steady state levels. Similar to the previous experiment, the O_2 levels became quite

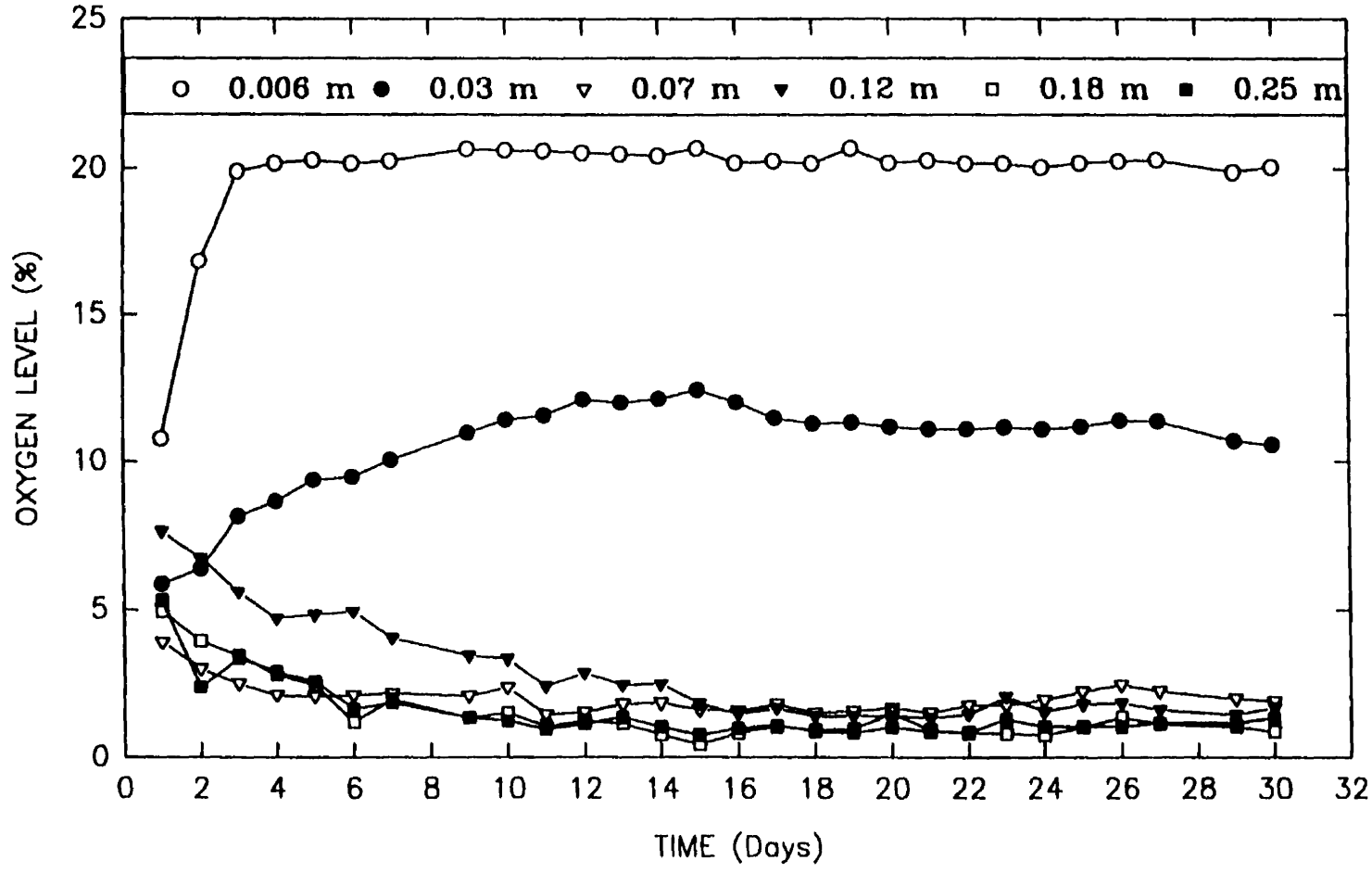

Figure 5.12 Progression of O_2 in Chambers with Channels of Six Varying Lengths and a CSA of $4*10^{-6}$ m^2

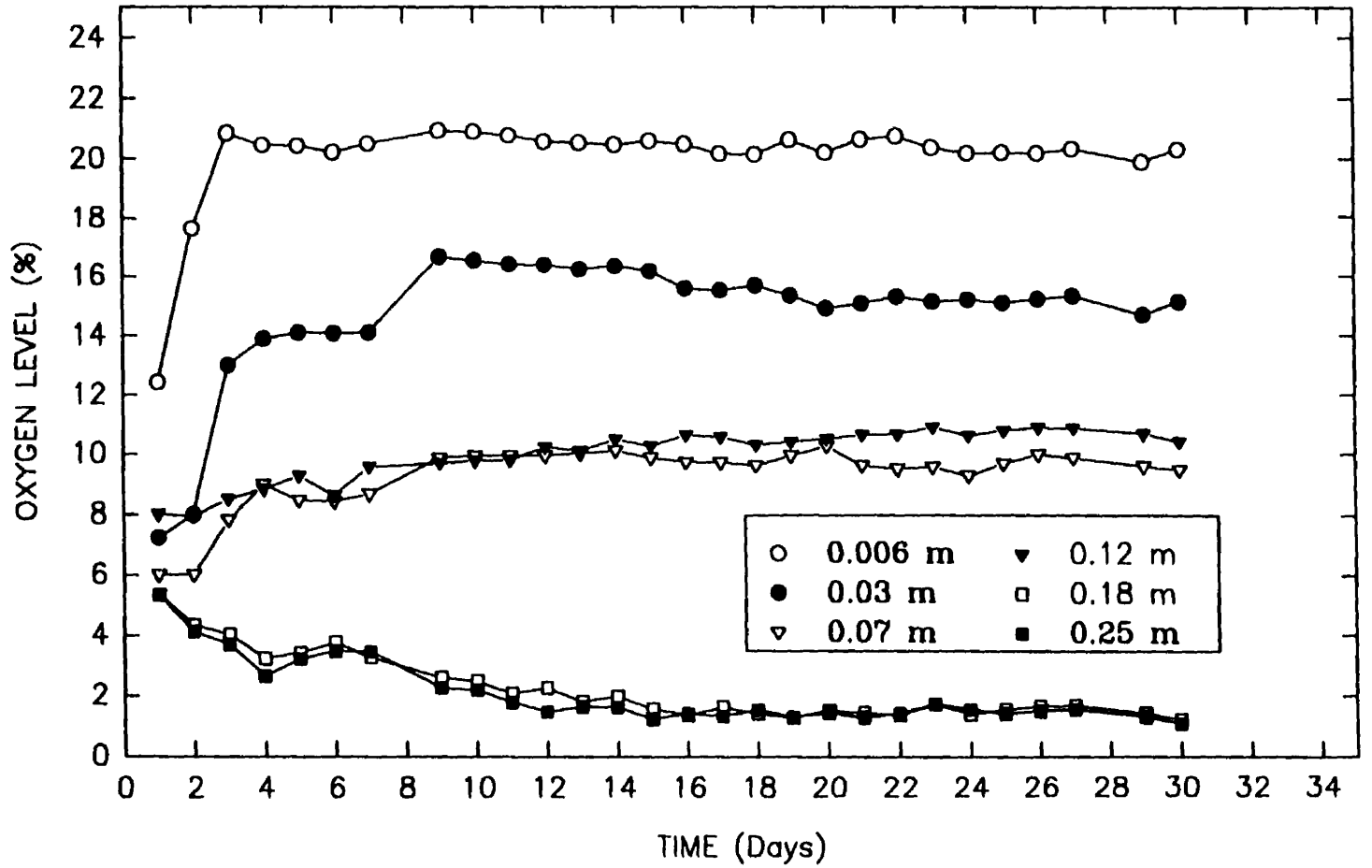

Figure 5.13 Progression of O_2 in Chambers with Channels of Six Varying Lengths and a CSA of $1.8*10^{-5}$ m^2

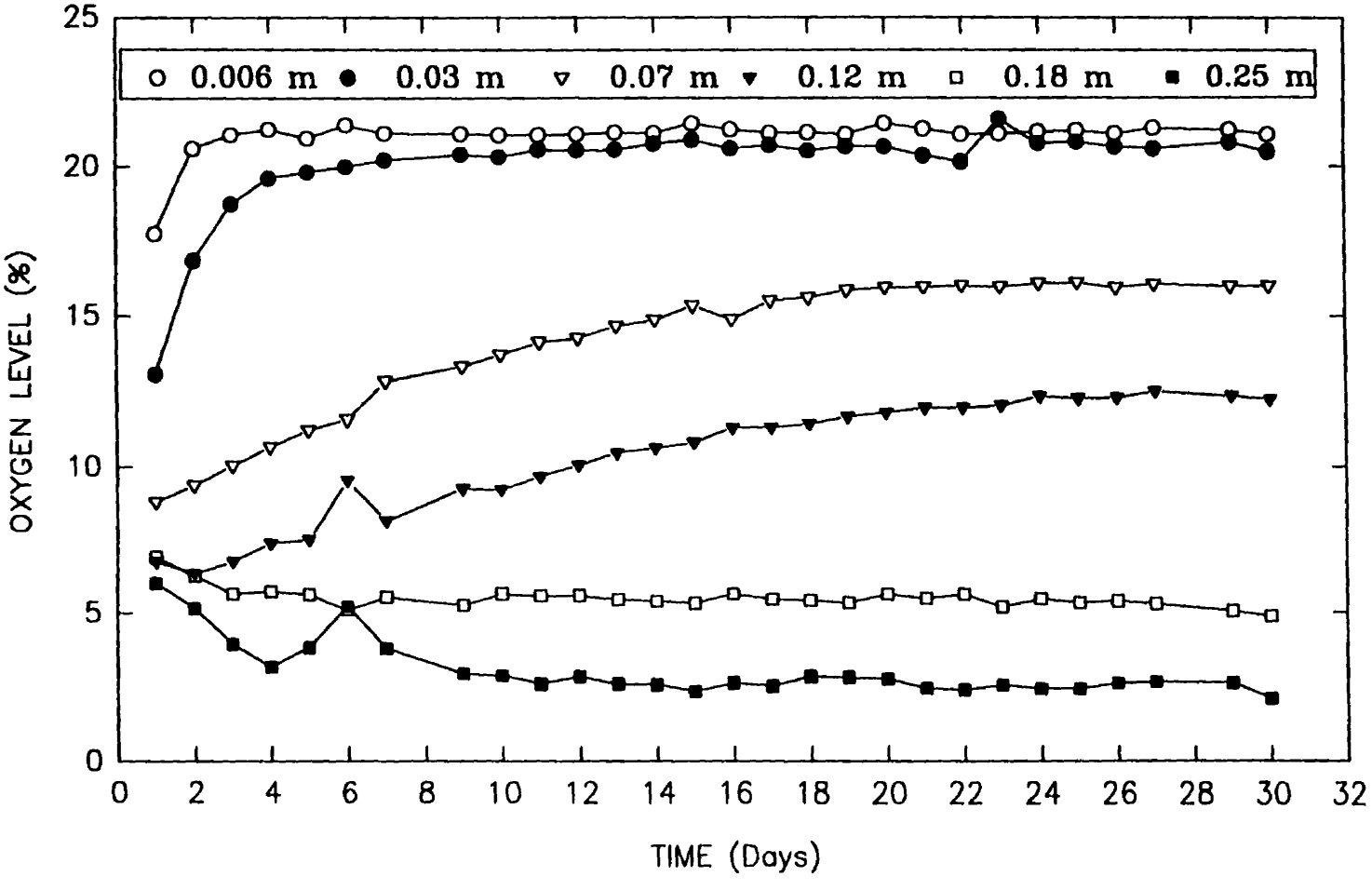

Figure 5.14 Progression of O_2 in Chambers with Channels of Six Varying Lengths and a CSA of $1.15*10^{-4}$ m^2

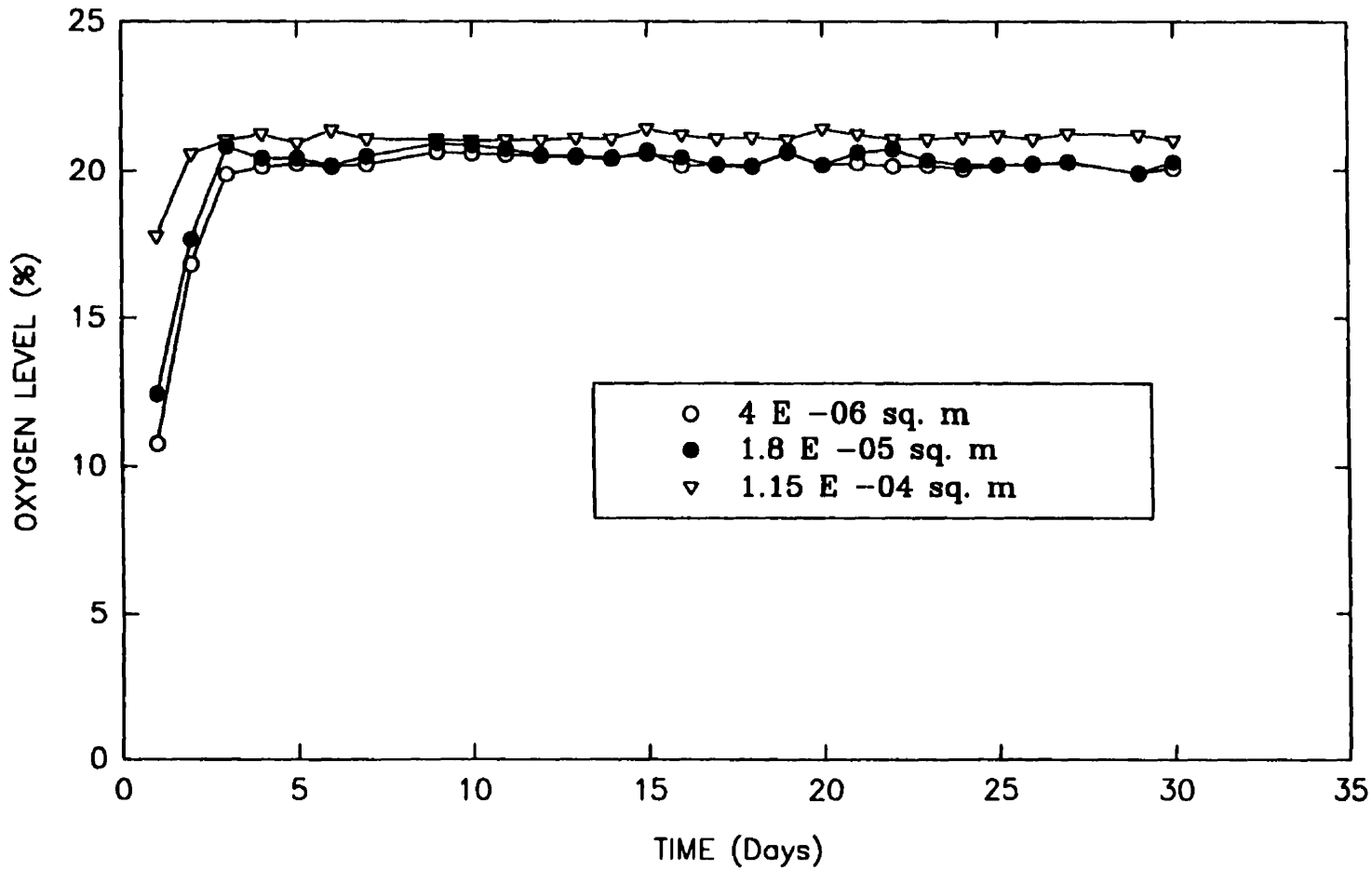

Figure 5.15 Comparison of Progression of O_2 for Chambers with Channels of 0.006 m in Length and Three Varying Cross Sectional Areas.

