A STUDY OF THE NUTRITIVE VALUE OF PASTURE HERBAGE WITH PARTICULAR REFERENCE TO THE EFFECTS OF STAGE OF MATURITY AT TIME OF HARVEST

by

Ivor Hugh Proverbs

A THESIS

presented to the Faculty of Graduate Studies and Research of McGill University in partial fulfilment of the requirements for the degree of Master of Science

May 1950.

ACKNOWLEDGMENT

This study was undertaken in the Nutrition Department, Macdonald College (McGill University) and was made possible by a grant from the Quebec Department of Agriculture, administered through the Macdonald College Pasture Committee.

The author's sincere appreciation is extended to Dr.E.W.Crampton, Professor of Animal Nutrition, for constant guidance in connection with this project.

Acknowledgment is made to Mr.J.P.Woods, Graduate Assistant in Animal Nutrition, in collaboration with whom the entire project was conducted.

To the staff of the Nutrition Department, and to the Graduate Assistants who willingly lent a hand from time to time in the agronomic phases of the work, a very cordial thank-you is extended.

TABLE OF CONTENTS

ABSTRACT

The influence of maturity on the nutritive value of the leaf, stem and head of timothy hay harvested at two stages of maturity has been investigated. Average digestibility coefficients by four Cheviot wethers were used as the criterion for measuring nutritive value.

The decreased nutritive value of the hay with maturity was caused by a serious decrease in availability of the stem nutrients. The leaf and head of the plant largely maintained their high nutritive value with maturity, except in the case of protein whose availability was in all cases very much lowered.

The head of the plant assumed greater importance with maturity as a source of available nutrients, especially protein, tending to offset the serious losses in digestibility incurred by the stem.

INTRODUCTION

Because of the importance of livestock in Eastern Canada Agricultural research has largely been focused on improvement of hay and pasture crops. The Macdonald College Pasture Committee was organized in 1931 in an effort to coordinate agronomic, chemical and nutritional research on this problem.

The ultimate aim of pasture research must be to provide a high and sustained yield of forage that is highly acceptable to the animal and of good nutritive value. Nutritional aspects of pasture research must therefore be directed towards the factors which affect the nutritive value of herbage with a view to bringing them, as far as possible, under the control of the operator. Among the more important factors affecting nutritive value are:

1. Species of plant
2. Stage of maturity
3. Meteorological conditions
4. Soil fertility.

This thesis will deal with the second of these factors - the effect of stage of maturity on nutritive value.

From the point of view of practical considerations it must be noted that the stage of development of the plant at time of harvesting has many and far-reaching effects. These include (a) yield (b) composition (c) digestibility (d) palatability (e) convenience of harvesting (f) permanence of the stand (g) the keeping qualities of the hay (h) the selling qualities of the hay (Waters 1915). Evidently then nutritive value alone cannot be taken as the criterion for deciding the best stage of maturity at which herbage should be harvested. Such a decision must be prompted by an accurate evaluation of all the factors involved, which in turn calls for an individual study of these factors.

At the outset it may be expedient to define nutritive value as the feeding value of a given quantity of material. Thus an increase in the stock-carrying capacity of a field does not necessarily imply an increase in the nutritive value of the herbage (Crampton 1934). In the studies covered by this thesis apparent digestibility as measured with sheep will be used as the criterion for evaluating nutritive value. The justification for this has been covered by Crampton and Jackson (1944) and will be further discussed at a later stage.

Pasture research workers have long sought after some standard or criterion for evaluating herbage material without having to resort to the trouble and expense of conducting feeding trials with the animals in question. Chemical methods have so far been found to be rather unreliable indices. Histological methods have lately been applied to the plant material with some degree of success. Another and far simpler method of evaluation is based on the premise that the leaf of the plant material is of far superior feeding value to the stem, and therefore a knowledge of the relative proportions of leaf and stem in the forage should be an indication of its feeding value. However there is very little experimental evidence on which such a conclusion can be based. Before this method of evaluation can be used with any degree of certainty the relative feeding value of the leaf and stem under various conditions must be established for the herbage under consideration.

In view of the importance of timothy (Phleum pratense) for hay production, grazing and silage production in Eastern Canada it was only natural that this grass should be chosen for the study.

The object of this thesis is not essentially to determine the most suitable stage at which timothy should be cut for hay. The mere fact that the design of the experiment includes only two stages of maturity is an indication of this. It was hoped to go a step further and by separating the hay into leaf, stem and head, and by determining their digestibilities separately, to establish the effect of time of cutting on the proportions and feeding value of these fractions. Finally it was hoped that such data might yield some information on the validity of using the leaf-stem ratio of herbage as an index of its nutritive value.

LITERATURE REVIEW

This review will deal first with the process of lignification insofar as it affects the nutritive value of the plant. This will be followed by a section describing the effect of maturity on the chemical composition and digestibility of hay and pasture crops. In the following section the plants will be broken down into their morphological components - leaf, stem and head, and the work done on the effect of maturity on these separate fractions described. Finally the problem of finding a yard-stick for measuring nutritive value of herbage, and the potentialities of the leaf-stem ratio will be introduced.

Lignification in plant tissue.

The importance of lignin as a limiting factor to the nutritive value of forage has been outlined by Watson and Horton (1936) and Patton and Gieseker (1942). They have shown that lignin limits digestibility because of its encrusting effect on otherwise digestible constituents. It can also combine chemically with the constituents forming unavailable compounds. A third method by which it may lower digestibility is through local inhibition of digestive enzymes due to the toxic action of phenol compounds resulting from partial decomposition of lignin. Crampton and Forshaw (1939) noted that in materials of high lignin content, the digestibility of dry matter, ether extract and cellulose were very much lowered. Even relatively small quantities of lignin had a pronounced effect on digestibility.

The mode of formation of lignin is still unknown, although many speculative theories have been put forward. Phillips et al (1942) have made a very thorough chemical analysis of the timothy plant (Phleum pratense) at successive stages of growth. They found that the per cent cellulose, crude lignin, and methoxyl in the lignin increased with maturity. The absolute quantity of these fractions together with pentosans and pectic substances also increased quite steadily with maturity. They found no evidence to indicate that the plant synthesizes lignin from either cellulose, pentosans or pectic substances. The results were more indicative of lignin being synthesized directly from sucrose or from glucose or fructose as proposed in an earlier study of the oat plant (Phillips et al 1939). Up to about 1932 it was believed that lignification was a senescent change. Woodman, Norman and French (1931) wrote "It is clear that fibre production may go on in growing herbage until the amount is as high as 24% of the dry matter without the chemical character of the fibre undergoing alteration. Throughout this period the digestible form of cellulose only is being elaborated in the herbage plant. At some stage in the subsequent growth of the herbage, marked by the production of but a further 7-8% of fibre, the character of the fibre being deposited changes from cellulose to lignocellulose and the herbage becomes lignified. Lignification therefore is apparently delayed until the final stages of fibre production. This process does not occur in the vegetative stage, but only in the late flowering or even the period of seed formation when the stems and leaves are being depleted of nutrients." However Woodman and Stewart (1932) revised this opinion somewhat after showing that lignin is produced, although in small amounts, in the earliest stages of development of the plant. Kerr and Bailey (1934) showed that lignification is initiated during the earlier stages of secondary wall formation and is first visible in the primary walls and in the intercellular substances. After this it spreads centripetally through the successively formed layers of the secondary wall with advancing maturity of the plant. Its structure can be likened to reinforced concrete in which the iron strands represent the cellulose framework and the concrete the lignin and other constituents. Drapala, Raymond and Crampton (1947) studied plant lignification by histological methods. They found that lignification proceeds regularly with advancing maturity, and the regions around the vascular bundles are primarily involved. They also confirmed an earlier observation of Woodman and Stewart (1932) that it is not only the amount of lignin but its mode of deposition that determines the extent of its effect on digestibility of pasture herbage.

Patton (1943) investigated various Montana grasses to determine the effect of maturity on lignin and cellulose values. The correlation between these two components was 0.9, indicating that the proportion of raw cellulose may serve to indicate the degree of lignification.

Effect of maturity on the chemical composition and digestibility of crops with special reference to timothy.