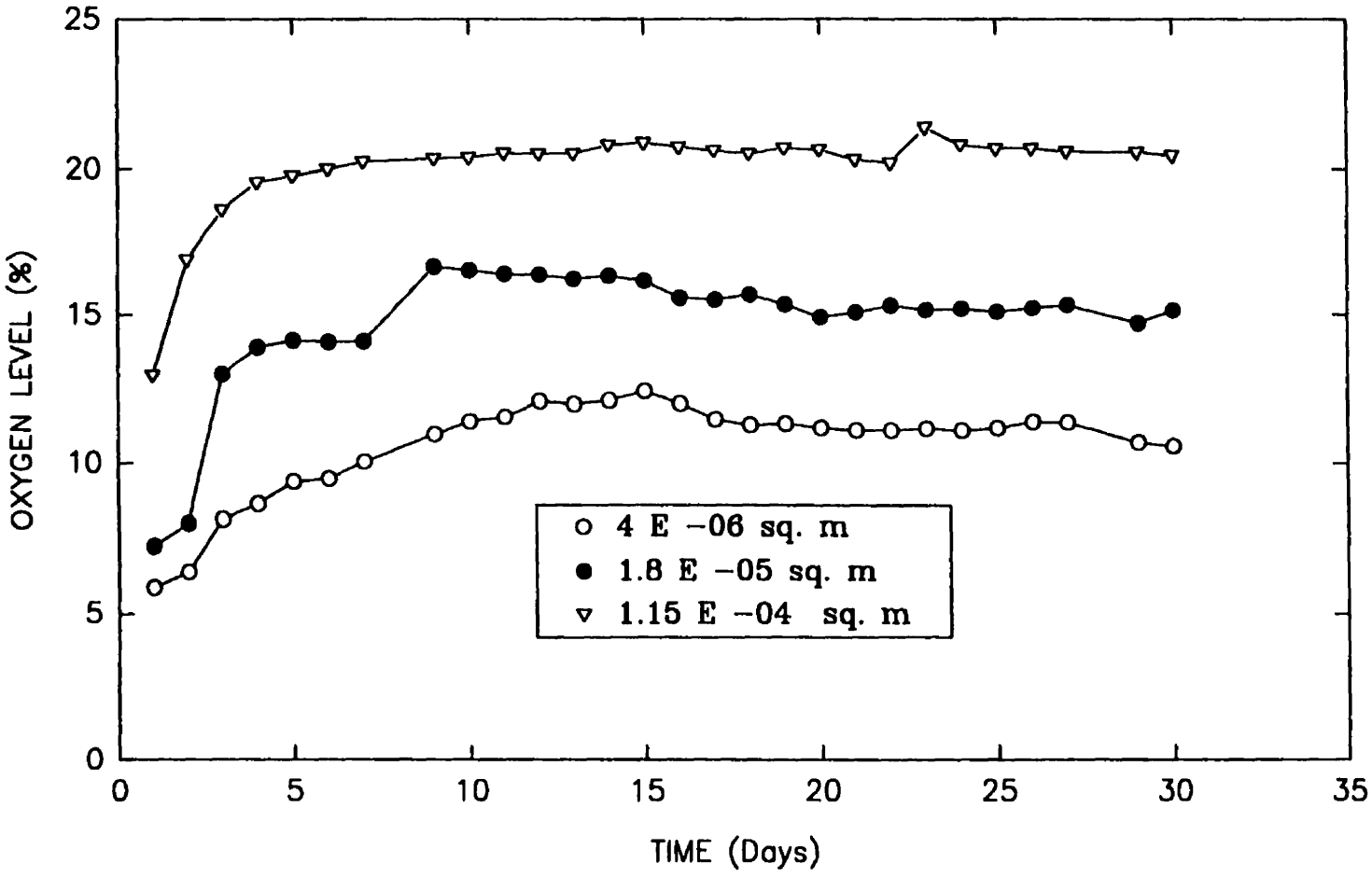

figure 5.16 Comparison of Progression of O_2 for Chambers with Channels of 0.03 m Length and Three Varying Cross Sectional Areas.

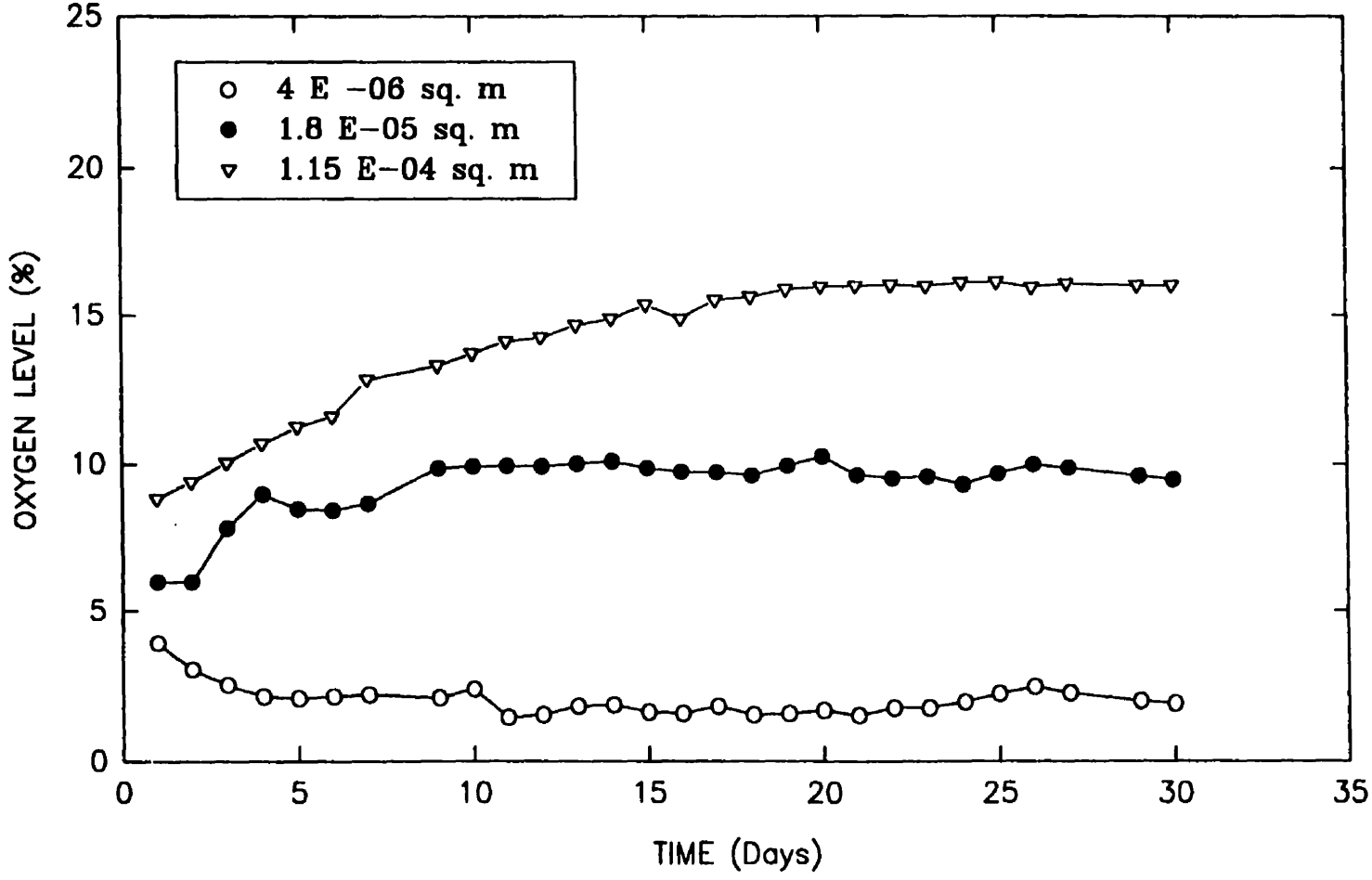

Figure 5.17 Comparison of Progression of O_2 for Chambers with Channels of 0.07 m in Length and Three Varying Cross Sectional Areas.

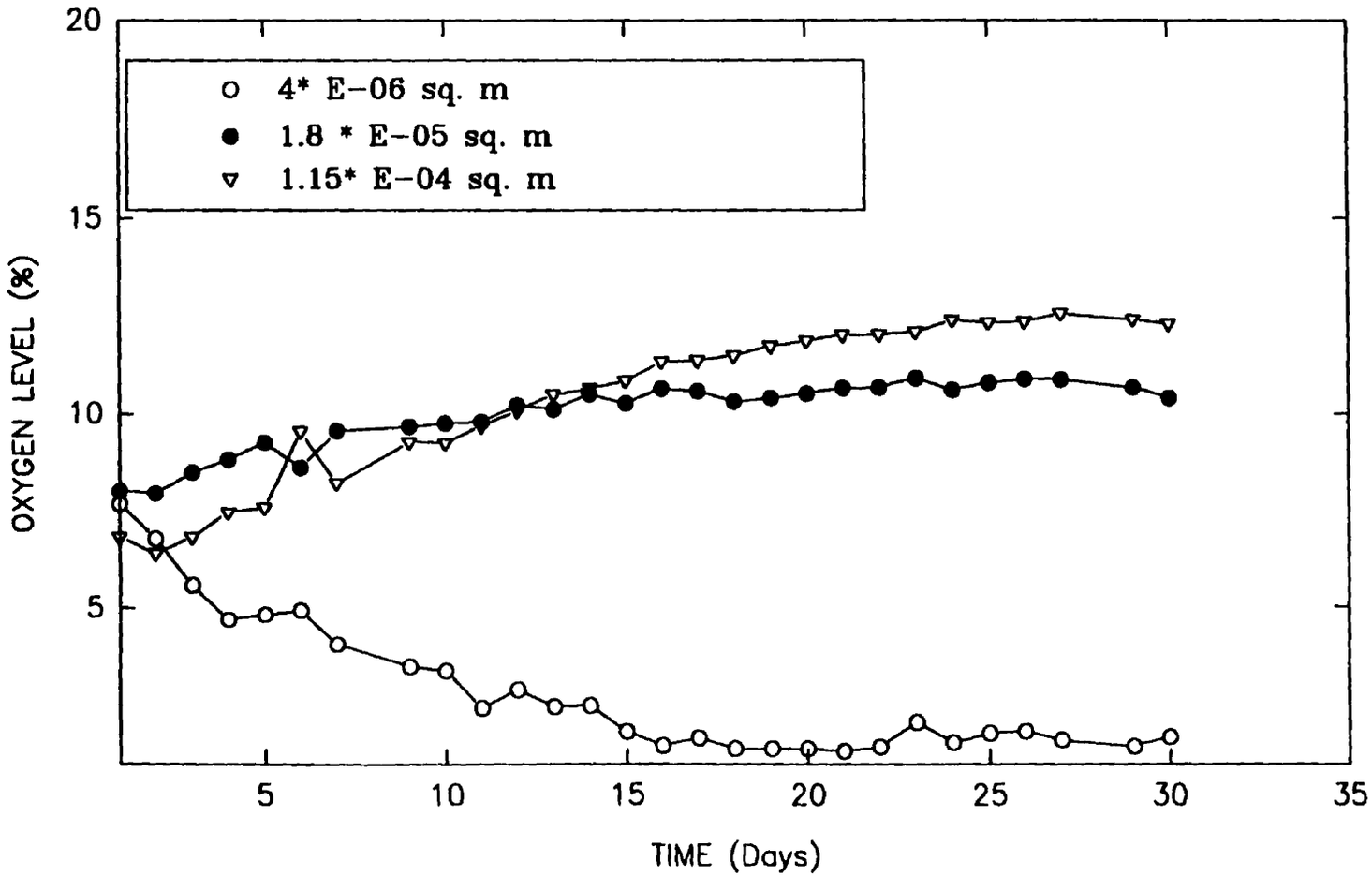

Figure 5.18 Comparison of Progression of O_2 for Chambers with Channels of 0.12 m in Length and Three Varying Cross Sectional Areas.

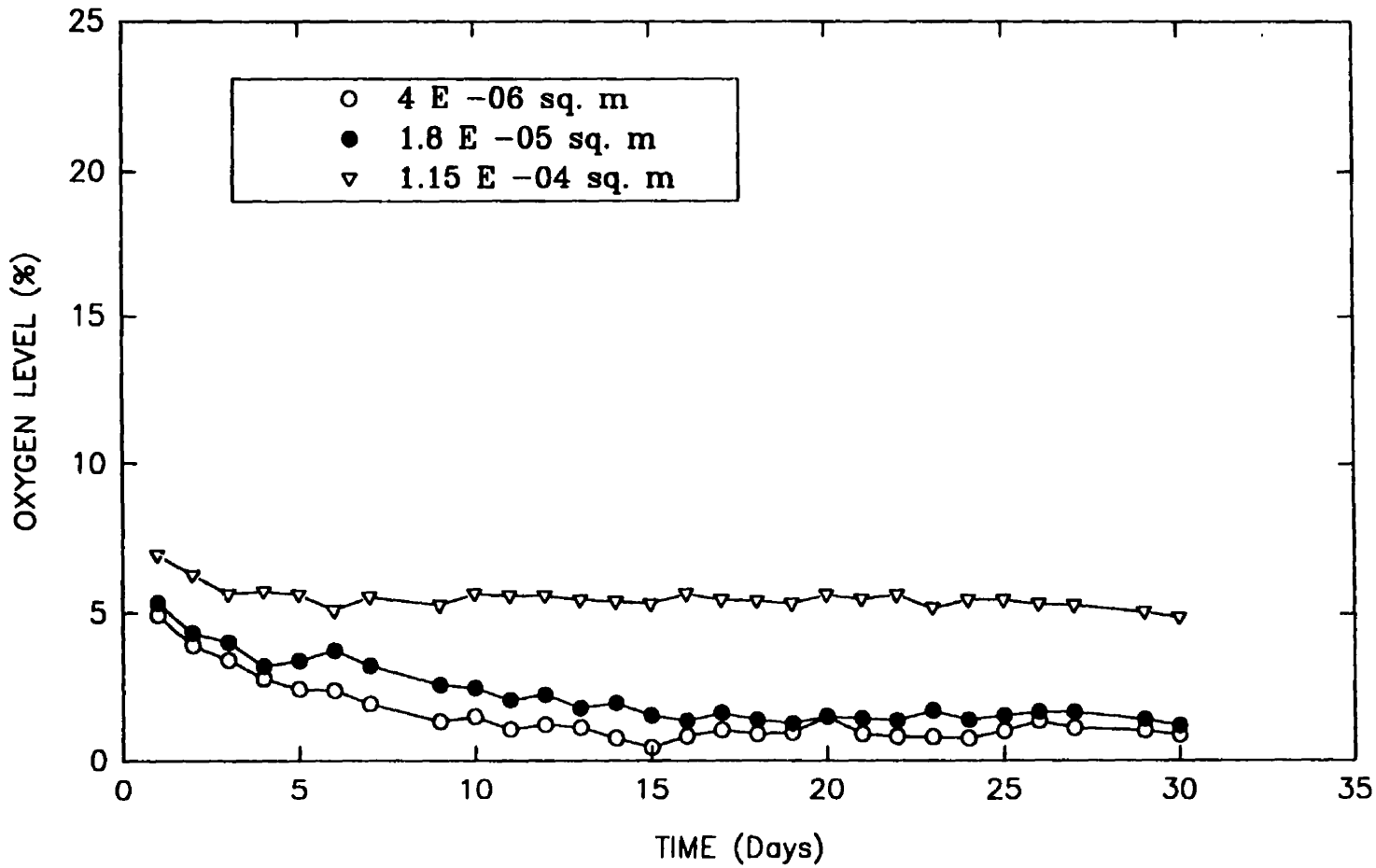

Figure 5.19 Comparison of Progression of O_2 for Chambers with Channels of 0.18 m in Length and Three Varying Cross Sectional Areas.

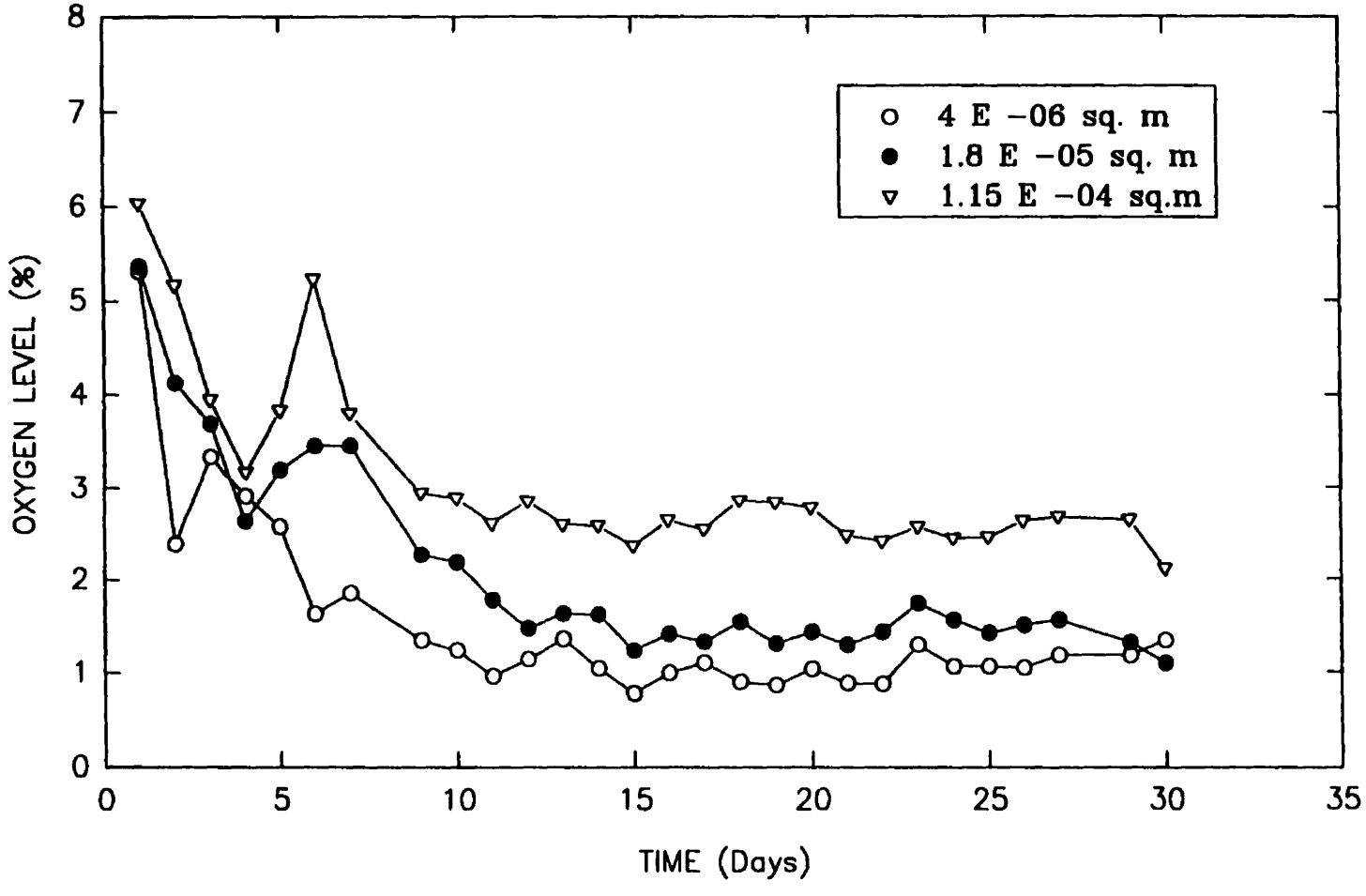

Figure 5.20 Comparison of Progression of O_2 for Chambers with Channels of 0.25 m in Length and three Varying Cross Sectional Areas.

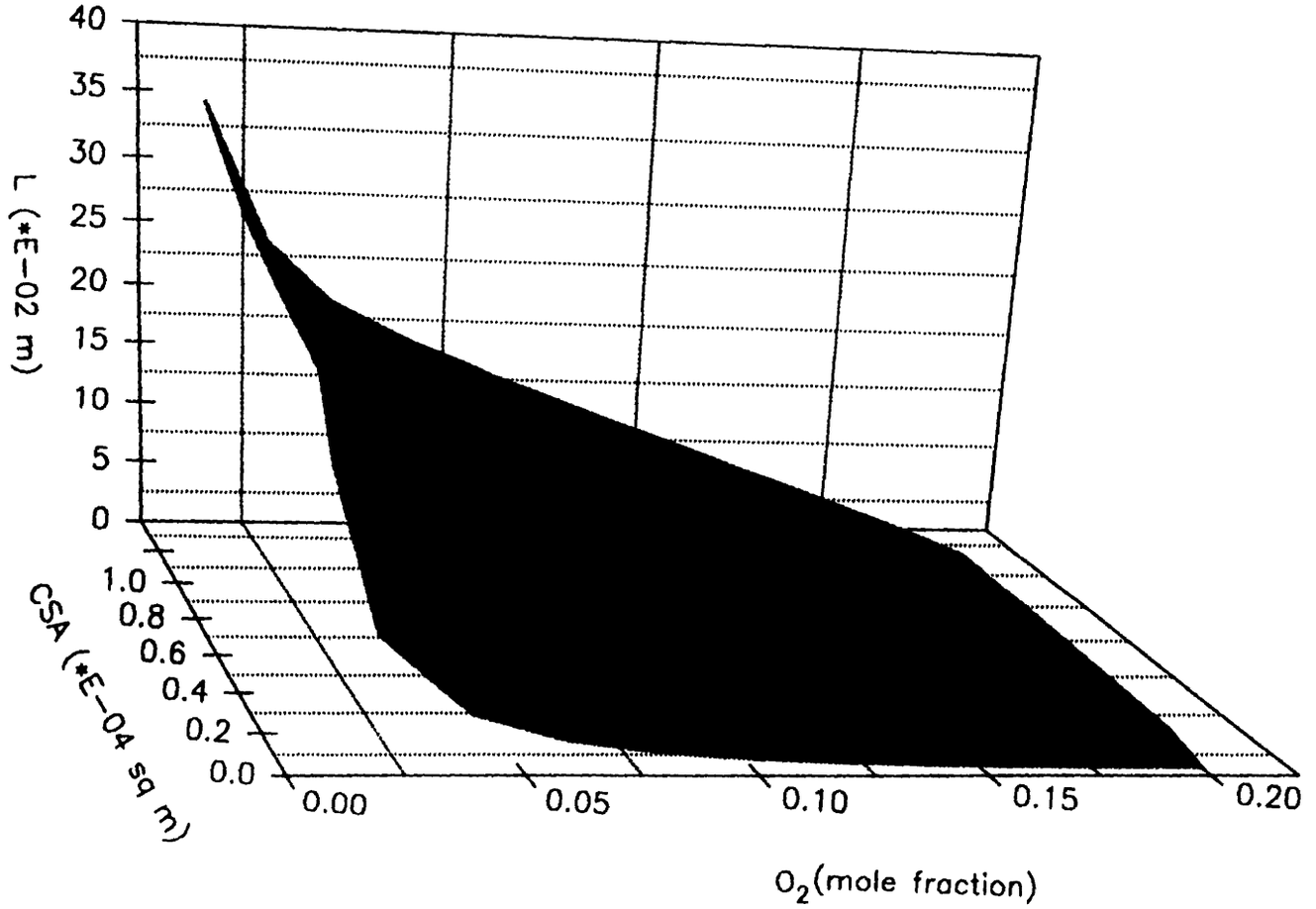

Figure 5.21 The Relationship of Length, Cross Sectional Area of Diffusion Channel and O_2 Steady State Level.

Table 5.3 Time (Days) Taken for O_2 to Reach Steady State Conditions in Chambers Installed with Diffusion Channels

Chamber	L (m)	CSA (m^2)	t (D)*
1	0.006	$4*10^{-6}$	3
2	0.006	$1.8*10^{-5}$	4
3	0.006	$1.15*10^{-4}$	2
4	0.03	$4*10^{-6}$	9
5	0.03	$1.8*10^{-5}$	9
6	0.03	$1.15*10^{-4}$	3
7	0.07	$4*10^{-6}$	11
8	0.07	$1.8*10^{-5}$	9
9	0.07	$1.15*10^{-4}$	17
10	0.12	$4*10^{-6}$	15
11	0.12	$1.8*10^{-5}$	15
12	0.12	$1.15*10^{-4}$	6
13	0.18	$4*10^{-6}$	11
14	0.18	$1.8*10^{-5}$	11
15	0.18	$1.15*10^{-4}$	3
16	0.25	$4*10^{-6}$	9
17	0.25	$1.8*10^{-5}$	9
18	0.25	$1.15*10^{-4}$	4

* t = time, D = days

distinguishable as the cross sectional area of the channels increased. The ratio of O_2 to CO_2 varied in the chambers.

(b) **Carbon dioxide**

Comparison is also made for the carbon dioxide levels in the CO_2 unscrubbed chambers installed with channels of three varying lengths and three cross sectional areas. Figures 5.28, 5.29 and 5.30 are plots for carbon dioxide levels of the three cross sectional areas $9*10^{-6}$, $4*10^{-5}$ and $1.7*10^{-4}$ m^2 respectively, as a function of time. Contrary to the O_2 steady state levels, CO_2 steady state levels were generally low for chambers with shorter diffusion channels for all the three cross sectional areas. However, the lowest level was observed in the chambers with diffusion channels of larger cross sectional areas. The difference in the levels of CO_2 were distinguishable as the cross sectional area increased.

5.2.2 Model for Length of Diffusion Channel for Establishing CA Conditions in Storage of Spinach

Non linear regression of the experimental data using Sigma Plot computer software for curve fitting by Jandel (1992) yielded constants a_1, a_2 and a_3 for the model (Equation 3.29). The constants are presented in Table 5.4. The best fit curves of the model for predicting the length of a diffusion channel which can maintain a desired oxygen concentration, are plotted in Figure 5.31 for the three cross sectional areas.

5.3 Quality Evaluation of Spinach After CA Storage

Spinach stored relatively well in CA conditions established and maintained by the diffusion channels. The quality of spinach was good where O_2 levels were in the range of 1 to 5%. After 30 days of CA storage at 2°C, the mass of spinach was on average less by 2% and the trimming losses ranged from 3 to 24%. The results for quality assessment upon opening the chambers are shown in Table 5.5.

The quality of spinach after 49 days of storage in CA was comparatively

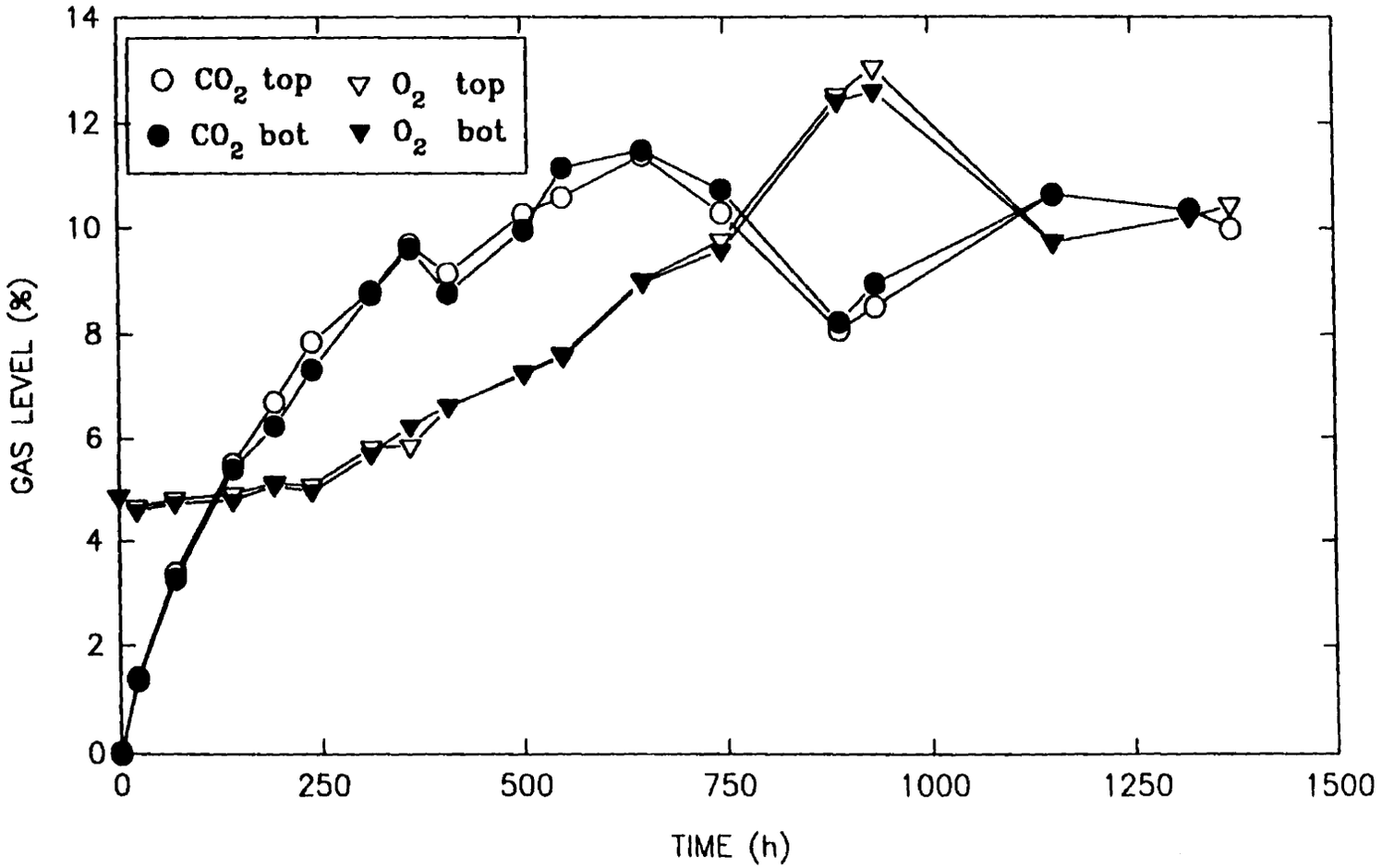

Figure 5.22 Comparison of Gas Sampled from Top and Bottom of the Storage Chambers with Diffusion Channel of 0.043 m and CSA of $9*10^{-6}$ m^2 as a Function of Time.