A considerable amount of work has been reported on the Timothy plant over the years. Kellner (1879), Jordan (1882), and Wilson (1884) reported that the nitrogen and ash of the Timothy plant were higher in the early stages of maturity and decreased in the ripening stages, but that the reverse condition prevailed with nitrogen-free extract. Ladd (1888) reported the analysis of four different stages of growth of Timothy hay. He confirmed the above observations and also noted an increase in sugar and starch after blossoming. Richardson (1883) noted the important fact that immature grasses are high in protein and low in fibre, and that the percentage of protein decreases while that of the fibre increases with maturity.

Morse (1888) analyzed timothy grass at various stages of maturity. The dry matter was found to increase steadily until seed formation. Crude protein decreased steadily with maturity although it remained fairly steady after the bloom began to fade.

Crude fibre increased steadily until seed formation after which it decreased slightly. Nitrogen-free extract remained nearly constant after the grass approached its full height. Ether extract decreased until blossoming, then increased until the seed began to form, when it again decreased, reaching its lowest point as the seed began to harden. Ash was more abundant during the rapid growth of the plant then after growth had ceased. The young grass was richer in nitrogen-free extract and fibre.

Waters (1915) and his associates Trowbridge et al (1915) investigated the influence of maturity upon the yield, composition, digestibility and feeding value of timothy hay using digestion trials involving steers. They found that the coefficients of digestion for total dry matter, crude protein, crude fibre, and N.F.E. declined steadily throughout the five stages of cutting (plants just headed, full bloom, seeds formed, seeds in dough and seeds ripe). They next calculated the T.D.N. per acre in the D.M. of the hay at the different stages. If this figure is taken as 100 at full bloom, the figures for the succeeding stages of maturity were 93.7, 87.9, and 81.6 respectively. From the standpoint of feeding value per acre they therefore favoured cutting at the full bloom stage. They point out however, that from the management standpoint, Timothy hay at full bloom is rather high in moisture and curing conditions are more favourable at a later stage. They also found that cutting at or before full bloom resulted in a reduction of the stand. They explain this as being the result of inadequate storage of reserve material in the bulbs so that the plants cannot reproduce normally. This is not likely to be the case in Eastern Canada for two reasons:

(a) Timothy is usually grown with clover in a rotation
(b) The rainfall is heavier and permits a better second growth than is produced in the corn belt. The second crop of hay stands long enough to store reserve material in sufficient amounts to make the growth vigorous the following spring.

Waters (1915) found that the increased yields of dry matter obtained by allowing the hay to mature past the full bloom stage were not as high as popularly believed. Actually the dry matter yield per acre was within 4-5% as high at full bloom as at any of the later stages of development. This is confirmed by Bird (1943) for four different species of grasses. In this study timothy was found to reach the peak of its protein yield at an earlier stage than the other grasses studied, viz. Brome, Redgrass and Kentucky bluegrass.

Bell, Thatcher and Hunt (1932) found that the time at which timothy was cut for hay had a definite influence on its value for sheep feeding. Timothy cut when not more than one-third of the heads were in blossom was fully one-third better than when cut after the bloosoms had fallen and the seed was ripening. A marked superiority of early over late cutting of timothy is indicated by the work of others (Prince, Blood and Percival 1933; Reed and Huffman 1930; Salisbury and Morrison 1933).

Arny (1926) sums up the situation as follows - "as far as yield of digestible nutrients is concerned it appears desirable to cut timothy hay not later than the full bloom stage. Where cutting at this stage as a regular practice thins the stand materially so that serious reductions in yield occur, some sacrifice in quality of hay must be made in the interest of keeping the fields in a productive condition.

The effect of maturity on the chemical composition of numerous forage grasses and legumes has been investigated. Crampton and Jackson (1944) found a steady decline in the dry matter digestibility of mixed pasturage from around 75% for early spring grass to 60% some 6 weeks later. The digestibility from this time on did not follow any definite pattern but closely paralleled local climatic conditions of moisture and temperature.

In an investigation of wild hay meadows Stewart and Clark (1944) concluded that by delaying cutting until early September, the yield of protein decreased by 60 lbs. per acre or 28.9%, while the gain in hay yield was 11.4%. They point out that the gain in yield by delaying cutting time is of doubtful advantage since it would probably have been exceeded by the growth of aftermath of higher nutritive value.

Briggs et al (1947) and Baker et al (1947) investigated the change in chemical composition of prairie hay with maturity and found that the crude protein content and digestibility decreased as the season advanced. Similar studies have been made on crested wheatgrass (Sotola 1940), bearded wheatgrass (Burkitt 1940), mixed herbage (Crampton and Forshaw 1938), and alfalfa (Dawson et al 1940; Sotola 1947; Woodman et al 1934). The general trend of effect was much the same as described for timothy, and there would be no advantage in repeating all the findings in this review. A point that should be noted is that there is a species difference in the rate of response to the effects of maturity. Thus alfalfa tends to become woody and of decreased digestibility much earlier than do the grasses (Woodman et al 1934). Similarly Sotola (1941) has shown that bromegrass retains its high nutritive qualities to a more advanced maturity than does crested wheatgrass, and at identical stages of growth is superior to it in D.C.P. and T.D.N.

It must be borne in mind that the process of aging in the plant is extremely complex. Rate of maturity is conditioned not only by the genetic characteristics of the plant, but by such factors as rainfall, temperature, and fertilization. When these factors vary throughout the growing season as much as they do in Eastern Canada, the problem of assessing the contribution of each becomes evident. Further, Norman (1939) points out that since the growth of the herbage may be checked by grazing or accelerated by fertilizer treatment, and at the same time the botanical composition of the sward may be radically altered, it is obvious that we are dealing not with a simple cycle of growth changes but a sequence of superimposed effects not necessarily in phase.

The inter-relationship between the factors affecting maturity is still far from being understood, and too many investigators have been prone to attribute changes over a period of time solely to maturity when there may be other factors pushing or retarding the genetic tendency to age. Thus some investigators have described the effects of maturity over an interval of a number of days or over a number of inches of growth. From what has been said it would appear that such descriptions are completely inadequate and it should be realized that the age of the plant must be described physiologically rather than chronologically.

Effect of maturity on the morphological components - leaf, stem and head.

The majority of the investigators mentioned have confined their efforts to an examination of the effect of maturity on the entire forage plant. Evidently a fuller understanding of the effects of maturity would be obtained by breaking the plant down into its gross morphological components - leaf, stem, and head and investigating them separately.

Trowbridge et al (1915) has reported on the yields and composition of the heads, stalks with attached leaves, stubble and bulbs for timothy cut at various stages of maturity. Fagan and

Jones (1923) under British conditions made a similar analysis of cocksfoot, perennial rye grass, and timothy. However these workers too did not make a complete morphological separation and included the heads with the stems, thus making interpretation of the results difficult. Hosterman and Hall (1938) went a step further and separated the heads, stems, leaf blades and leaf sheaths of timothy. They used five stages of maturity and conducted analyses for protein, fibre and ether extract. Their results showed that the proportion of leaf blades decreased progressively from 38% for the early cut nearly full-headed timothy to 10% when fully matured. Little decrease took place after the stage when 10% of the heads were straw coloured. The relative weight of the stems increased from 27% for the nearly fully headed timothy to a maximum of 47% at full bloom and then decreased to 39% at full maturity. The relative weights of heads increased from 12% to 40% with the greatest increase occurring after the plants had passed full bloom.

Trowbridge et al (1915) attempted to account for these various losses and gains. They state that losses may in general be incurred by three methods.

1. Some of the leaves of the lower end of the stem wither and fall off at maturity or are beaten off by rains.
2. At the time of seed formation and ripening the stems and leaves contribute nutrients not only to the seed but also to the bulbs.

3. Soluble mineral matter exudes from the leaves and stems and is washed away by rain and dew. This occurs in large part at maturity.

The per cent protein of the leaf blades remained fairly constant until the closing stages of maturity when they suffered a sharp drop. The stems also decreased in per cent protein but more uniformly, while the heads underwent no appreciable change with maturity. When cut prior to full bloom 70% of the total protein was in the leaf and stem of the plant, whereas at full maturity 70% was in the heads. The authors state that in the latter stage the seeds are so easily shattered in curing and storage that it is doubtful if the heads are of much value.

The per cent fibre of the leaves did not change materially with maturity while that of the stems increased steadily. The fibre content of the heads was about half as great at maturity as in the early stages. The stems contained 40% of the total crude fibre in the early bloom stage and 56% at full maturity.

It is obvious that in the final analysis feeding trials of some kind must be done in order to evaluate the effect of time of cutting on the nutritive value of leaf, stem and head. Sotola (1933, 1946) investigated the digestibility of alfalfa and sweet clover leaves and stems over three and two seasons respectively. However he used only a single time of cutting for each season.

Thus while his results are indicative of the difference in feeding value of leaf and stem for these two crops, they do not show the effect of maturity within a season on the nutritive value of these components. A search of the literature failed to reveal any work along these lines for the grasses.

Indices of nutritive value.

A continuous search has been in progress over the years for some method of assessing and predicting the nutritive value of herbage without having to go through the laborious and expensive procedure of feeding trials with the animals in question. Laboratory animals such as rabbits do not necessarily give results that are applicable to ruminants.

The shortcomings of the ordinary feedstuffs analysis as an index of nutritive value have been previously dealt with by Norman (1935, 1939) and Crampton and Maynard (1938).

Quantity of protein and/or minerals has often been taken as being an index of nutritive value of forage. Crampton (1934) points out that only where these are liable to be limiting factors in terms of the requirements of the animal would this be true. A feeding trial reported by Graves (1933) does not indicate that the high protein of pasture herbage when fed to dairy cows increased its nutritive value over a ration of grass hay in which the protein level was much lower (9.9 - 14.9%).

Crampton (1934) has shown that quality of protein affects the nutritive value of herbage for growing rabbits. It seems doubtful if similar results would be obtained with ruminants whose microflora are capable of amino acid synthesis from various dietary nitrogenous sources.

There has been some attempt made to correlate the energy value of herbage with its nutritive value. Gross energy values are probably good indices only when the herbage is young. As the carbohydrate constituents become less available with maturity, gross energy values, unless coupled with digestibility data, fail to follow this trend. Available energy, on the other hand, is closely correlated with feeding value. It has been shown that the digestibility of pasture may vary from 80% to 60% during a season's growth, and such a decrease will diminish the availability of the energy yielding components. Thus using digestibility as a criterion of nutritive value the search has led to a study of the factors affecting digestibility with a view to being able to predict the nutritive value of herbage under varying conditions. The complexity of this problem is discussed by Norman (1939) and Crampton and Jackson (1944).

In view of its adverse effect on digestibility the degree of lignification of the plant has long been considered one of the most promising avenues of approach to the problem of evaluating nutritive value. However chemical methods for separating lignin have not been entirely satisfactory. Drapala, Raymond and Crampton (1947) have recently applied histological methods with promising results. Their support of an earlier observation of Woodman and Stewart (1932) that the mode of deposition of lignin as well as its amount influenced digestibility of herbage would appear to sound the death knell to any future hope of determining nutritive value through chemical estimation of lignin: mode of deposition could never be measured chemically. If lignin is to be used as an index of nutritive value, then histological methods would seem to be the logical method of approach.

Interest in the relative proportions of leaf and stem as an index of nutritive value has been stimulated ever since recording of the fact that a leafy as opposed to a stemmy habit of growth characterized the famous fatting fields of Romney (Fagan and Jones 1923). These authors, after analysis of leaves and stems of many grasses concluded that "A knowledge of the relative proportions of leaf and stem will prove a fair guide to the nutritive value of a pasture at any period of the year." This was substantiated by Stapledon (1933). Woodman and Evans (1935) using alfalfa, found that changes in the leaf-stem ratio largely accounted for the fall in protein content, the rise in fibre, and the running off in digestibility that takes place as the crop passes on to the flowering stage. Sotola (1937), also on the basis of chemical composition, concluded that the stem-leaf-head ratio of cereal hays is a fairly reliable index of their nutritive values.

Reliable conclusions based on results of the actual feeding value of leaf as compared with stem are difficult to find. Sotola (1933, 1946) has compared the feeding value of the leaf and stem of alfalfa and of sweet clover hay. In the latter case he found that the leaf protein was 45% more digestible than that of the stems. Apparently there is no comparable work on the relative feeding value of leaf and stem reported for the grasses. The reason for this has probably been the difficulty of separating the two components in sufficient quantity for conducting feeding trials.

EXPERIMENTAL PROCEDURE

General.

Timothy hay was harvested at two stages of maturity which were chosen to represent the approximate limits between which most hay is harvested in Eastern Canada. After curing, each cut of hay was separated into leaf, stem and head. The digestibility by sheep of each of these fractions and of the entire hay was determined. In this way it was hoped to establish the effect of stage of cutting on the feeding value of these fractions. The relative proportions of leaf, stem and head at the two stages of cutting were also determined to establish the relationship between their proportions (which are known to change with maturity) and the nutritive value of the whole forage.

Preliminary trials and previous experience had indicated that some of the forage fractions were not very acceptable to the sheep, and they were unable to maintain their body weight over the long period required for the trials. This ruled out the possibility of determining digestibilities by direct feeding of the leaf, stem and head. Consequently the method proposed was that of Carbery, Chatterjee and Hye (1934). This method is based on the fact that when a given foodstuff (in this case any of the timothy fractions) is fed in combination with a basal ration, but at different levels, the amounts of feed consumed in each digestion trial and the resulting digestion coefficients for the total ration are easily calculated. The unknown factors are the digestibilities of each of the component feeds. This may be stated algebraically and solved for the unknown by means of a multiple regression equation:

$$y = b_1x_1 + b_2x_2$$

where,

x_1 = Amount of basal ration consumed
x_2 = Amount of forage consumed
b_1 = Digestible fraction of the basal ration
b_2 = Digestible fraction of the forage
y = Total amount digested from the forage + basal ration.

This equation is solved by means of the two following simultaneous equations:

$$\Sigma(x_1y) = b_1\ \Sigma(x_1)^2 + b_2\ \Sigma(x_1x_2)$$

$$\Sigma(x_2y) = b_1\ \Sigma(x_1x_2) + b_2\ \Sigma(x_2)^2$$

A graphical illustration of this method is shown in Appendix Figure I.

In order to increase the acceptability of the ration and raise its energy value, a combination of barley and molasses was used as a basal ration. To this the timothy fraction whose digestibility was to be determined was added at three successive levels.

The proportion of barley to molasses in the basal ration was kept constant throughout all tests. Preliminary trials indicated that a ratio of 4 to 1 by weight (air-dry basis) of barley to molasses was most satisfactory. The timothy fractions were added to this fixed basal ration at three different levels as outlined in the following scheme. (All figures on an air-dry basis).

Level	Timothy fraction	Basal ration
1	1.5	1.0
2	1.0	1.0
3	0.5	1.0

For convenience a feeding chart was made up to show the amounts of the different feeds that would fulfill the above conditions for a wide range of possible feed intakes at each level. In this way the feed allowance for any trial was set by the appetite of the individual sheep, and the amounts of hay, barley and molasses corresponding to any amount of total feed could be read off the chart.

The allotment plan used was as follows:

TABLE I - Allotment Plan (4 sheep per trial)

Stage of maturity	Forage fraction	Trial No.	Proportion of forage	Proportion of basal ration
Early bloom	Stem	1	1.5	1.0
		2	1.0	1.0
		3	0.5	1.0
	Entire	4	1.5	1.0
		5	1.0	1.0
		6	0.5	1.0
	Head	7	1.5	1.0
		8	1.0	1.0
		9	0.5	1.0
	Leaf	10	1.5	1.0
		11	1.0	1.0
		12	0.5	1.0
Seed almost mature	Stem	13	1.5	1.0
		14	1.0	1.0
		15	0.5	1.0
	Entire	16	1.5	1.0
		17	1.0	1.0
		18	0.5	1.0
	Head	19	1.5	1.0
		20	1.0	1.0
		21	0.5	1.0
	Leaf	22	1.5	1.0
		23	1.0	1.0
		24	0.5	1.0

Two lots of 4 sheep each were used for the 2 stages of maturity respectively.

Preparation of forage.

Timothy hay was harvested at two stages of maturity, and after being field cured was separated into leaf, stem and head. Enough was taken to provide about 150 lbs. each of these fractions for conducting digestibility trials with sheep. The hay was taken from a pure stand of timothy (Milton variety) in which there was no more than 1-2% of weeds. In order to facilitate later separation of the hay into leaf, stem and head a binder was used to harvest the crop. Justification for this unusual method of harvesting hay will be apparent when the method of separation is described.