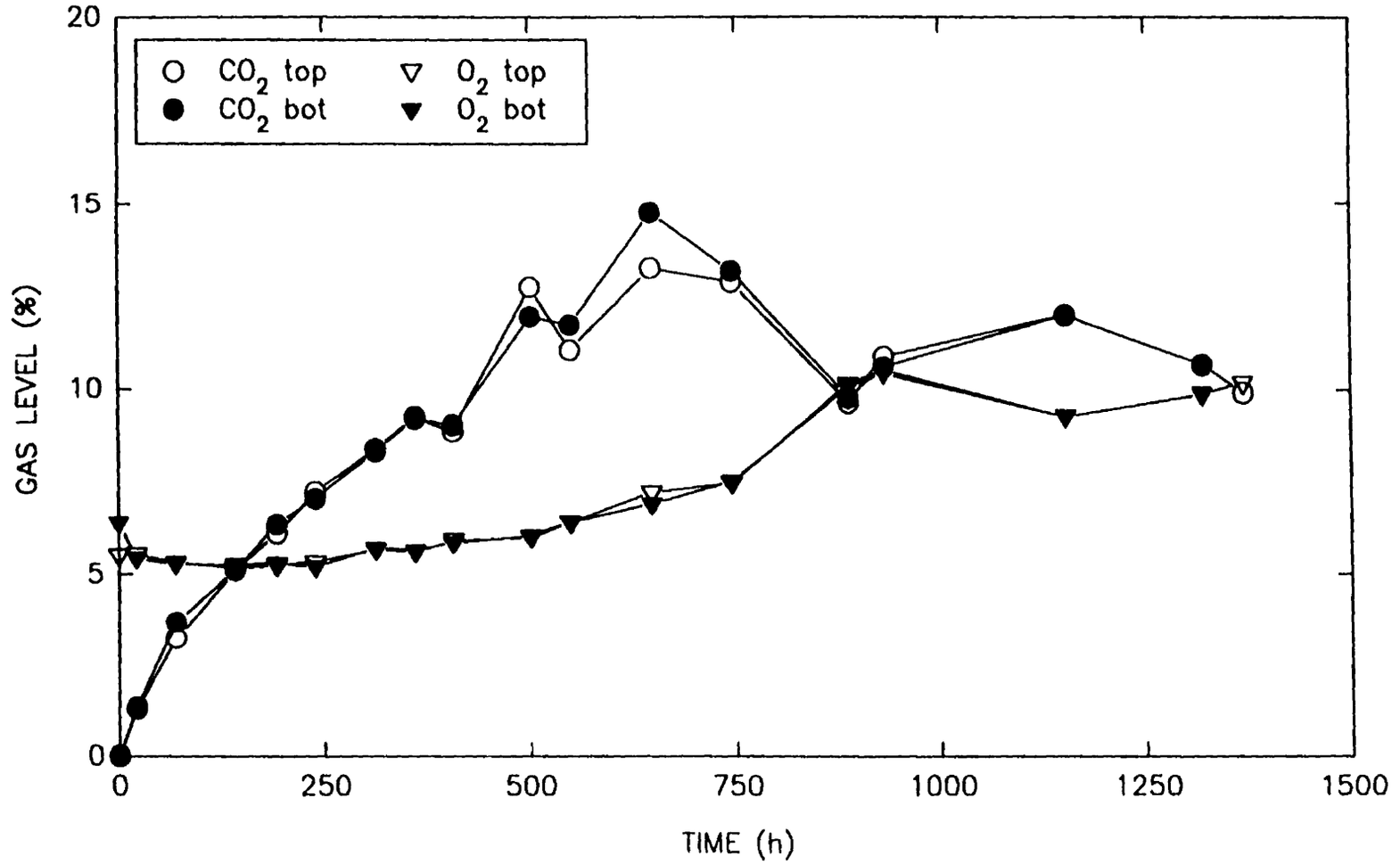

Figure 5.23 Comparison of Gas Sampled from Top and Bottom of the Storage Chambers with Diffusion Channel of 0.11 m and CSA of $9*10^{-6}$ m^2 as a Function of Time.

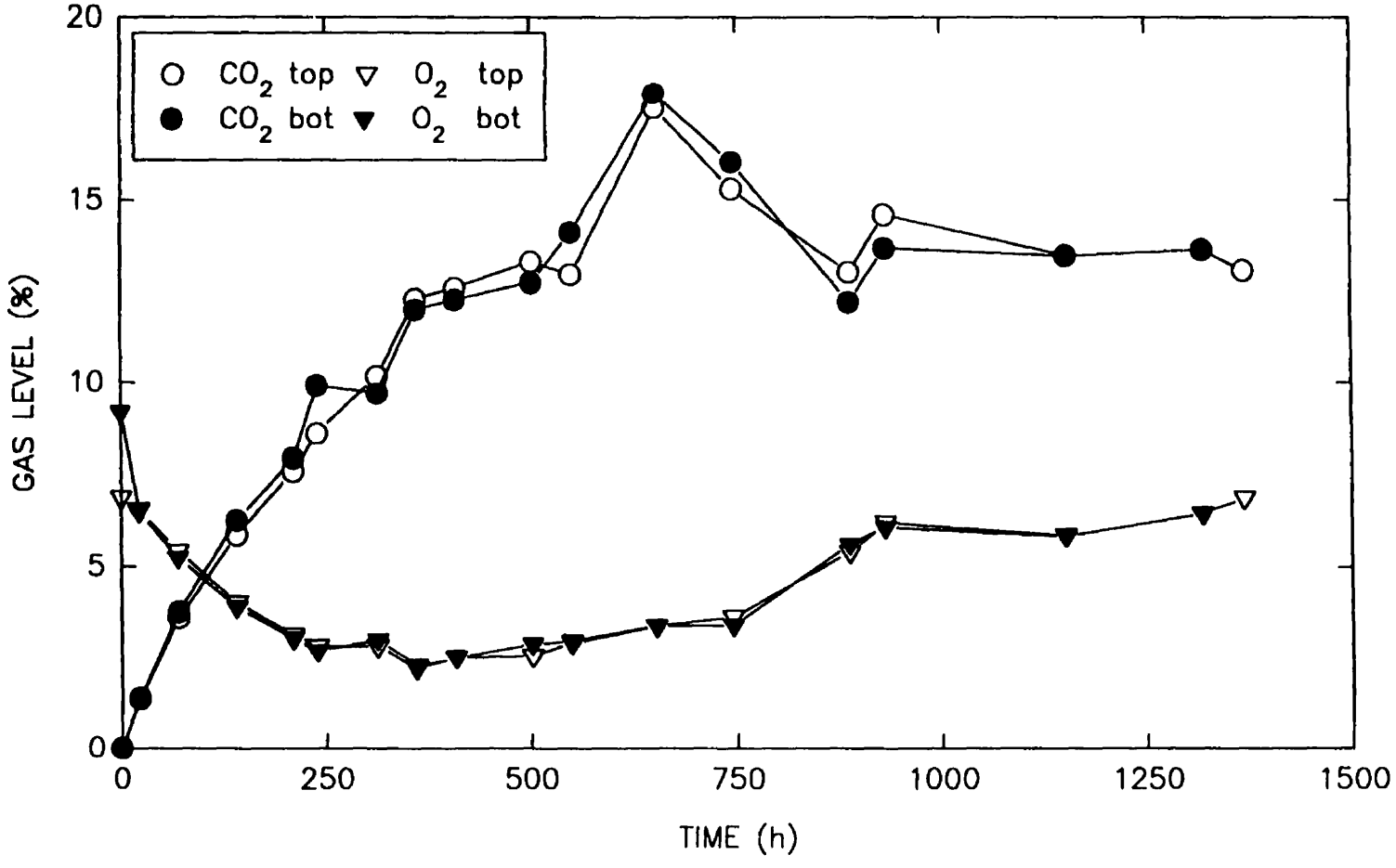

Figure 5.24 Comparison of Gas Sampled from Top and Bottom of the Storage Chambers with Diffusion Channel of 0.19 m and a CSA of $9*10^{-6}$ m^2 as a Function of Time.

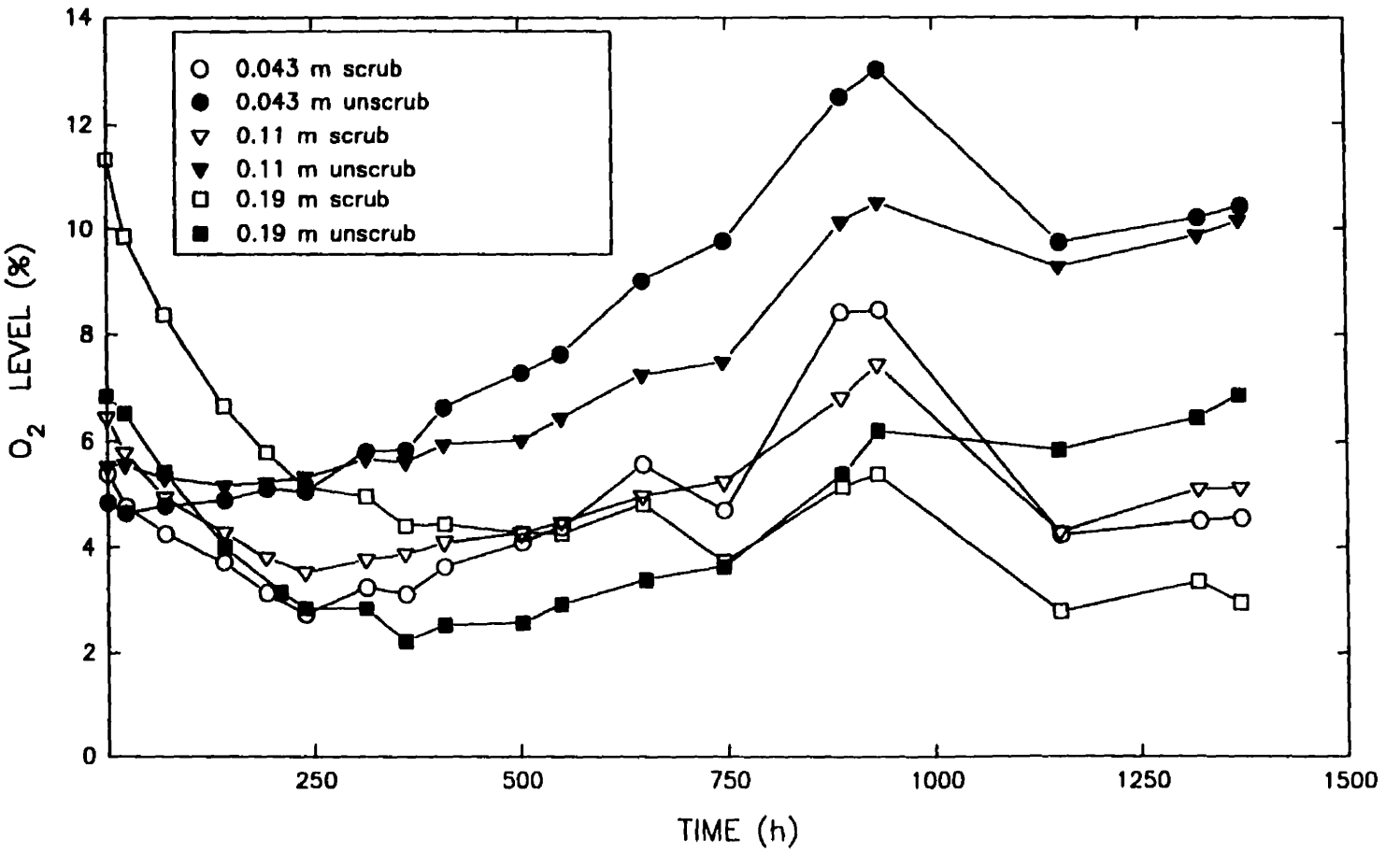

Figure 5.25 Comparison of O_2 Levels in CO_2 Scrubbed and Unscrubbed Chamber for CSA of $9*10^{-6}$ m^2.

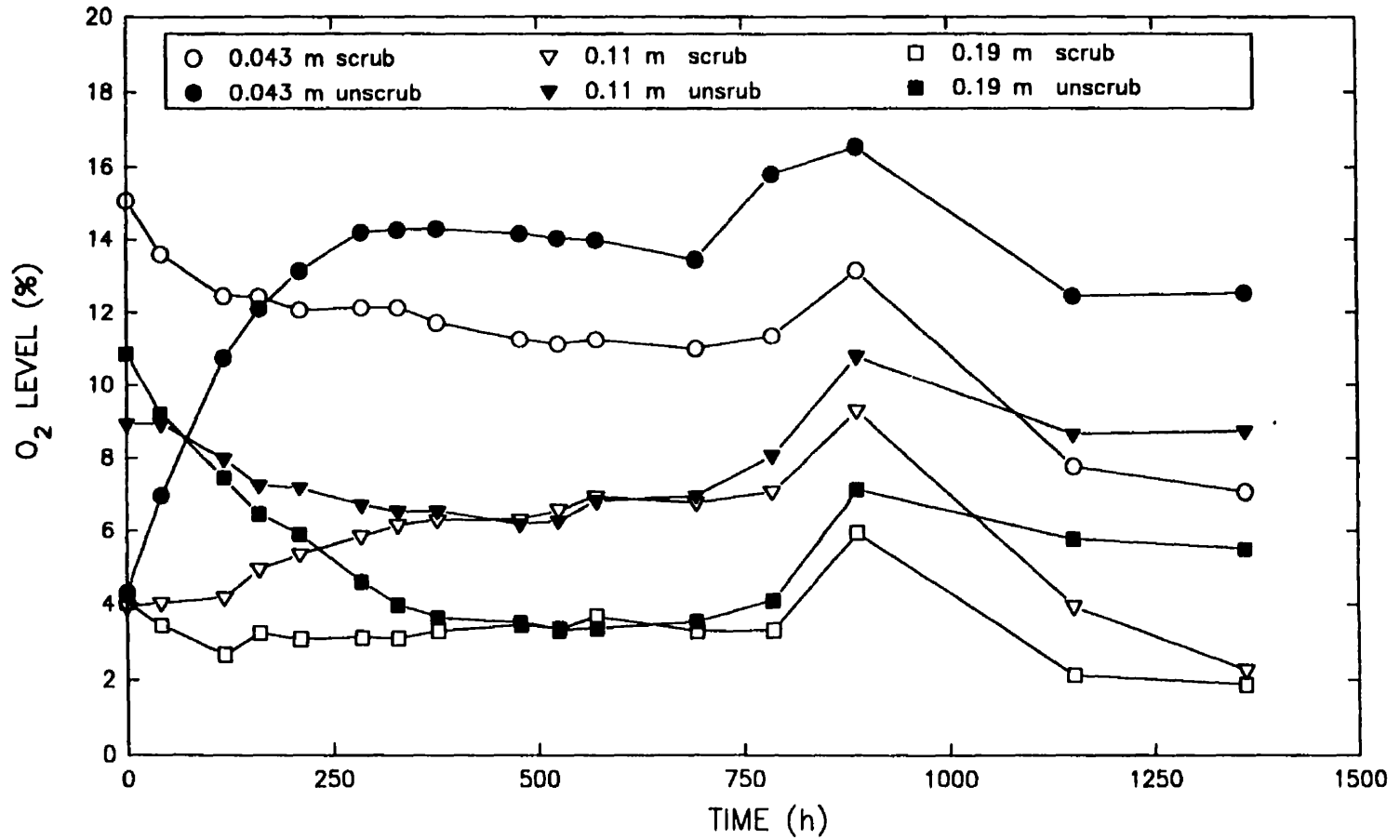

Figure 5.26 Comparison of O_2 Levels in CO_2 Scrubbed and Unscrubbed Chambers for CSA of $4*10^{-5}$ m^2.