Part of the stand was cut on June 29th,1949 just after the flower of early bloom had appeared. The plants were smaller than average due to an exceptionally dry Spring. The remainder of the stand was left to mature 23 days longer before being cut. At this stage the crop was almost fully mature. Most of the heads had turned brown and the seeds were almost fully mature. The binder was set so that it cut the forage about three inches above the ground. The hay was field cured by a gradual process of turning the sheaves inside out and exposing them to the wind and sun. They were stooked and covered with canvas whenever there was a threat of rain, so that they escaped the effects of leaching.

After curing was completed the sheaves were assembled for separation into leaf, stem and head. This presented a problem which was solved only after considerable trial and error.

Manual separation as Sotola (1933) had done for alfalfa was impossible in this case. Eventually a method was evolved which gave a good and relatively rapid separation. The heads were first cut off by guiding the sheaves into a cutter box and withdrawing them at an appropriate time so that a minimum of stalks were included with the heads. This process removed the heads and chopped them into one to two inch lengths suitable for feeding in a single operation. About 10% stem stalks were inevitably included with the heads.

Owing to the difference in length of the timothy plants within a sheaf it was impossible to remove all the heads by the above operation. Their removal was accomplished by placing the sheaf on a block and making a cut with a meat cleaver which removed that part of the sheaf containing most of the remaining heads. This section was discarded and any heads remaining in the sheaves were removed by hand.

The next operation was to separate leaf and stem from the headless sheaves. The latter were first put through the cutter box which chopped them into short lengths. A threshing machine was then used to effect the separation of this material into leaf and stem. The chopped foraged was fed into the thresher where a combination of spiked, rapidly revolving rollers shattered it and loosened the leaf sheaths from the stems. The material was thrown around in its passage along the thresher through which a strong current of air was swept. The effect was that the lighter leaf and leaf sheath were blown along while the heavier stem gravitated downwards and was collected through spouts leading from the bottom of the machine. A barrier placed about two feet from the end of the machine (which was completely open) ensured that only the lighter leaf which was blown over the barrier was collected. Any stem which was blown through was stopped by the barrier and run through the machine a second time. The overall result was that the stem was collected in virtually a pure state, and the leaf and leaf sheath were together relatively free from stem.

Portions of the entire sheaves from both early and late cuts were also chopped and bagged, thus giving four feeding fractions for each stage of maturity.

Some loss of seed from the heads was encountered with the late cut hay in curing and in later handling. This loss was probably at least as great as would be encountered under practical farming conditions.

The relative proportions of leaf, stem and head in the entire forage were determined on representative samples taken from the cured sheaves. The separation was made by hand and the results computed to a dry matter basis. Leaf sheath was included with the leaf.

Management of digestibility trials.

Eight Cheviot wether lambs, about five months of age, and in good condition were used in the trials. They were divided into two equal groups, four lambs being fed the various fractions of the early bloom forage, while the other group was fed the late cut material. This procedure was necessary in order to economize on time by feeding the early and late cuts simultaneously. The forage was in all cases combined with the basal ration as previously outlined.

The lambs were placed in metabolism crates and fitted with leather harnesses to which removable canvas sacks could be attached for collection of faeces. The floors of the crates were so designed that the urine passed through a grill under which a large funnel leading into a collecting bottle was placed.

The animals were fed and their faeces sacks emptied once daily. Water was available at all times and was changed daily. Iodized salt licks were provided at first, but it was found that some of the animals ate them and developed diarrhea. Consequently the licks were removed and salt was fed with the ration at the rate of one teaspoonful per animal daily.

In preparing the feed it was found most convenient to weigh out the hay portion first. The molasses was then added and mixed thoroughly with the hay. The coarsely ground barley was added last and mixed to form a coating over the hay. When mixed in this way the ration appeared to be most acceptable to the animals. The molasses was diluted with water 2:1 by weight in order to facilitate weighing and mixing it with the forage. For convenience a stock solution of the diluted molasses was made up periodically and stored in a large bottle.

The trials were each of thirteen days duration during which the daily feed intake was kept constant. This was made up of a preliminary period of 5 days followed by an 8 day collection period during which total faeces output by each animal was recorded. During this period the faeces were sampled each day and a 20% aliquot was placed in a small dish and oven-dried at 105°C for 24 hours to determine the dry matter output. These dry aliquots were weighed and composited daily for each animal, and at the end of each test they were ground in a hammer-mill using a fine mesh and bottled for ehemical analysis. Samples of all feeds were also composited daily for chemical analysis.

Some difficulty was encountered at level 3 of the various trials where the proportion of basal ration to forage was relatively high. It would appear that the total crude fibre of the ration was insufficient at this level to allow normal rumination, and some cases of diarrhea and vomiting occurred. However these cases were relatively few and after a change to a roughage diet for a few days the animals concerned were able to complete their tests successfully.

Chewing of the wooden crates by the animals was suddenly encountered when the trials were about three-quarters completed. It started while feeding the third level of heads and continued in varying degree throughout the feeding of the leaf. Uncomplicated phosphorus deficiency or an insufficiency of fibre in the ration may have been contributing factors. However no serious effect on the results of the trials could be detected.

Chemical analyses.

Standard A.O.A.C. procedures were used in determinations of the proximate principles of feed and faeces samples. Complete proximate analyses were made of the pure leaf, stem and head separated by hand, and of the entire forage for both stages of maturity. Energy values were also determined by means of the bomb calorimeter.

Since digestibilities of dry matter, crude protein, crude fibre, and energy were to be calculated, analysis of feeds and faces samples were limited to these components.

The faeces samples from each test were composited into a single unit so that the analytical results and digestibilities obtained were averages for the four animals concerned.

RESULTS AND DISCUSSION

General.

A series of tabulated results are presented in the following pages which show the morphological and chemical analysis, and the nutritive value as measured by digestibility of the leaf, stem and head of timothy hay at two stages of maturity. The two stages shown are early bloom and that where the seed was almost mature, these representing the approximate limits between which most hay is harvested in Eastern Canada. The data shown are therefore the net results of changes that have occurred in leaf, stem and head between these stages of maturity. Since no intermediate stages were considered it is impossible to trace completely the sequence of changes.

For convenience the changes in leaf, stem and head will be discussed separately. The net effect of all these changes should be reflected in the results obtained for the entire hay, which will then be presented.

At this point it must be made clear that the digestibility coefficients which were obtained by the multiple regression method of Carbery et al (1934) do not appear to be entirely reliable. This is especially true for the crude fibre coefficients. The method is based on the premise that the digestibility of the basal ration (barley and molasses) remains constant throughout the tests. Actually calculations of these coefficients (Appendix Table II) showed that they varied quite considerably for the basal ration over the trials. However since the digestibility of the roughage is used in calculation of these coefficients it is impossible to say if these differences in digestibility of the basal ration are real. In view of some of the recent literature (Briggs et al 1938-40, Burroughs et al, 1949, Hamilton 1942, Swift et al 1947, Burroughs et al 1949) on the effects of sources of readily available carbohydrate in depressing roughage digestibility, it seems quite possible that the variable results obtained were a reflection of fluctuating microfloral activity with changing amounts and proportions of molasses in the diet. This would tend to support the rapidly growing belief that digestibility by ruminants is conditioned by the selective activity of a very dynamic microfloral population which is very sensitive to changes in diet and to fulfillment of their own dietary requirements.

Unfortunately the design of the experiment did not permit an estimate of error of the regression coefficients. An interesting comparison might have been made by feeding one or more of the hay fractions without the basal diet and determining the digestibility coefficients directly. However time did not permit such a study.

It seems probable that while the absolute values of the coefficients are not entirely reliable, they none the less serve to illustrate relative changes in digestibility with maturity. Only in the case of the crude fibre values must extreme reservation be placed on the results shown.

TABLE II - Analysis and Digestibility of Timothy leaf (Blade and Sheath) (Dry matter basis)

Factor	Early bloom	Seed almost mature
Morphological		
% of entire plant	39.3	32.8
Chemical composition (%)		
Gross energy (Cals/gm)	4.88	4.65
Crude Protein	8.88	5.67
Crude Fibre	32.3	34.3
Ether extract	3.12	2.00
Ash	5.47	6.19
N.F.E. (by diff.)	50.2	51.9
Digestibility % (av. of 4 sheep)		
Dry matter	56	51
Energy	53	47
Crude Protein	50 (44)*	30
Crude Fibre	58	59

*Indicates a value that is probably more reliable as indicated by the graphical method (See Appendix Figure I).

Effect of maturity on the composition and digestibility of timothy hay.

Leaf.