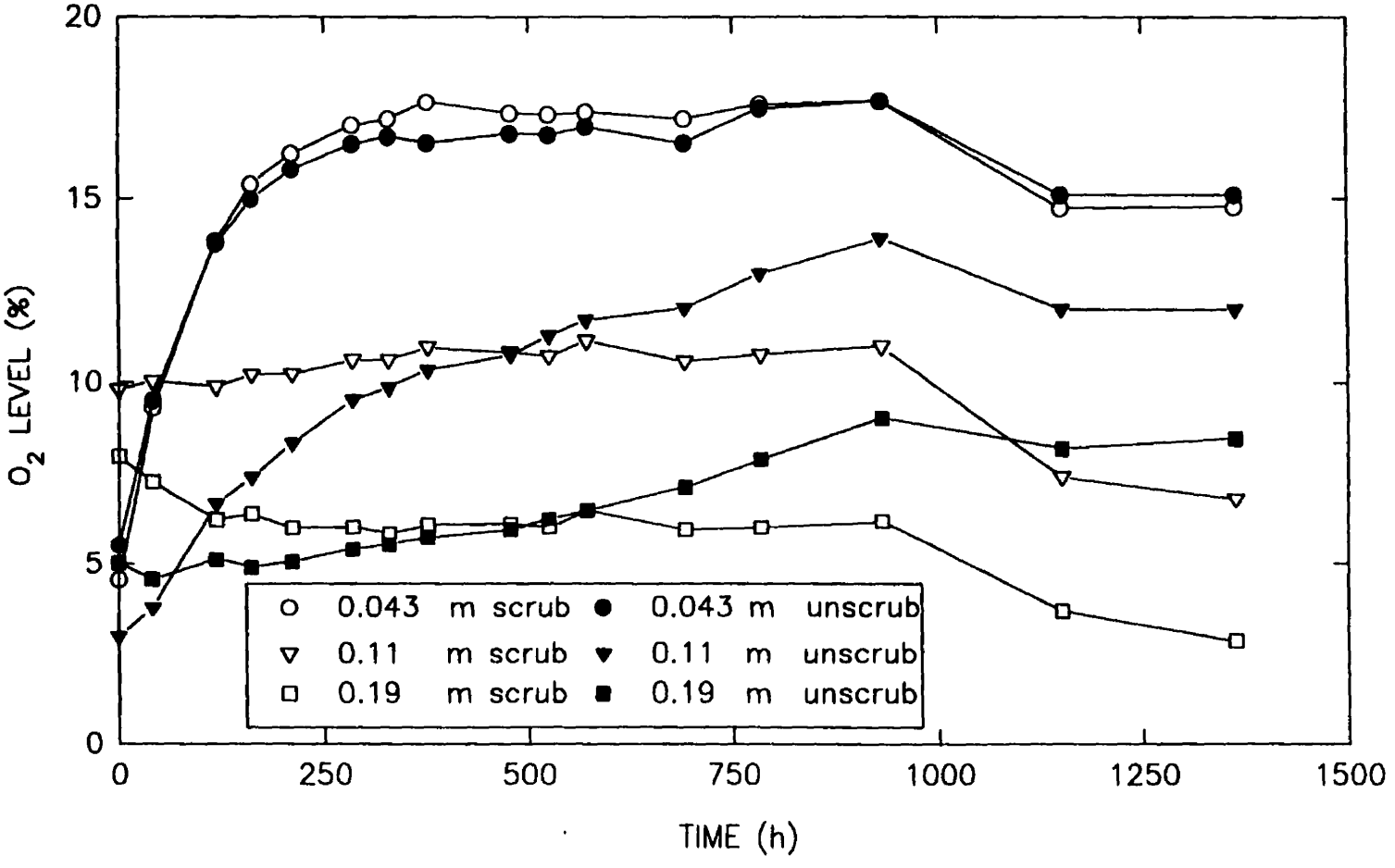

Figure 5.27 Comparison of O_2 Levels in CO_2 Scrubbed and Unscrubbed Chambers for CSA of $1.7*10^{-4}$ m^2.

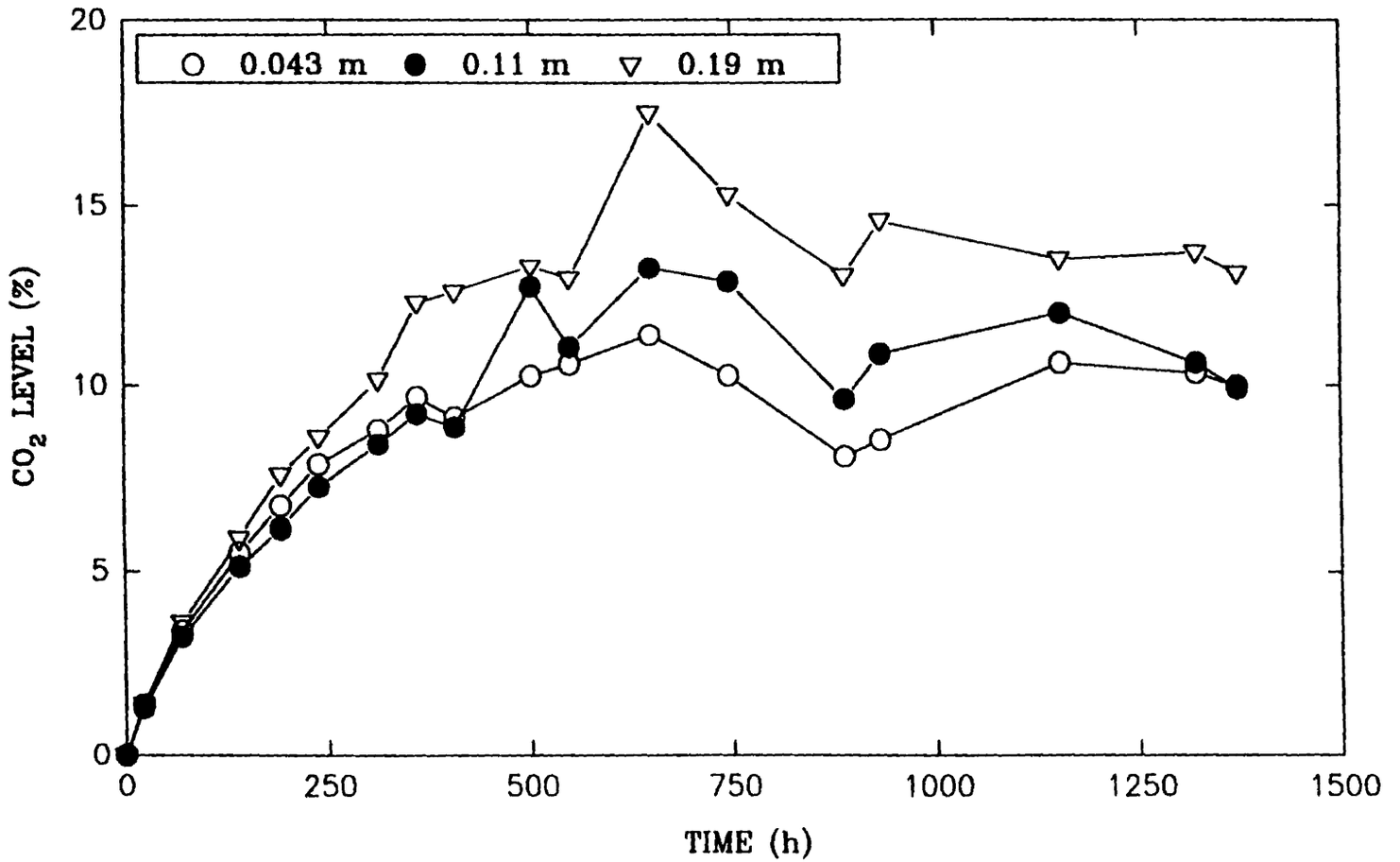

Figure 5.28 Progression of CO_2 in Chambers with Channels of Three Varying Lengths and CSA of $9*10^{-6}$ cm^2.

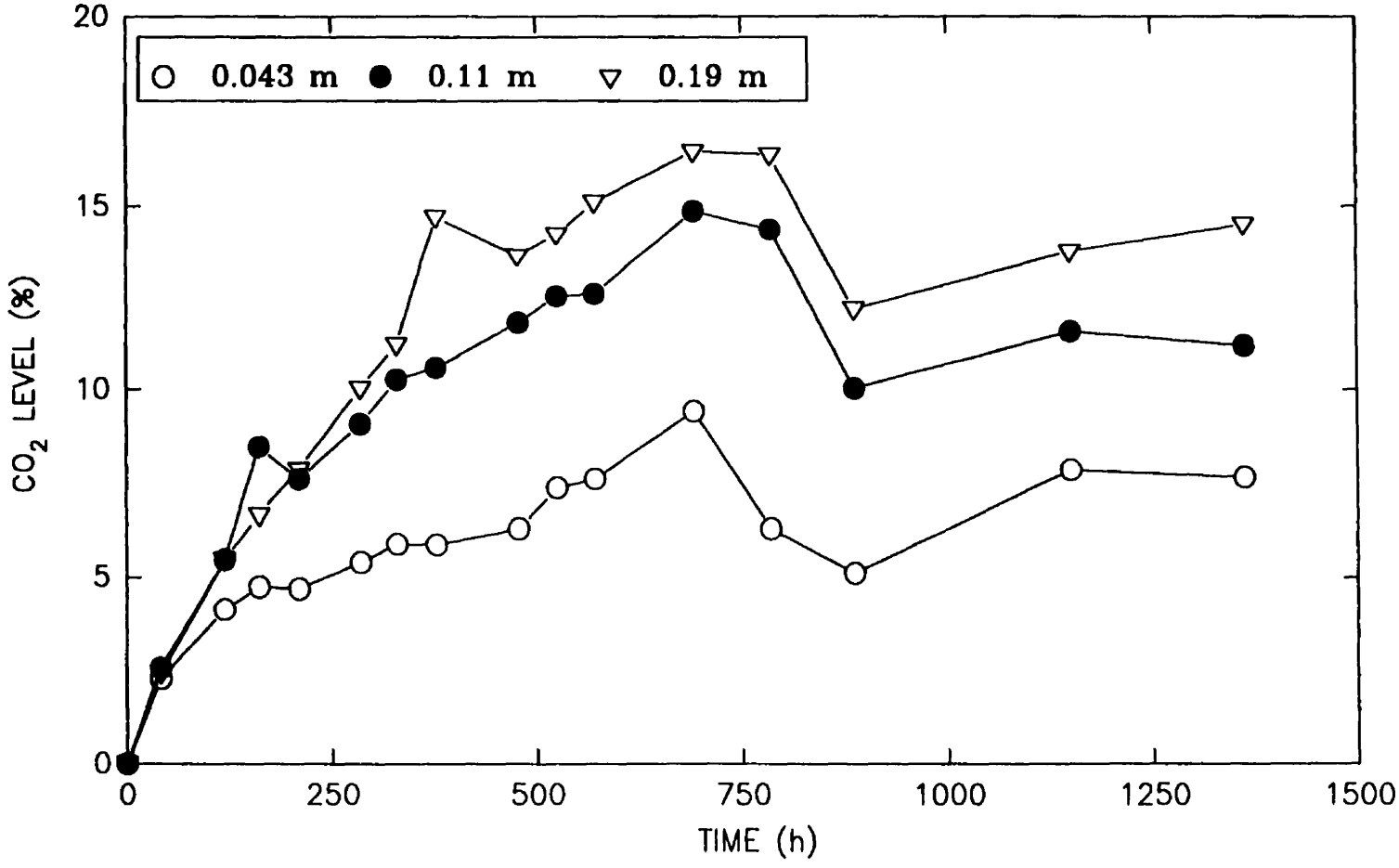

Figure 5.29 Progression of CO_2 in Chambers with Channels of Three Varying Lengths and CSA of $4*10^{-5}$ m^2.

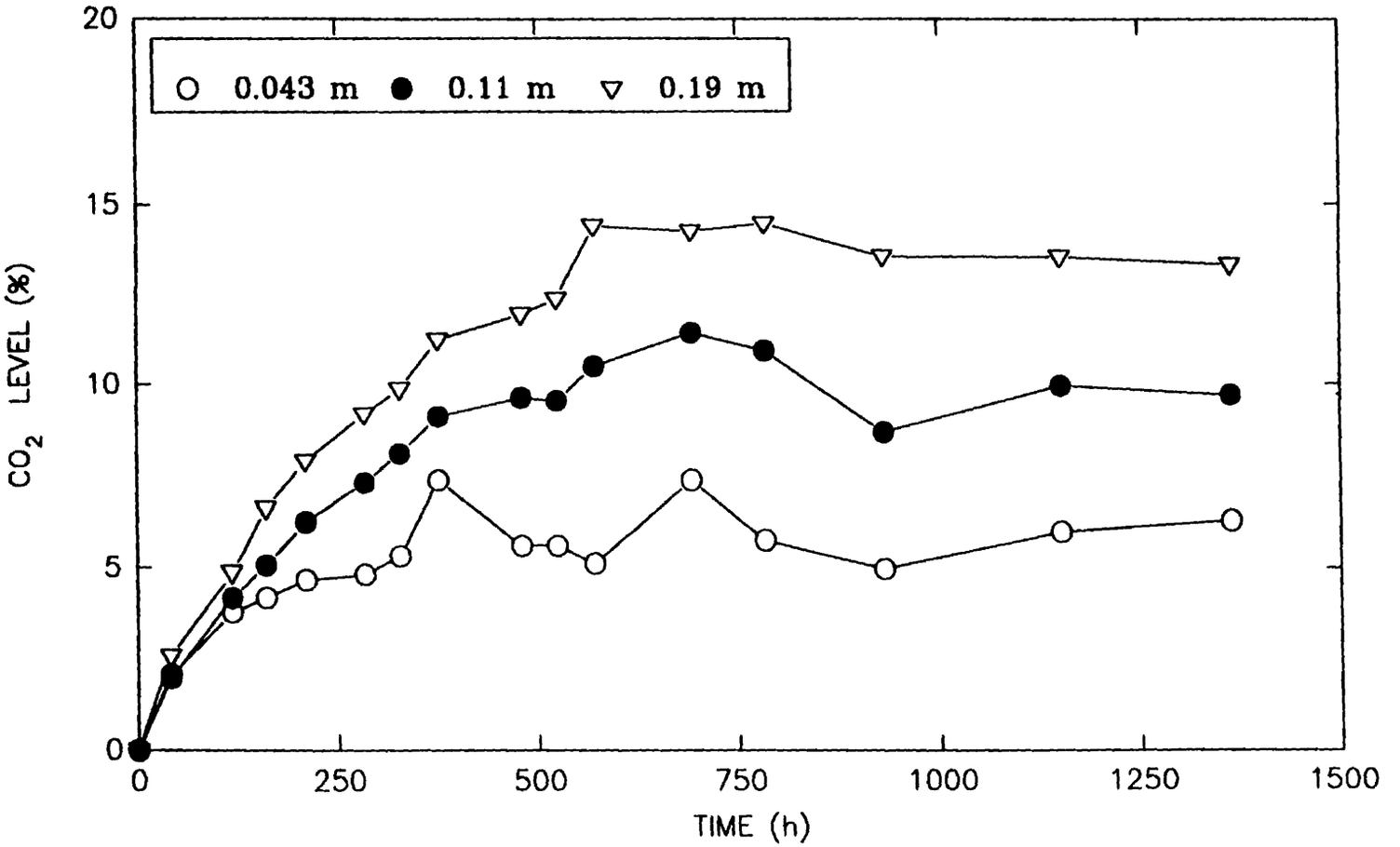

Figure 5.30 Progression of CO_2 in Chambers with Channels of Three Varying Lengths and CSA of $1.7*10^{-4}$ m^2.