Table II shows that the proportion of leaf blades and sheath decreased by about 6% betweem early bloom and the stage where the seed was almost mature. This differential was not as high as that noted for the same variety of timothy taken from an adjoining field in the previous year. Differences of this type would be largely a result of soil and weather conditions.

Maturity resulted in rather a high drop in the proportion of leaf protein. Hosterman and Hall (1938) have shown that the decrease starts only after full bloom, is very sudden in the final stages of maturity, and is confined to the leaf blades rather than the sheaths. The caloric value of the leaf decreased somewhat with maturity due to a decrease in ether extract and an increase in ash. This is in contrast to the results of the above workers who reported a definite increase in the proportion of ether extract of the leaf with maturity. The crude fibre percentage increased only slightly with maturity.

The digestibility studies show that the nutritive value of the leaf remained comparatively high with maturity. The digestibility of dry matter and of calories dropped only about 5%. However the drop in digestibility of the leaf protein was rather high, amounting to about 15%. No decrease in availability of crude fibre was noted but it must be remembered that the validity of the fibre coefficients are very questionable.

The data point to the fact that lignification of leaf does not take place to any great extent with maturity. This is indicated by the observation that the chemical components studied, with the exception of protein, did not lose much of their availability with maturity. The immature leaves are an excellent source of digestible protein. However towards the end of maturity their stores of protein are depleted and appear to be taken to the head of the plant where they are incorporated into the seeds. The protein remaining in the leaf appears to be altered in such a way that its availability is impaired.

TABLE III - Analysis and Digestibility of Timothy Stems
(Dry matter basis)

Factor	Early bloom	Seed almost mature
Morphological		
% of Entire plant	40.8	40.2
Chemical composition (%)		
Gross energy (Cals/gm)	4.70	4.68
Crude Protein	4.60	3.06
Crude Fibre	41.5	41.1
Ether extract	1.30	0.82
Ash	3.20	3.80
N.F.E. (By diff.)	49.4	51.2
Digestibility % (av. of 4 sheep)		
Dry matter	51	38
Energy	47	37
Crude Protein	19 (28)*	9
Crude Fibre	57	32

*Indicates a value that is probably more reliable as indicated by the graphical method.

Stem.

Table III shows that the proportion of stem in the entire plant was about the same at the two stages of maturity considered. However Hosterman and Hall (1938) have shown that there is a considerable change in proportion of stem between these two stages. It rises gradually to a maximum around the time of full bloom after which it drops quite rapidly with the increase in dry matter of the heads in the final stages of maturity.

The crude fibre analysis and digestibility are especially interesting. They indicate that while the proportion of crude fibre in the stem is the same at both stages of maturity, its digestibility has dropped by about 25%. This shows very clearly the fallacy of using percent crude fibre as an index of nutritive value. Evidently lignification of the stems with maturity was responsible for the marked drop in digestibility of crude fibre. This laying down of lignin with maturity probably protected all other cell nutrients from bacterial action so that they passed through the alimentary tract largely unchanged by the digestive juices. The net result was a serious decrease in digestibility of all chemical components of the mature stem. It may be concluded that while the stem of early bloom timothy is a good source of digestible nutrients, the mature stems are lignified and of very much diminished value.

TABLE IV - Analysis and Digestibility of Timothy Heads
(Dry matter basis)

Factor	Early bloom	Seed almost mature
Morphological		
% of Entire plant	19.9	27.0
Chemical composition (%)		
Gross energy (Cals/gm)	4.88	4.85
Crude Protein	12.25	14.95
Crude Fibre	28.5	14.9
Ether extract	2.79	3.44
Ash	5.72	5.24
N.F.E. (By diff.)	50.7	61.4
Digestibility % (Av. of 4 sheep)		
Dry matter	54	52
Energy	51	48
Crude Protein	58 (49)*	41
Crude Fibre	60	58

*Indicates a value that is probably more reliable as indicated by the graphical method.

Heads.

Table IV shows that the proportion of dry matter of the heads in the hay increases substantially with maturity. This increase is most sudden towards the end of maturity as the seeds begin to fill and ripen. It has been shown that with maturity there is a drop in the nutrients of the leaf and stem, and it is probable therefore that they are being translocated to the head. The percent fibre of the head shows an increase, but this is undoubtedly due to the higher proportions of other nutrients as the head matures.

The digestibility studies revealed the interesting fact that the heads of the plant retain their high nutritive value with maturity. Only a 3% drop of dry matter digestibility was noted, and this was closely paralleled by the energy digestibility. The indication is that the seeds of mature hay are well digested by sheep. It is true that they were not quite mature, but their seed coats were sufficiently hard that they could not be fractured by pressure from a finger nail. It is doubtful if any serious lowering of digestibility would have occurred with full maturity of the seed.

The only chemical component that underwent any serious decrease in digestibility was crude protein where a drop of 10% was noted. Since about 10% stem stalks were included with the heads it is possible that the observed lowering of digestibility was entirely due to this contamination. It may be concluded then that the heads suffer little or no loss of availability with maturity, and with their increasing content of dry matter become an important source of digestible nutrients with maturity.

TABLE V - Analysis and Digestibility of the Entire Timothy Plant. (Dry matter basis)

Factor	Early bloom	Seed almost mature
Morphological		
% Leaf	39.3	32.8
% Stem	40.8	40.2
% Head	19.9	27.0
Chemical composition (%)		
Gross Energy (Cal/gm)	4.76	4.67
Crude Protein	6.61	6.72
Crude Fibre	38.7	32.6
Ether extract	2.26	2.44
Ash	4.31	4.55
N.F.E. (by diff.)	48.1	53.7
Digestibility % (av. of 4 sheep)		
Dry matter	58	49
Energy	55	51
Crude Protein	49	35
Crude Fibre	63	44

Entire plant.

Table V shows the morphological and chemical analyses and the digestion coefficients for the entire hay at two stages of maturity. Obviously these results should summarize the effect of maturity on the weighted mean of the leaf, stem and head, and therefore give the net changes in nutritive value of the hay.

The chemical composition of the plant as measured by the feedingstuffs analysis does not change appreciably, and certainly is not indicative of the decrease in nutritive value that occurs with maturity. The protein digestibility dropped by almost 15% while that of crude fibre suffered about a 20% decrease. However the energy digestibility was only lowered by about 5% indicating that the soluble carbohydrates and ether extract could not have undergone very serious lowering of digestibility.

It has been shown that the marked decrease in availability of protein with maturity must be attributed to both leaf and stem, while the drop in crude fibre digestibility is due almost entirely to the stem.

The problem of estimating nutritive value.

Nutritive value of grass taken alone is not of too much significance. A grass may be of high nutritive value at a certain time of the season, but if its growth habits do not permit harvesting at this time then the usefulness of its high feeding value is eclipsed.

Stapledon (1933) considers that at the same stage of maturity - rapid growth - all grasses have about the same feeding value as measured by grazing sheep. Thus the nutritive value of any grass must be stated in terms of some definite stage of its growth, and in general the usefulness of a high nutritive value will be confined to that time at which the grass can economically and conveniently be harvested or grazed without detriment to the sward. Nutritive value is a dynamic factor and subject to many varying conditions. Meteorological conditions above all determine rate of growth over and above the inherent growth characteristics of the plant under "standard" conditions. It seems very probable that the interaction of genetic and meteorological factors is responsible to a major degree for the energy value (if not nutritive value) of the plant at any particular stage of its growth.

To carry this point further: the nutritive value of a plant must be conditioned by the ability of the plant to absorb nutrients from the soil and convert them to their tissues in a form readily given up to the digestive processes of the animal. If Stapledon (1933) is correct in stating that the young rapidly growing grasses are all of equal nutritive value, then it follows that the decrease in nutritive value that exists after this stage of rapid growth should be proportional to the tendency of the plant to stop vegetative growth and proceed to seed formation. Slowing of vegetative growth is accompanied by lignification, encrustation and walling off of the nutrients, especially of the stem, thereby making them less vulnerable to the digestive processes and decreasing the nutritive value of the plant. It follows therefore that processes, whether inherent to the plant or of outside origin, that will maintain vegetative growth will serve to maintain nutritive value. Water, more than any other factor is likely to limit rate of growth. In this connection it would be of interest to calculate the correlation between nutritive value and

(a) Rate of growth

(b) Depth of root system (where water is a limiting factor).

Such methods of approach may possibly elucidate the differences in nutritive value between species.