Table 5.4 Constants for The Diffusion Channel Model

CSA	a_1	a_2	a_3
$4*10^{-6}$	32.71	-620.50	127.53
$1.8*10^{-5}$	28.21	2019.20	20.76
$1.15*10^{-4}$	14.47	944.96	6.9483
ALL CSA	17.38	131.52	36.76

poorer than in the first set. The physiological mass losses ranged from 3 to 24%, whereas the trimming losses were as high as 80%. The losses were generally higher for spinach stored in CO_2 scrubbed chambers. Overall, appearance was fair in the chambers with CO_2 levels above 10%. Fungal growth was predominant in chambers with O_2 levels above 1.5%. The post-CA storage quality of spinach after 49 days of storage is shown in Table 5.6.

The results from Duncan's Multiple Range Test (DMRT) performed on the means of spinach colour measurements are shown in Tables 5.7 and 5.8 for storage durations of 30 and 49 days, respectively.

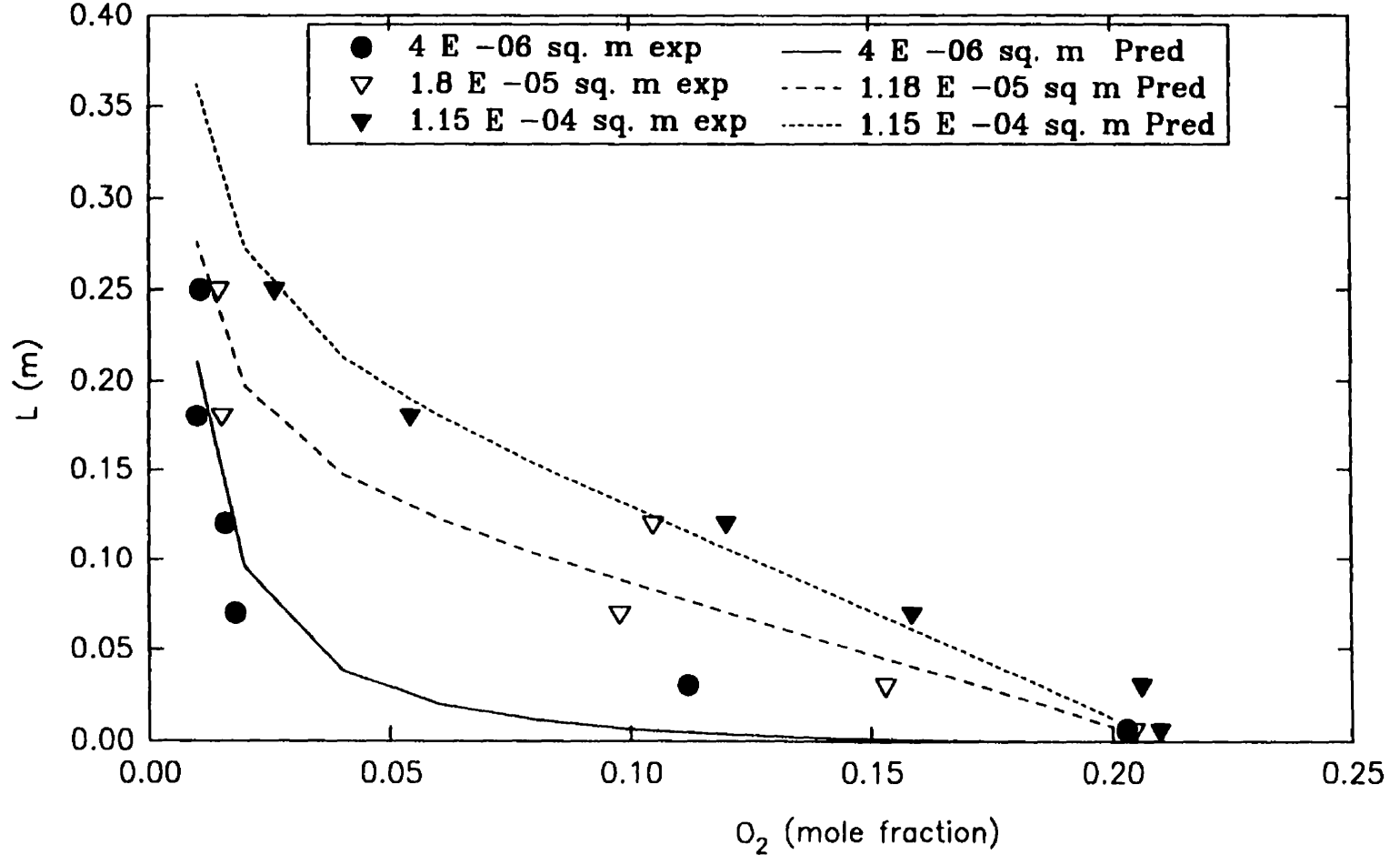

Figure 5.31 Comparison of Predicted Length of Diffusion Channel with Experimental Data for Three Varying Cross Sectional Areas.

Table 5.5 Quality of Spinach After 30 Days of CA Storage at 2°C

Ch #	O_2 (%)	Phy* (%)	Tr** (%)	Appearance of Leaves	Fungal Growth***
1	20.30	2.0	16.0	fresh and green	+
2	20.45	3.0	19.0	fluffy and green	+
3	21.15	2.0	21.0	fluffy and green	+
4	11.21	3.0	21.0	fresh and green	+
5	15.3	2.0	17.0	fresh and green	+
6	20.61	3.0	23.0	fluffy and green	+
7	1.79	2.0	18.0	fresh and green	+
8	9.80	2.0	19.0	fresh and green	+
9	15.85	2.0	24.0	fresh and green	+
10	1.60	2.0	14.0	fresh and green	+
11	10.5	2.0	15.0	fresh and green	+
12	12.01	2.0	13.0	fresh and green	+
13	1.0	2.0	3.0	fresh and green[1]	-
14	1.53	2.0	9.0	fresh and green	-
15	5.43	2.0	19.0	fresh and green	+
16	1.08	2.0	8.0	fresh and green	-
17	1.45	2.0	8.0	fresh and green	-
18	2.61	2.0	21.0	fresh and green	+

* Physiological mass loss; ** = Trimming mass Loss

*** + = Present and - = Absent; 1 = Excellent

Table 5.6 Quality of Spinach After 49 Days of CA Storage at 2°C

Ch #	O_2 (%)	CO_2 (%)	Phy* (%)	Tr** (%)	Appearance of Leaves	Fungal Growth***
1	3.0	0	4	60	fresh and green	-
2	4.5	10.5	3.0	31	fresh and green	-
3	5.0	0	5.0	60	turgid and yellow	-
4	6.0	12.5	4.0	34	fresh and green	-
5	4.5	0	5.0	66	fresh and green	-
6	3.0	13.0	3.0	16	fresh and green[1]	-
7	11.0	0	4.0	52	fluffy and brown	-
8	14.0	5.5	3.0	35	fluffy and green	+
9	6.5	0	5.0	62	fluffy and brown	-
10	7.0	12.0	3.0	20	fluffy and green	-
11	3.0	0	5.0	62	fluffy and brown	-
12	3.5	16.0	5.0	67	fluffy and green	-
13	17.0	0	14.0	64	fluffy and yellow	+
14	16.0	4.5	3.0	54	fresh and green	+
15	10.5	0	5.0	80	fluffy and yellow	-
16	11.5	11	3	41	fresh and green	-
17	6	0	4	56	fluffy and yellow	+
18	6	14	5	16	fluffy and green	-

* = Physiological mass loss; ** = Trimming mass loss

*** + = Present and - = Absent; 1 = Excellent

Table 5.7 Colour (L, a and b) Values* of Spinach After 30 Days of CA Storage

Ch #	L (Value)	a (Hue)	b (Chroma)
1	39.10^{bc}	-13.73^{abc}	20.63^{abcd}
2	40.33^{abc}	-13.97^{ab}	20.90^{ab}
3	38.57^{c}	-11.87^{de}	17.23^{ef}
4	40.40^{abc}	-11.67^{e}	18.00^{cdef}
5	41.57^{abc}	-12.27^{bcde}	18.60^{bcdef}
6	41.47^{abc}	-13.53^{abcd}	21.20^{ab}
7	39.77^{bc}	-13.60^{abc}	18.80^{bcdef}
8	39.47^{bc}	-12.27^{bcde}	19.07^{bcdef}
9	35.50^{d}	-11.17^{e}	16.80^{f}
10	42.37^{ab}	-12.10^{cde}	18.83^{bcdef}
11	42.53^{ab}	-13.50^{abcd}	20.83^{abc}
12	43.27^{a}	-12.37^{bcde}	19.97^{abcde}
13	41.13^{abc}	-12.57^{bcde}	18.57^{bcdef}
14	41.53^{abc}	-12.70^{abcde}	19.40^{bcdef}
15	41.80^{abc}	-14.33^{a}	22.37^{a}
16	39.90^{abc}	-11.40^{e}	17.87^{def}
17	40.73^{abc}	-13.40^{abcd}	20.00^{abcde}
18	39.83^{abc}	-13.47^{abcd}	20.67^{abcd}

* = Mean of three samples

Note: Means with same letter in the same column are not significantly different at $P \leq 0.05$.

Table 5.8. Colour (L, a and b) Values* of Spinach After 49 Days of CA Storage

Ch #	L (Value)	a (Hue)	b (Chroma)
1	33.57^{c}	-11.63^{bc}	21.80^{abc}
2	35.73^{abc}	-11.63^{bc}	18.80^{bc}
3	38.83^{abc}	-13.77^{ab}	21.80^{abc}
4	39.83^{a}	-13.97^{ab}	21.83^{abc}
5	34.07^{bc}	-10.40^{c}	17.27^{c}
6	34.27^{bc}	-12.10^{abc}	18.93^{bc}
7	37.17^{abc}	-12.83^{abc}	19.80^{abc}
8	37.47^{abc}	-13.23^{ab}	20.83^{abc}
9	36.70^{abc}	-12.17^{abc}	17.90^{c}
10	38.37^{abc}	-13.07^{ab}	19.10^{bc}
11	38.40^{abc}	-14.10^{ab}	21.63^{abc}
12	34.20^{bc}	-12.20^{abc}	18.63^{bc}
13	39.93^{a}	-13.83^{ab}	24.00^{a}
14	34.13^{bc}	-12.27^{abc}	18.60^{bc}
15	37.70^{abc}	-14.13^{ab}	23.83^{a}
16	33.60^{bc}	-11.63^{bc}	18.53^{c}
17	38.87^{ab}	-14.27^{a}	23.17^{ab}
18	38.83^{abc}	-12.57^{abc}	20.10^{abc}

* = Mean of three Samples

Note: Means with same letters in the same column are not significantly different at $P \leq 0.05$

CHAPTER VI

DISCUSSION

6.1 Effect of Temperature on the Respiration Rate of Spinach

It is evident from the results that the respiration rate of spinach is affected by the changes in temperature. At 23°C, the respiration rate of spinach declined to almost zero after only 40 hours of storage (Figure 5.1). Such a decline corresponds to many typical biochemical and chemical reactions (Cornish-Bowden, 1979 and Burton, 1974). The effect can be attributed to the rapid depletion of the substrate concentration during the respiration process. Spinach, like many leafy vegetables, has limited reserves of substrates (Burton, 1974), as such, the respiration rate is likely to start declining just a few hours after storage. The high temperature hastens the respiration process, with subsequent increase in substrate breakdown. Considering the fact that survival of plant cells depends greatly on the energy generated by the respiration process (Ramaswamy and Raghavan, 1995), a zero respiration rate in storage is not desired because it indicates death of the plant cells. The respiration rate decreased gradually at 2°C (Figure 5.1). Such a behaviour signifies that low temperature is desirable to slow down the respiration process and allow the minimum possible respiration rate that could keep the tissues alive for longer periods. The fast decline in the respiration rate at high temperature could also be attributed to the malfunctioning of the enzymes which catalyses the respiration process. Enzymes are made of protein compounds which are easily denatured if the surrounding temperature is beyond their optimum level (Wills et al., 1981; Burton, 1982 and Cornish-Bowden, 1979).

6.1.2 Respiration Rate at Different Temperature Levels as a Function of O_2 Concentration

If a produce is stored in a gas sealed container, it is expected that the

oxygen level decrease and carbon dioxide level increase because oxygen and carbon dioxide are reactant and product of the respiration process, respectively (Figure 5.2). The decline in the oxygen concentration is what is attributed to the depressive effect on the respiration rate of spinach (Figure 5.3). Such an effect has previously been reported (Platenius, 1943; Burton, 1974; Henig and Gilbert, 1975; Burton, 1982; Beaudry and Gran, 1993) and is extremely important from a postharvest point of view. It is this effect that led early researchers suggest storage of produce where oxygen is limited (Kays, 1991). Limited oxygen could reduce the respiration rate by interfering with the enzymatic activities that occur simultaneously with the respiration process. In the glycolytic pathway, the enzyme which catalyses conversion of fructose-6-phosphate to fructose biphosphate is reported to be inhibited by ATP and citric acid. Both compounds are formed in an oxygen dependent TCA cycle (Burton, 1982 and Kays, 1991).

It is noticeable that the depressive effect of oxygen on the respiration rate ceased with a concentration of below or above 48 mg/l (3.4%) (Figure 5.3). Such an effect is reported to be widely found in many plants (Kays, 1991). The point at which the respiration rate starts to increase is what most researchers call Extinction Point (EP) or Critical concentration and it signifies the termination of the aerobic respiration via the TCA cycle and the commencement of anaerobic respiration (Burton, 1982; Kays, 1991 and Ramaswamy and Raghavan, 1995). The increase in the rate of the respiration at this point (EP) is what is referred to as the Pasteur effect (Isenberg, 1979). Characteristics of it are off flavours and odours.

The critical O_2 concentration for spinach at 2°C, of 48 mg/litre, is within the O_2 critical concentration range for many commodities (Kays, 1991). At high temperatures the EP was not clearly defined (Figure 5.5). The reason could be that the tissues respired so fast and that may have resulted in occurrence of some other biochemical reactions. It is also possible that the time interval for gas sampling

and analysis at such temperatures was not close enough to pick up such an effect. The problem could also be attributed to mere mathematical errors. The EP is however expected to vary with temperature (Kays, 1991). Its value could be high at high temperatures. Some researchers have compiled data on the effect of oxygen and temperature on the respiration rate of various products (Table A-2) cited by Kays (1991).

Low O_2 concentration is desirable to slow down metabolic activities in the produce, however, care has to be taken in ensuring that the concentration is not below the (EP). Knowledge of the critical O_2 concentration of a produce at specific temperatures is therefore very important in postharvest technology. It influences the designing of ventilation or gas diffusion systems in CA storage rooms. It determines the desired levels of O_2 and temperature to be maintained in the storage room. It can also be a deciding factor for the type of storage system to be used for the particular commodity. For instance, in a mixed load of commodities with different critical O_2 levels, at the same temperature, it is appropriate to choose systems such as the hypobaric system which is reported to keep commodities below their EP in a good condition for a longer period (Burg and Kosson, 1983 and Ramaswamy and Raghavan, 1995). Such systems allow rapid diffusion of the O_2 into the tissues with subsequent reduction in the concentration gradient between the interior and the exterior atmospheres. In a diffusion channel system, the pressure is atmospheric (1atm), as such the diffusion of gases into the plant tissue is likely to be slow. The diffusion channel system might therefore, not be suitable for storage of mixed loads. When storing a particular commodity, the dimensions of the diffusion channel should allow and maintain the oxygen level in the storage room of close to its EP and NEVER below.