For the individual plant it has been shown in this study that the decrease in nutritive value as the plant matures is mainly due to changes in the nature of the stem. Chemical methods have so far failed to indicate these changes in the stem, and have therefore failed as indices of nutritive value. Obviously too the amount of stem present will be no index since its proportion does not change in the same way as its own nutritive value or that of the entire plant.

The proportion of leaf appears to be promising as an index of nutritive value. The results of this study definitely indicate that leafier species will better maintain their nutritive value through the season since the leaf does not show the drastic decrease in digestibility with maturity characteristic of the stem. Further the proportion of leaf in the entire plant falls off with increasing maturity, and at the same time there is a decrease in the nutritive value of the plant. There may possibly be a physiological basis behind this observation whereby the rate at which the proportion of leaf falls off is proportional to the rate at which the plant in general and the stem in particular becomes unavailable. No conclusions can be drawn on this point from the present study since only two stages of maturity were considered. Such a relationship could be investigated by taking a large number of cuts of hay throughout the season and determining their proportions of leaf and digestibility.

Another promising method of study may be based on the fact that the decrease in nutritive value of the plant with maturity is mainly due to the stems. Histological methods of the type initiated by Drapala, Raymond and Crampton (1947) aimed at correlating digestibility with the microscopic appearance of stem sections may possibly lead to the use of a standard for evaluating nutritive value of grasses.

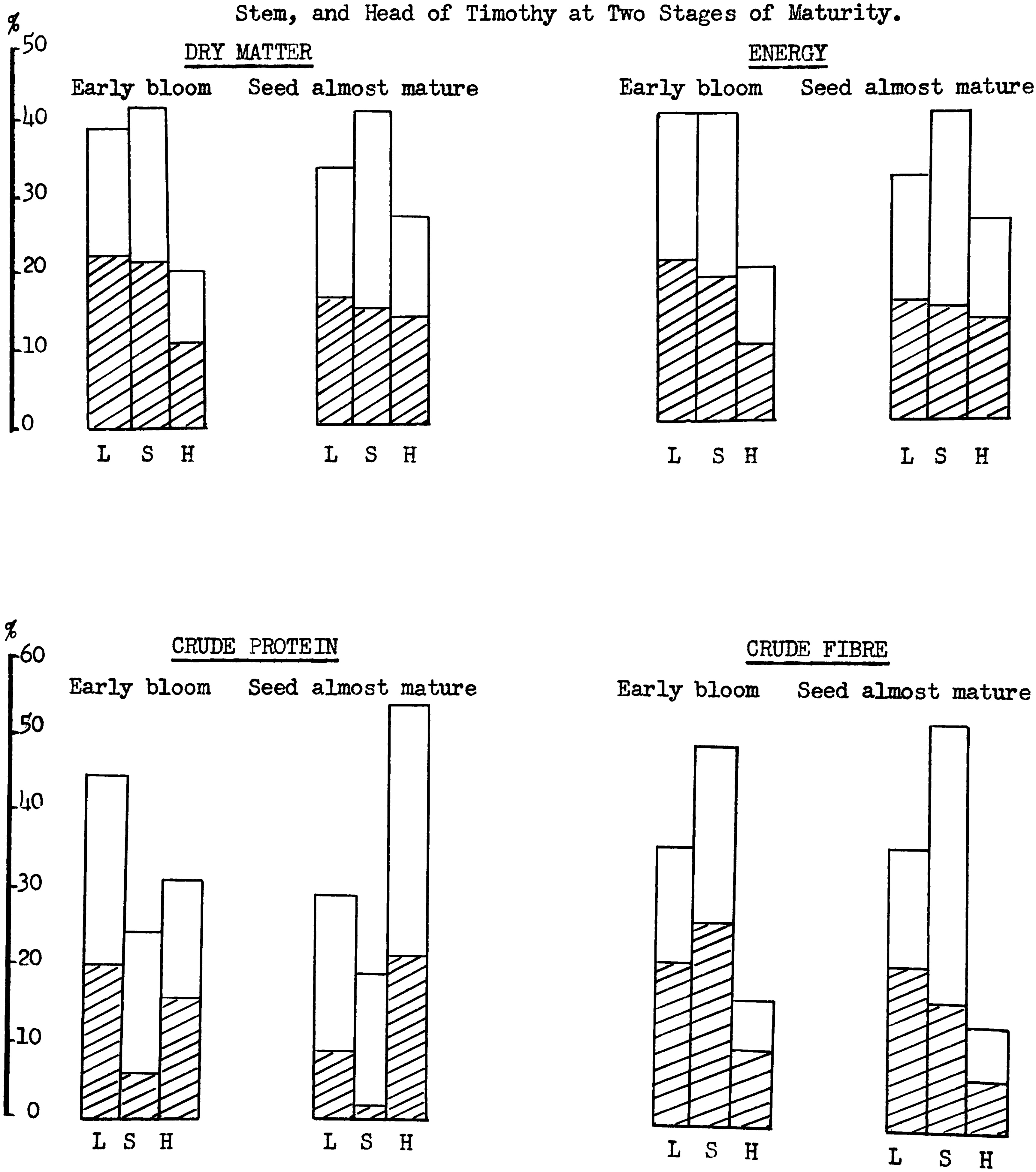
SUMMARY

The net changes in nutritive value of the leaf, stem and head of timothy hay between two stages of maturity have been investigated. The stages studied represented approximately those between which most hay is harvested in Eastern Canada. Supporting data were obtained on the net changes in morphological and chemical composition of the hay between these stages.

Average digestibility coefficients by four sheep were used as the criterion for measuring nutritive value. Digestibility of dry matter, energy, crude protein and crude fibre were determined using the multiple regression method of Carbery et al (1934), in which a basal ration of barley and molasses was fed at three successive levels along with the forage fractions whose digestibility was to be determined.

The results are summarized in Figure I (based on Appendix Table V) where the relative availability of the various proximate principles is projected onto the relative proportions in which these nutrients occur in the leaf, stem and head. The effect of maturity on the relationship between gross morphology, chemical composition and nutritive value of the plant is brought into clear focus.

FIG. I - Total and Digestible Nutrients Supplied by Leaf, Stem, and Head of Timothy at Two Stages of Maturity.



Key

Full blocks	=	% Total nutrients
Shaded "	=	% digestible nutrients
L	=	Leaf
S	=	Stem
H	=	Head

The proportion of leaf in the plant fell while there was a rise in the dry matter of the heads with seed formation. The availability of the stems was almost as high as that of the leaves up to the early bloom stage of maturity. However towards the end of maturity the digestibility of stem had markedly decreased.

The digestibility of the leaf decreased somewhat while that of the head remained nearly constant with maturity. (Interpolating for the fact that the heads were contaminated with about 10% stem). However it is only in the mature hay that the proportion of head is high enough to attain real significance as a contributor of digestible nutrients, and especially of protein.

The digestibility of protein was low for all parts of the hay, even at the early bloom stage. By maturity its availability had fallen seriously except in the heads, which at this time were supplying 68% of the digestible protein.

The possibility of using proportion of leaf, or histological examination of the stem as an index of nutritive value has been discussed.

CONCLUSIONS

1. The stemmy portions of timothy, except for a low contribution of digestible protein, are of fairly high nutritive value up to the early bloom stage. However at maturity a serious drop in digestibility of all stem nutrients has occurred which is not indicated by the conventional proximate analysis.
2. The leaf of the plant does not suffer nearly as high a drop in digestibility with maturity, indicating that lignification is not as active as in the stems.
3. The heads of the plant have a surprisingly high nutritive value which they retain with maturity. In the mature hay they make up about 30% of the plant and contribute about 68% of the digestible protein.
4. The digestibility of the stem and leaf protein is seriously lowered with maturity. The stem contributes little of the total digestible protein at or after early bloom.

LITERATURE CITED

Arny, A.C. 1926
The influence of time of cutting on the quality of crops.
Jour. Amer. Soc. Agron., 18:684-703.

Baker, M.L., V.H.Arthand, E.C. Conard, and L.C.Newell 1947
Effects of time of cutting on yield and feeding value of prairie hay. A progress report for 1945-46.
Nebr. Agr. Exp. Sta. Bull. 385.

Bell, D.S., L.E.Thatcher, and C.H.Hunt 1932
Experiments with timothy hay for pregnant and nursing ewes.
Proc. Amer. Soc. Animal Production:176-180.

Bird, J.N. 1943
Stage of cutting studies. 1.Grasses.
Journ. Amer. Soc. Agron. 35:845-861.