The extinction point can be determined by RQ. Beaudry and Gran (1993)

indicated that the point at which the RQ rises drastically may indicate that the critical O_2 concentration has been reached. However, when using RQ as an EP determinant, it should be borne in mind that changes in RQ are not always due to the changes in the oxygen concentration. Sometimes, the type of substrate being metabolized and the degree of substrate metabolism have a role to play (Bidwell, 1979 and Wills et al., 1981). It is therefore, important that the RQ be interpreted with caution. In this study, the RQ value varied with time (Figure 5.7) as well as with changes in O_2 concentration (Figure 5.8). The drastic increase in the RQ after 150 hours of storage could either be associated with oxidation of organic acids or an indication of O_2 being retained or failing to diffuse into the tissues, a consequence of which is fermentation (Wills et al., 1981; Bidwell, 1979 and Smock, 1979).

6.1.3 Respiration Rate at Different Temperature Levels as a Function of CO_2 Concentration

The decrease in the respiration rate of spinach with an increase in carbon dioxide concentration (Figure 5.4) could be related to the effect of accumulation of products that occur in many reversible or partly reversible biochemical and chemical reactions. The high CO_2 may have impeded the forward reaction which breaks down sugar (Equation 2.1). Some researchers have also attributed the effect to the increase in the concentration of CO_2 in the cell sap, which normally changes the balance of reactants and products of other subsidiary metabolic pathways prevailing in the plant cells (Burton, 1982). However, such a condition may just result in a temporary decrease in the CO_2 output with subsequent temporary decrease in the respiration rate (Burton, 1982). Sometimes, there could be a balance between the subsidiary reactions and the respiration rate, such that CO_2 output remains constant despite the increase in the concentration of the intermediates. If that happens, unnoticeable changes in the respiration rate are

expected. It has been reported by other researchers that the high carbon dioxide concentration does not necessarily depress respiration rate in all tissues and sometimes where such a concentration is expected to inhibit the process, may actually enhance it. The precise mechanism for the inhibitory effect of CO_2 has not been adequately explored (Kays, 1991). Lee et al. (1991) indicated that the depressive effect of CO_2 on the respiration rate sometimes depends on the tolerance limit of the commodity. At 2°C, the CO_2 depressive effect on the respiration rate ceased with a concentration above 280 mg/litre (Figure 5.4). Beyond that concentration, there was a drastic increase in the RQ (Figure 5.7). The results are in agreement with Burton (1982). The increase in the respiration rate beyond certain levels of CO_2 concentration is partly related to the carboxylation reactions occurring in the TCA cycle (Kays, 1991).

High temperature increased the impact of high carbon dioxide on the respiration rate of spinach. Figure 5.7 shows that at 15°C the rise in the respiration rate commenced after only 50 hours whereas at 2°C it was 100 hours later. It is however, apparent in Figure 5.6 that the concentration of CO_2 at the points of rising respiration rate coincides at a CO_2 concentration of 280 mg/l. This emphasizes the importance of keeping the temperature low enough so as to reduce CO_2 injury. The CO_2 level also has to be kept to optimum levels so as to reduce the impact of high storage temperature on the shelf life of the produce. The results are in agreement with most researchers (Platenius, 1943; Bidwell, 1979;, Wills et al., 1981; Kays, 1991 and Beaudry and Gran, 1993; Ratti et al., 1993 and Ramachandra, 1995).

The results discussed in this section are a guide in establishing proper CA storage for spinach. They can also assist in determining the extent to which environmental factors such as temperature, O_2 and CO_2 can be controlled in CA storage rooms. A model for predicting the respiration rate of spinach is therefore

necessary.

6.2 Modelling Respiration Rate of Spinach

The model for the respiration rate of spinach was in fair agreement with the experimental data. The regression coefficient (R^2) was 0.99. The fitting was fair even when respiration rate was expressed as the rate of O_2 consumption. However, the regression constants differed with the basis on which the respiration rate was expressed as shown in Table 5.1. The inhibition constant K_1, calculated as the rate of CO_2 evolution, is inversely related to temperature, whereas, the one calculated as the rate of O_2 consumption increases with an increase in temperature. Such results are expected because the rate of carbon dioxide evolution during the respiration process is not the same as the rate of oxygen consumption. The differences observed in the regression constants suggest that, when using the model, it is important to define the basis on which the respiration rate is being expressed. The value of K_3 was negligible in either case which nullifies the effect of changing carbon dioxide concentration on the respiration rate. Ratti et al. (1993) and Ramachandra (1995) reported similar results. For practical purposes, respiration rate of spinach can reliably be predicted by Equation 5.1. Such a model could be applicable for predicting respiration rate of other commodities of similar respiratory behaviour.

It is worthy to note that vegetables are biological materials and, depending on the prevailing conditions, their response to environmental factors might not be predictable.

6.3 Diffusion Channel for Establishing CA Conditions

6.3.1 Progression of The Gases in The Storage Chambers

Varying length and cross sectional area of the diffusion channels, successfully created and maintained different stable atmospheric conditions. The results (Figures 5.12 through 5.14) suggest that the diffusion channel system can

be used for CA storage of different commodities of different gas concentration requirements. The findings concur with Ratti et al. (1994) and Ramachandra (1995). However, it has been observed in Figures 5.15 through 5.20 that if the length of the diffusion channel is shorter than 0.03 m or longer than 0.07 m, changing its cross sectional area may result in gas variations that are not appreciable. In Figures 5.18 and 5.19, oxygen levels maintained in the chambers with diffusion channels of lengths 0.12 m, 0.18 m and 0.25 m respectively, were not quite distinct for cross sectional areas of $4*10^{-6}$ and $1.8*10^{-5}$ m^2. The findings give an indication that, there should be an optimum length of the diffusion channel for which the gas level inside the chamber can be a function of the channel cross sectional area. Based on Figures 5.15 through 5.20, the optimum length could lie between 0.03 and 0.07 m.

The optimum dimensions of a diffusion channel could be influenced by the respiration characteristics of the produce. Depending on the respiration rate, a channel of an optimum length has to allow gas to diffuse into the chamber at the same rate as it is being utilised in the respiration process. The equilibrium established between the two processes is likely to change with changes in the diffusion channel cross sectional area.

The mass flux equation in Chapter III (Equation 3.12) shows that the mass flux of a gas is inversely related to length of the diffusion path, therefore, a short diffusion path could allow, if other factors are held constant, gas to diffuse at a faster rate than the rate of the respiration process. This is the reason why the oxygen concentration was high in the chambers installed with a diffusion channel of 0.006 m in length (Figure 5.15). The shorter diffusion path also contributed to the fast attainment of O_2 steady state conditions (Table 5.3). Since a long diffusion path offers a lot of resistance to the diffusion of gases, it is expected that the chambers installed with longer diffusion channels maintain low levels of oxygen.

Most likely, the long gas diffusion path offsets the effect of changing the cross sectional area of the diffusion channel on the steady state gas (O_2) level to be maintained in the chambers. The steady state gas level in a chamber installed with a long diffusion channel could only be influenced by the difference in the gas concentration between the inside and the outside that exists because of the on going respiration process.

Oxygen level in the chambers with longer diffusion channels (above 0.18 m) attained steadiness relatively in less days than chambers with medium sized channels (0.07 m) (Table 5.3). Such an effect is expected because when the diffusion path is long, the gas molecules move slowly into the chambers resulting in less collision among them.

Since the respiratory characteristics of a produce influence the optimum dimensions of the diffusion channel, it is expected that varying storage temperature could also change the dimensions of the channels. However, Baugerod, (1980) and Ratti et al. (1994) indicated that diffusion channels are capable of absorbing fluctuations in storage temperature. The effect has not been adequately researched to warrant any concrete conclusion. Hence, there is need for further investigation.

Changing the dimensions of the diffusion channel also maintained variable CO_2 levels. This shows that the diffusion channel system is flexible. When both gases (O_2 and CO_2) are desired, unfixed ratios (Table 5.6) can be obtained. Such a characteristic makes the diffusion channel system more versatile than the membrane system. It is apparent from Figures 5.25, 5.26 and 5.27 that O_2 levels generally decreased with an increase in length whereas Figures 5.28, 5.29 and 5.30 show that CO_2 increased with an increase in length of the diffusion channel. This is expected, because a long diffusion channel path offers high resistance to the diffusion of gases, be it inward or outward. As a consequence, such gases as carbon dioxide, which are continuously being released during the respiration

process, tend to accumulate in the chambers despite the concentration gradient that exists between the inside and the outside. It is also noticeable that the increase in the carbon dioxide level with an increase in the length of the diffusion channel was not proportional to the decrease in the oxygen level. However, the ratio of O_2:CO_2 was generally high for chambers with diffusion channels of a large cross sectional area and low for chambers with long diffusion channels (Table 5.6). The reason is obvious. Gases diffuse according to their chemical and physical characteristics (Kays, 1991).

6.3.2 Effect of Position of Sampling Port on the Gas Levels

It is clear from the results (Figures 5.22, 5.23 and 5.24) that there was no distinguishable difference in the gas levels within the storage chambers. Uniformity in the distribution of the gases in the chambers is expected so that such gas samples can be drawn from any point on the chambers. The results have shown that the inclusion of the cylindrical wire meshes in the middle of the storage chambers is not necessary. The space occupied by the meshes can therefore be recovered and their cost be avoided.

The uniformity in the gas distribution inside the chambers indicates that the spinach was well packed and that permitted free air circulation.

6.3.3 Effect of Scrubbing CO_2 on O_2 Concentration

Scrubbing CO_2 in the chambers resulted in higher steady state levels of O_2 (Figures 5.25, 5.26 and 5.27). The variations in the O_2 levels between CO_2 scrubbed and unscrubbed chambers could be because of the outward diffusion of the CO_2 which changed the gas diffusion system from a one dimensional to a two dimensional. It has been noted in the same figures that it took more time for O_2 in CO_2 unscrubbed chambers to reach steady state conditions than in the CO_2 scrubbed chambers. The effect could partly be attributed to the chemical and physical properties of CO_2 which are so dynamic. The RQ values of spinach show

that not the same amount of CO_2 is produced despite the same amount of O_2 consumed. Since diffusion of a gas is affected by its concentration, which is a function of volume (Gebhart, 1993), changes in the volume of the CO_2 diffusing outside is expected to affect the volume of O_2 diffusing inside the chambers, hence the fluctuations. The unsteadiness in the O_2 levels of the unscrubbed chambers could also be because the CO_2 and O_2 molecules move in opposite directions and that might have caused some friction between them, as they bump into each other. In establishing CA conditions by diffusion channel system, it is therefore important to note that scrubbing CO_2 changes the steady state level of O_2 and also the time such conditions can be achieved.

6.3.4 Modelling of The Length of The Diffusion Channel

The model developed (Equation 3.29) for predicting length of diffusion channel in establishing appropriate CA conditions for storage of spinach, fits the experimental data well when individual channel cross sectional areas are considered. The analysis shows that the constants a_1, a_2 and a_3 are different among the three channel cross sectional areas. Non linear regression analysis of the combined data from all the three cross sectional areas, in some instances, underestimates the dimensions of the lengths. The reason could be because oxygen level in the chambers, where the channel length is too short or too long (Figures 5.15 and 5.20), is insensitive to the changes in the channel cross sectional area. The model therefore, suggests that, the desired oxygen level in the storage chambers for any length can be precisely obtained if the cross sectional area of the channel is known. To get the same oxygen level as in a chamber installed with a short diffusion channel of a small cross sectional area, one can use a longer diffusion channel of a larger cross sectional area.

It is also important to note that, in some chambers, the steady state oxygen levels are higher (>21%) than in the air. This problem is associated with the

failure of the gas chromatograph to separate argon from oxygen (Goyette et al., 1994 and Ratti et al., 1994). As a consequence, oxygen levels in some cases is over estimated. Such discrepancies may have had an effect on the predicted values of the length of the diffusion channel. It could be better if there is a way of separating argon from oxygen.

6.3.5 Quality of Spinach after CA Storage

The results in Table 5.5, clearly show that spinach stored well at 2°C for 30 days in CA conditions where oxygen level was in the range of 1-1.5%. The limited oxygen played a role in reducing the respiration rate of spinach hence, delayed the senescence process. The results from the Figure 5.3 showed that, at 2°C, a minimum respiration rate was exhibited when oxygen level was 3.4% (48 mg/litre). Platenius (1943) also found a significant depressive effect on the respiration rate of spinach with O_2 level below 5%. It is noticeable also that oxygen levels in the range of 1-1.5% resulted in less trimming losses. Oxygen levels above 5% but lower than the ambient level (21%), were able to maintain freshness of spinach, however, pathogenic infections could not be retarded. Consequently, there was high trimming losses (Table 5.5). This effect indicates that limited oxygen supply (< 5 %) prevents pathogenic infection. The results in Table 5.5 show that fungal growth was absent in chambers with low levels of oxygen. The findings prove the potential of using CA conditions in retarding microbial infection during postharvest handling of crops. Oxygen concentration of 1% or low has been reported to have an appreciable effect in reducing growth, spore formation and germination of many postharvest fungi such as *Rhizopus stolonifer* (El-Goorani and Sommer, 1981). The chief benefit of low oxygen levels in CA storage of spinach, was mainly realised in retention of the green colour. The results are in conformity with Singh et al. (1972) on lettuce, Gariépy et al. (1984b) on cabbage and Ramachandra (1995) on broccoli.

After the 30 days of CA storage, limpness in the leaves was generally predominant in the chambers with high oxygen levels. The effect is attributed to the accelerated respiration rate which led to degradation of the organic compounds, including those responsible for maintaining tissue integrity.

Most recommended CA storage conditions for spinach quoted in literature mention of the combined effect of high CO_2 and low O_2 concentrations as being more effective in maintaining quality of spinach for 2 weeks (Hardenburg et al., 1990). However, it is apparent from this experiment that presence of low O_2 (1-1.5%) alone is capable of extending the shelf life of spinach with subsequent reduction in microbial infection for a month (30 days).

The results have also proved that diffusion channel system is capable of maintaining desirable oxygen concentration for CA storage of spinach. Excellent results were obtained with a diffusion channel of 0.18 m in length and a cross sectional area of $4*10^{-6}$ m^2. Ratti et al. (1994) found a diffusion channel of 0.25 m to maintain the best conditions for CA storage of Cauliflower.

After 49 days of CA storage, both physiological and trimming mass losses were higher than after 30 days of CA storage. Duration for storage had a greater impact on the post storage quality of the spinach. However, microbial infection also played a greater role. Oxygen levels of 5-20%, which were tolerated for 30 days, resulted in excessive browning after 49 days. It is worth to note that after 49 days of storage spinach was relatively in good condition in CO_2 scrubbed chambers with O_2 levels below 5%. The quality was even superior where there was a combination of low O_2 and high CO_2. The results concur with Platenius (1943). Ulrich (1975) reported also that a combination of low O_2 and high CO_2 is more effective in reducing respiration rate of harvested produce. The most predominant characteristic with the presence of CO_2 was retention of the green colour. Carbon dioxide has a role to play in preventing chlorophyll degradation

(Platenius, 1943 and Burton, 1982). The presence of high CO_2 prevented chlorophyll degradation in green beans (Groeschel et al., 1966). Do and Salunkhe (1975) reported that CO_2 suppressed degradation of chlorophyll in some fruits. McGill et al. (1966) indicated that in CA storage, where both gases were present, delayed degradation of chrollophyll in spinach. It was also observed in this study that there was a complete absence of fungal infection with CO_2 of above 5.5% for any O_2 level. Such results suggest that, although the presence of both O_2 and CO_2 gases has an impact on retarding microbial infection during CA storage, the effect of augmented CO_2 is more pronounced. El-Goorani and Sommer (1981) and Kader (1992b) reported of similar findings. There is therefore, potential to use CA for preventing pathogenic infections in storage and that could definitely minimise use of chemicals.