Briggs, H.M., V.G.Heller, and R. Wall 1938-40
Molasses reduces the digestibility of other components of rations.
Okla. Agr. Exp. Sta. Biennial Report. Vol.62.

_______ W.D.Gallup, J.S. Dinning and A.E.Darlow 1947
The yield and feeding value of prairie hay cut at different stages of maturity.
Jour. Animal Sci. 6:483.

Burkitt, W.H. 1940
The apparent digestibility and nutritive value of beardless wheatgrass at three stages of maturity.
Jour. Agr. Rsh. 61:471.

Burroughs, Wise, P. Gerlaugh and C.H.Edgington 1949
The influence of cornstarch upon roughage digestion in cattle.
Jour. Animal Sci. 8:271-278.

Carbery, M., I. Chatterjee and Md Abdul Hye 1934
Studies on the determination of digestibility coefficients. 1. A new method of experimentation and computation for directly obtaining the digestibility coefficients of individual feed nutrients in a mixed ration.
Indian Jour. Vet.Sci. and Animal Husb. 4:295.

Crampton, E.W. 1934 — The nutritive value of pasture herbage. Quality of protein. Emp. Jour. of Exp. Agr. 11:8.

________ and L.A.Maynard 1938 — Nutritive value of feeds. Jour. Nutr. 15:4.

________ and R.P.Forshaw 1939 — The intra-seasonal changes in the nutritive value of pasture herbage. Sci. Agr. 19:(12):701.

________ amd I.R.C.Jackson 1944 — Seasonal variation in chemical composition of pasture herbage and the relation to its digestibility by steers and sheep. Jour. Animal Sci. 3: (4): 333.

Dawson, J.R., D.V. Kopland and R.R.Graves 1940 — Yield, chemical composition, and feeding value for milk production of alfalfa hay cut at 3 stages of maturity. U.S.D.A. Tech. Bull. 739.

Drapala, W.J., L.C.Raymond and E.W.Crampton 1947 — The effects of maturity of the plant and its lignification and subsequent digestibility by animals as indicated by methods of plant histology. Sci. Agr. 27:1.

Fagan, F.W., and H.T.Jones 1923 — The nutritive value of grasses as shown by their chemical composition. Welsh plant breeding station. Series H No.3, Seasons 1920-23, University College of Wales, Aberystwyth.

Graves, R.R., J.R.Dawson, D.V.Kopland and T.W.Moseley 1933 — Feeding value for milk production of pasture grasses when grazed, when fed green, and when fed as hay or silage. U.S.D.A. Tech. Bull. 381.

Hamilton, T.S. 1942 — The effect of added glucose upon the digestibility of protein and of fibre in rations for sheep. Jour. Nutr. 23: 101-110.

Hosterman, W.H. and W.L. Hall 1938 — Time of cutting timothy: Effect on the proportion of leaf blades, leaf sheaths, stems and heads, and on their crude protein, ether extract and crude fibre contents.
Jour. Amer. Soc. Agron. 30:564-68.

Jordan. 1882 — Cited by Trowbridge P.F., L.D.Haigh, and C.R.Moulton.
Mo. Agr. Exp. Sta. Rsh. Bul. 20.1915.

Kellner, O. 1879 — Cited by Trowbridge, P.F., L.D. Haigh, and C.R.Moulton.
Mo. Agr. Exp. Sta. Rsh. Bul. 20:1915.

Kerr, W., and I.W. Bailey 1934 — Structure, optical properties, and chemical composition of the so-called middle lamella.
Jour. Arnold Arboretum. 15:(4):327.

Ladd. 1888 — Cited by Trowbridge, P.F., L.D.Haigh, and C.R.Moulton.
Mo. Agr. Exp. Sta. Rsh. Bul.20.1915.

Morse, F.W. 1891 — Growth of timothy grass.
N.H. Agr. Exp. Sta. Ann: Report 2:63-9.

Norman, A.G. 1935 — The composition of crude fibre.
Jour. Agr. Sci. 25: 529-40.

________ 1939 — Biochemical approach to grass problems,
Jour. Amer. Soc. Agron. 31: 751.

Patton, A.R. 1943 — Seasonal changes in the lignin and cellulose content of some Montana grasses.
Jour. Animal Sci. 2: (1):59.

________ and L. Gieseker 1942 — Seasonal changes in the lignin and cellulose content of some Montana grasses.
Journ. Animal Sci. 1: (1): 22.

Phillips, Max., M.J.Goss, B.L.Davis and H. Stevens 1939 — Composition of the various parts of the oak plant at successive stages of growth, with special reference to the formation of lignin.
Jour. Agr. Rsh. 59: 319-66.

Phillips, Max., B.L.Davis and H.D.Weihe
1942
Composition of the tops and roots of the timothy plant at successive stages of growth.
Jour. Agr. Rsh. 64: 533-46.

Phillips, T.G., T.O.Smith, and R.H.Harper
1943
The composition of timothy.
N.H. Agr. Exp. Sta. Tech. Bul.81.

Prince, F.S., P.T.Blood and G.P.Percival
1933
Studies of the feed value of early hay.
N.H. Agr. Exp. Sta. Cir. 41.

Reed, O.E., and C.F.Huffman
1930
The results of a five year mineral feeding investigation with dairy cattle.
Mich. Agr. Exp. Sta. Tech. Bul. 105.

Richardson, C.
1883
U.S.D.A. Annual Report 231.

Salisbury, G.W. and F.B. Morrison
1933
Value for dairy cows of early cut timothy hay fertilized with a nitrogenous fertilizer.
N.Y. (Cornell) Agr. Exp. Sta. Ann. Rpt. 1932-33: 86-87.

Sotola, J.
1927
Relation of maturity to the nutritive value of first, second and third cuttings of irrigated alfalfa.
Jour. Agr. Rsh. 35: 361-83.

——— 1933
The nutritive value of alfalfa leaves and stems.
Jour. Agr. Rsh. 47: 919-45.

——— 1937
The chemical composition and nutritive value of certain cereal hays as affected by plant maturity.
Jour. Agr. Rsh. 54: 399-415.

——— 1940
The chemical composition and apparent digestibility of nutrients in crested wheatgrass harvested in three stages of maturity.
Jour. Agr. Rsh. 61:4:303-311.

——— 1941
The chemical composition and apparent digestibility of nutrients in smooth bromegrass harvested in three stages of maturity.
Jour. Agr. Rsh. 63, 427.

Sotola, J.
1946
The effect of maturity on the chemical composition and digestibility of the stems and leaves of sweet clover hay.
Jour. Agr. Rsh. 72:12:365-71.

Stapledon, R.G.
1933
Leaf-stem ratio and digestibility.
Account of the organization and work of the station from Apr.1919 to July 1933.
Univ. College of Wales, Aberystwyth, Welsh plant breeding station.

Stewart, G. and J. Clark
1944
Effect of prolonged spring grazing on the yield amd quality of forage from wild hay meadows.
Jour. Amer. Soc. Agron. 36:238.

Swift, R.W., E.J. Thacker, A. Black, J.W. Bratzler and W.H.James
1947
Digestibility of rations for ruminants as affected by proportions of the nutrients.
Jour. Animal Sci. 6:432-44.

Trowbridge, P.F., L.D.Haigh and C.R.Moulton
1915
,Studies of the timothy plant. Part 2.
Mo. Agr. Exp. Sta. Rsh.Bul.20.

Waters, H.J.
1915
Studies of the timothy plant. Part 1.
Mo. Agr. Exp. Sta. Rsh.Bul.19.

Watson, S.J. and E.A.Horton
1936
The technique of digestibility trials with sheep and its application to rabbits.
Emp. J. Expt. Agr. 4:25.

Wilson
1884
Cited by Trowbridge P.F., L.D.Haigh and C.R.Moulton.
Mo. Agr. Exp. Sta. Rsh. Bul.20.