Statistical analysis using Duncan's Multiple Range Tests (DMRT) on the means for the colour measurements, has shown that "L" was significantly different in chamber "9" of the first set. The reason could be that the fungal infection which change the surface colour of the leaves. Chamber 9, as shown in Table 5.5 had the highest trimming losses. Most of the leaves were extensively damaged. The different gas levels maintained by the diffusion channels, statistically, did not change the hue and chromaticity, "a" and "b" respectively as shown in Tables 5.7 and 5.8, for 30 and 49 days of CA storage, respectively.

The spinach leaves after 49 days of CA storage were more flaccid with O_2 levels above 5%. The effect could be attributed to direct loss of moisture through transpiration. However, when opening the chambers it was found that the inside was nearly saturated. The limpness in the leaves could therefore be associated with the high respiration rate of the spinach due to the presence of high oxygen levels. Consequently, there was continuous breakdown of the structural organic compounds. Excellent quality in spinach was obtained with a gas combination of

5.5% O_2 and 13% CO_2. The quality of spinach was also quite satisfactory where both oxygen and carbon dioxide were almost equal to 11%. Such a 1:1 ratio of CO_2 to O_2 has been reported by Ueda and Murata cited by Hardenburg et al. (1990). Singh et al. (1972) found that such a ratio to be suitable for CA storage of lettuce.

Overall, the results have shown that shelf life of spinach can be extended by more than 100% of the RA, if stored in a controlled atmosphere. The chamber installed with a diffusion channel of 0.19 m in length and a cross sectional area of $9*10^{-6}$ m^2, maintained the best gas combination of O_2 and CO_2 concentration for CA storage of spinach (Table 5.6).

CHAPTER VII

CONCLUSION

The respiration rate of spinach was low at 2°C and resulted in more than a tenfold increase at ambient temperature (23°C). It is therefore beneficial to store spinach at low temperature for it to have a longer shelf life. Since spinach is in most cases harvested from fields far away from storage, precooling soon after harvest is essential to reduce the impact of the ambient temperature.

High temperature facilitated the depletion and accumulation of oxygen and carbon dioxide concentrations, respectively, in the chambers, which further led to a rapid decline of the respiration rate. Storing spinach in limited oxygen and/ or elevated carbon dioxide atmospheres, soon after harvest, could also limit the impact of the high temperature by allowing the spinach to respire at a lower rate.

A model based on enzymatic kinetics has been developed and could reliably predict the respiration rate of spinach at any given storage temperature. The model is useful in determining the oxygen and carbon dioxide consumption and production rates, respectively, in CA storage and it is fundamental in designing of any CA storage system for spinach. The model, however, shows that the changes in CO_2 concentration has little effect on the respiration rate as compared to the changes in O_2 concentration.

Diffusion channels are capable of establishing desirable CA conditions for storage of spinach. Both length and cross sectional area of a channel have an influence on the steady state gas level in the chambers. However, if the cross sectional area is small, increasing the length might have no appreciable effect on steady state gas level. Similarly, if the length is too short, in this case, less than 0.03 m or too long, greater than 0.07 m, no significant difference in the gas levels is obtained by changing the CSA. In general, the longer the diffusion channel the

lower the oxygen level and the higher the carbon dioxide level in the chambers. Variable combinations of the oxygen and carbon dioxide in the storage chambers can also be obtained by simply varying the channel length and cross sectional area. It is therefore possible to use the diffusion channel system for CA storage of commodities of various gas composition requirements.

A model based on Fick's first Law of "Molecular Diffusion" has been developed to precisely predict the dimensions of the diffusion channel which can allow and maintain a desired gas composition for CA storage of spinach. The model matches the respiration rate of the spinach (in terms of oxygen consumption rate) and the dimensions of the diffusion channel.

A diffusion channel of 0.18 m in length with a cross sectional area of $4*10^{-6}$ m^2 maintained desired O_2 level for CA storage of 1.7 kg of spinach. A diffusion channel of 0.19 m with a cross sectional area of $9*10^{-6}$ m^2 also maintained the best combination of oxygen and carbon dioxide levels for CA storage of the same amount of spinach. Overall, the storage life of spinach was extended to more than 100% of the RA.

It is noteworthy that the presence of CO_2 in the CA storage chambers, installed with diffusion channels, has an effect on the final O_2 steady state concentration and it also affects the time taken for O_2 to attain steadiness.

The gas was well distributed in the storage chambers as such there is no requirement for including wire meshes when packing the spinach.

CA conditions established and maintained by the diffusion channels in addition to extending the postharvest life of spinach, retarded storage microbial infection. The diffusion channels are capable of maintaining O_2 as low as 1% and CO_2 as high as 16% both of which are effective in controlling some problematic pathogens during storage. Use of the diffusion channel could therefore minimize use of chemicals such as fungistat.

Based on the results from this study, a diffusion channel system is effective in controlling diffusion of gas in CA storage chambers. As such, it could be a more flexible, less expensive and simpler way of altering gas composition in CA storage. The principle behind this system could lead to a more versatile technology for CA/MA storage of fresh produce.

CHAPTER VIII

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APPENDIX

Table A-1 Maximum Permissible Mass Loss in Some Selected Commodities

Commodity	Maximum Permissible Mass Loss (%)
Apples (*Malus slyvestris*, Mill.) (4 cultivar)	7.5
Asparagus (*Asparagus officinalis*, L.)	8.0
Beans	
Broad (*Vicia faba*, L.)	6.0
Runner (*Phaseolus coccineus*, L.)	5.0
Snap (*Phaseolus vulgaris*, L.) (4 cultivars)	41.0
Beetroot (*Beta vulgaris*, L.)	7.0
Beetroot with tops	5.0
Blackberries (*Rubus spp.*)	6.0
Broccoli (*Brassica oleracea*, L. Italica group)	4.0
Brussels sprouts (*Brassica Oleracea*, L. Gemmifera group)	8.0
Cabbage (*Brassica oleracea*, L. Capitata group) (3 cultivars)	8.0/10.9
Carrots (*Dacus carota*, L.)	8.0
Carrots with leaves	4.0
Cauliflower (*Brassica oleracea*, Botrytis group)	7.0
Celery (*Apium graveolens* var. *dulce*, Pers.)	10.0
Cucumber (*Cucumis sativus*, L.)	5.0
Leek (*Allium ampeloprasum*, L. Porrum group)	7.0
Lettuce (*Lactuca sativa*, L.) (3 cultivars)	3.7
Nectarine (*Prunus persica* var. *nucipersica*, (Suckow) Schneid.)	21.1
Onion (*Allium cepa*, L.)	10.0
Parsinip (*Pastinaca sativa*, L.)	7.0
Peach (*Prunus persica*, (L.) Batsch.)	16.4
Pea (*Pisum sativum*, L.)	5.0
Pear (*Pyrus communis*, L.) (3 cultivars)	5.9
Peper, green (*Capsicum annuum*, L. Grossum group)	7.0/12.2
Persimmons (*Diospyros kaki*, L.f.)	13.3
Potato (*Solanum tuberosum*, L.)	7.0
Raspberries (*Rubus idaeus* var. *strigosus*, (Michx.) Maxim.)	6.0
Rhubarb, forced (*Rheum rhabarbarum*, L.)	5.0
Spinach (*Spinacia oleracea*, L.)	3.0
Squash, summer (*Cucurbita* spp.)	23.9
Sweetcorn (*Zea mays* var. *rugosa*, Bonaf.)	7.0
Tomato (*Lycopersicon esculentum*, Mill)	7.0/6.2
Turnip with leaves (*Brassica rapa*, L. Rapifera group)	5.0
Watercress (*Nasturtium officinale*, R.Br.)	7.0

Source: Kays (1991)

Table A-2 Effect of Oxygen Concentration and Temperature on The Respiration Rate of Some Selected Commodities

	Carbon dioxide Production (mg/kg.h)					
	In Air			In 3% O_2		
Temperature °C	0	10	20	0	10	20
Asparagus	28	63	127	25	45	75
Beans(Broad)	35	87	145	40	55	80
Beans (Runner)	21	36	90	15	25	46
Beetroot storing	4	11	19	6	7	10
Beetroot Buching with leaves	11	22	40	7	14	32
Blackberries, Bedford Giant	22	62	155	15	50	125
Blackcurrants, Baldwin	16	39	130	12	30	74
Brussels sprouts	17	50	90	14	35	70
Cabbage						
Primo	11	30	40	8	15	30
January King	6	26	57	6	18	28
Deccma	3	8	20	2	6	12
Carrots						
Storing	13	19	33	7	11	25
buching with leaves	35	74	121	28	54	85
Calabrese	42	105	240	-	70	120
Cauliflower, April Glory	20	45	126	14	45	60
Celery, white	7	12	33	5	9	22
Cucumber	6	13	15	5	8	10
Gooseberries, Leveller	10	23	58	7	16	26
Leeks, Musselburgh	20	50	110	10	30	57
Lettuce						
Unrivalled	18	26	85	15	20	55
Kordaata	9	17	37	7	12	25
Klock	16	31	80	15	25	45
Onion Bedfordshire Champion	3	7	8	2	4	4
Parsinip, Hollow Crown	7	26	49	6	12	30
Potato						
Main crop (King Edward)	6	4	6	5	3	4
new immature	10	20	40	10	18	30

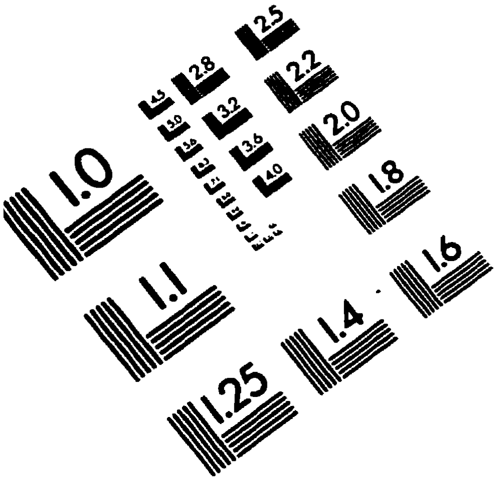
Contd.....

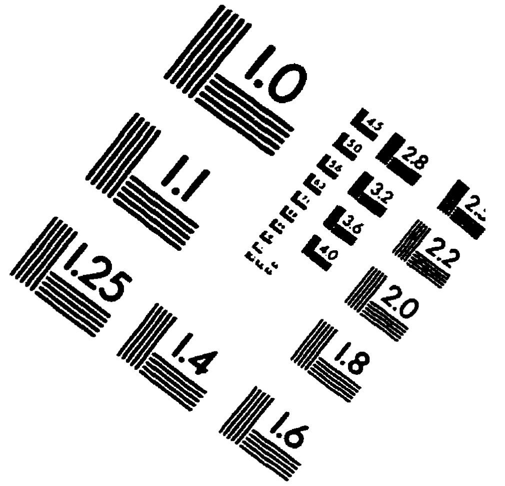
Table A-2 (Contd.) Effect of Oxygen Concentration and Temperature on The Respiration Rate of Some Selected Commodities

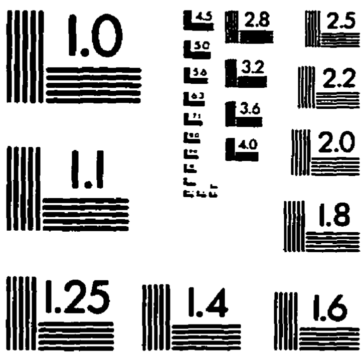
	Carbon dioxide Production (mg/kg.h)					
	In Air			In 3% O_2		
Temperature °C	0	10	20	0	10	20
Peas (in pod) Early (Kelvedon Wonder)	40	130	255	29	84	160
Main Crop (Dark Green Perfection)	47	120	250	45	60	160
Peppers, green	8	20	35	9	14	17
Raspberries, Mailing, and Jewel	24	92	200	22	56	130
Rhubarb (forced)	14	35	54	11	20	42
Spinach (prickly True)	50	80	150	51	87	137
Sprouting Broccoli	77	170	425	65	115	215
Strawberries, Cambridge and Favourite	15	52	127	12	45	86
Sweetcorn	31	90	210	27	60	120
Tomato, Eurocross BB	6	15	30	4	6	12
Turnip, bunching with leaves	15	30	52	10	19	39
Watercress	18	80	207	19	72	168

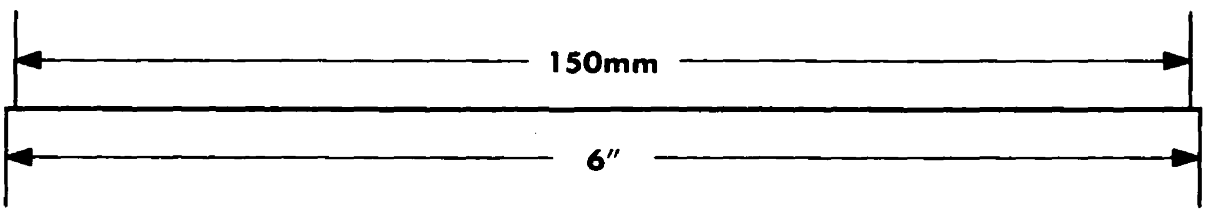
Source: Kays (1991)

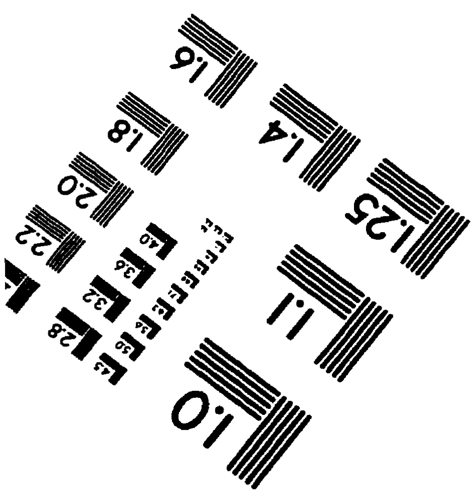
IMAGE EVALUATION TEST TARGET (QA-3)

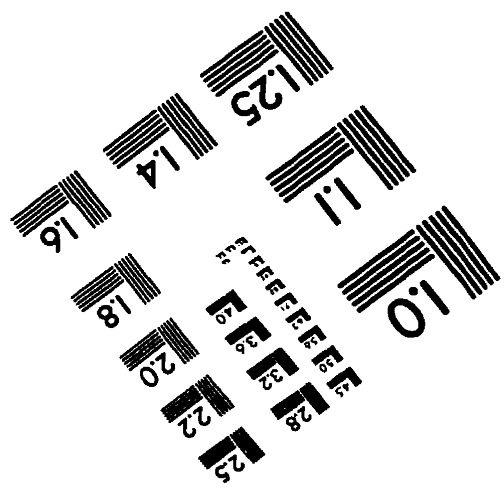












APPLIED IMAGE, Inc
1653 East Main Street
Rochester, NY 14609 USA
Phone: 716/482-0300
Fax: 716/288-5989