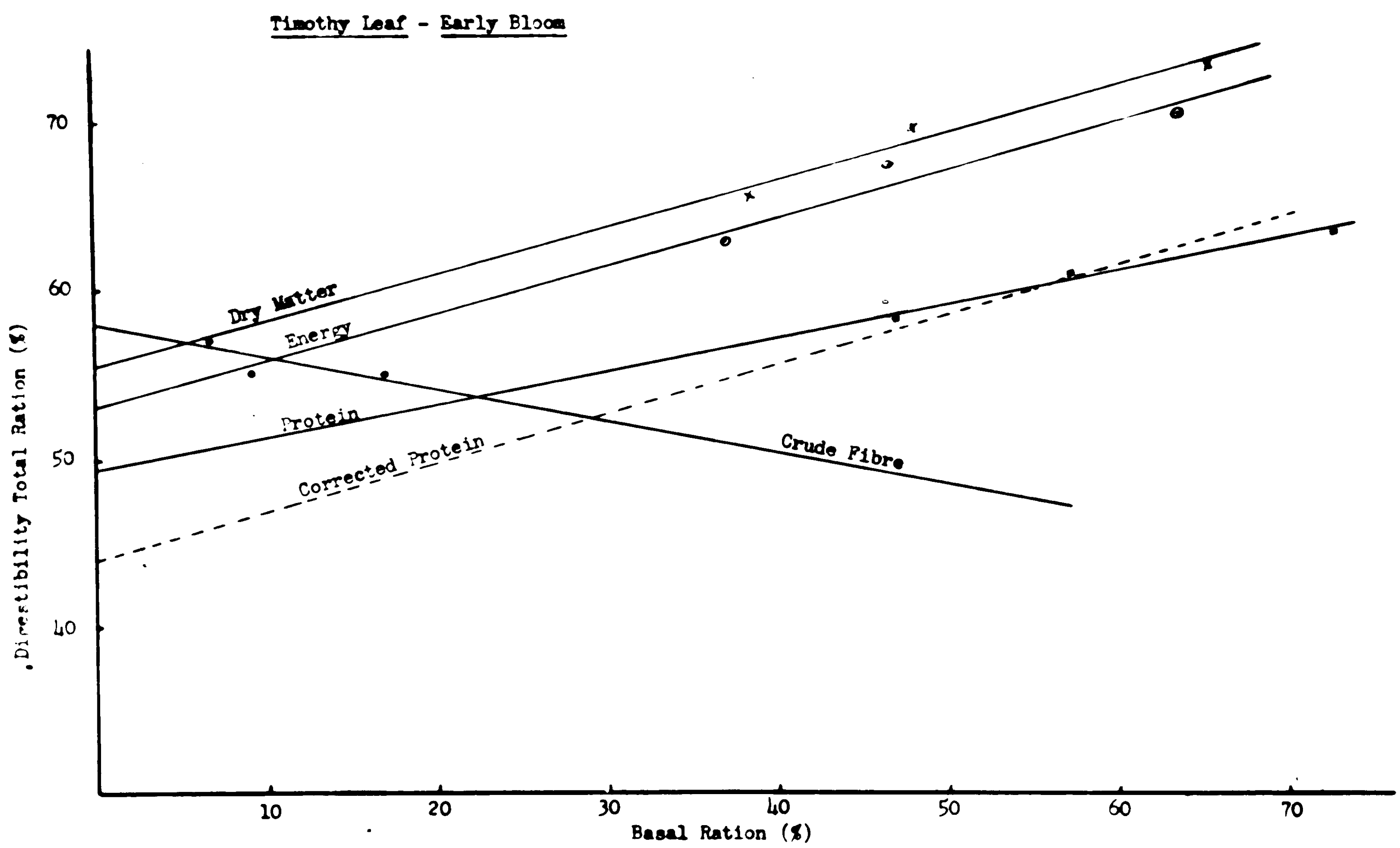
Woodman, H.E., D.B.Norman and M.H.French
1931
Nutritive value of pasture VII. The influence of intensity of grazing on the yield, composition, and nutritive value of pasture herbage.
Jour. Agr. Sci. 21:267-321.

__________ and J. Stewart
1932
The mechanism of cellulose digestion in the ruminant organism. III. The action of cellulose-splitting bacteria on the fibre of certain typical feedingstuffs.
Jour. Agr. Sci. 22:527-47.

Woodman, H.E., R.E.Evans and D.B.Norman
1934 Nutritive value of lucerne. II. Investigations into the influence of systematic cutting at three different stages of growth on the yield, composition, and nutritive value of lucerne.
Jour. Agr. Sci. 24:283-311.

__________, and R.E.Evans
1935 The nutritive value of lucerne. IV. The leaf-stem ratio.
Jour. Agr. Sci. 25:579.

APPENDIX



APPENDIX FIG. I - Graphical Illustration of Regression Method

APPENDIX TABLE I - Digestion Coefficients (%) for Whole Ration Including Early Bloom Timothy Hay. (Dry matter basis)

Forage fraction	Proportion forage to basal ration	Animal Number				Average
		1	2	3	4	
Stem	1.5/1	64.2	63.0	65.3	64.0	64.1
	1.0/1	62.8	66.8	67.0	66.0	65.6
	0.5/1	71.3	74.6	70.5	71.9	72.1
Entire	1.5/1	59.9	68.0	66.7	66.7	65.3
	1.0/1	62.1	70.2	66.8	70.4	67.4
	0.5/1	65.4	71.5	71.6	73.4	70.5
Head	1.5/1	59.4	69.1	66.0	66.6	65.3
	1.0/1	66.8	72.9	72.2	72.8	71.2
	0.5/1	69.5	77.0	73.9	73.4	73.4
Leaf	1.5/1	63.6	66.6	64.7	65.4	65.1
	1.0/1	69.2	70.1	69.0	70.1	69.6
	0.5/1	70.4	72.9	74.5	71.6	72.3

APPENDIX TABLE IA - Digestion Coefficients (%) for Whole Ration Including Mature Timothy Hay. (Dry matter basis)

Forage fraction	Proportion forage to basal ration	Animal Number				Average
		5	6	7	8	
Stem	1.5/1	58.4	60.8	57.8	57.8	58.7
	1.0/1	63.5	64.2	62.7	65.8	64.0
	0.5/1	71.9	71.9	74.7	75.1	73.4
Entire	1.5/1	59.1	60.9	62.8	60.2	60.5
	1.0/1	65.4	65.7	64.3	67.9	65.7
	0.5/1	68.5	71.2	69.3	69.8	69.7
Head	1.5/1	63.1	63.4	62.8	62.5	62.9
	1.0/1	64.1	65.1	64.6	67.1	65.2
	0.5/1	70.7	72.6	68.2	71.4	70.7
Leaf	1.5/1	63.2	63.4	64.5	66.1	64.3
	1.0/1	69.0	66.5	67.5	67.1	67.5
	0.5/1	72.1	73.4	72.8	74.9	73.3

APPENDIX TABLE II - Digestibility Coefficients (%) of Basal Ration (Barley and Molasses) When Fed with Hay Fractions. (Dry matter basis)

Hay fraction	Maturity	Dry matter	Energy	Crude protein	Crude* fibre
Leaf	Early	80	80	68	41
	Late	85	85	77	11
Stem	Early	83	83	74	-9
	Late	92	90	78	115
Head	Early	85	85	74	33
	Late	81	81	76	-21
Entire	Early	77	76	67	-6
	Late	80	76	76	-15

To indicate the reliability of the digestibility data for the hay fractions. Theoretically values in each column should be constant.

* Values unreliable due to relatively small amounts of fibre in the basal ration.

APPENDIX TABLE III - Chemical Analysis of Feeds (Dry matter basis).

Feed	Stage of maturity	Gross Energy (Cals/gm)	Crude protein %	Crude fibre %	Ether extract %	Ash %	N.F.E. (by diff.) %
Leaf	Early	4.84	8.02	35.3	3.05	5.32	48.3
	Late	4.77	6.03	35.7	2.62	6.18	49.4
Stem	Early	4.70	5.02	40.8	1.42	4.10	48.7
	Late	4.76	4.12	37.7	1.52	4.06	52.6
Head	Early	4.86	10.82	33.0	2.50	4.78	48.9
	Late	4.66	11.12	24.2	3.22	5.11	56.3
Entire	Early	4.76	6.61	38.7	2.26	4.31	48.1
	Late	4.67	6.72	32.6	2.44	4.55	53.7
Barley		4.74	13.36	4.9			
Molasses		3.72	2.66	-			

APPENDIX TABLE IV - Faeces Analysis (average of 4 sheep)
(Dry matter basis)

Feeding trial number	Crude Protein %	Crude Fibre %	Gross Energy (Cals/gm)
1	9.56	32.5	5.01
2	11.06	33.5	4.95
3	11.68	29.3	4.88
4	10.49	30.3	5.03
5	10.71	28.5	4.95
6	12.75	26.2	4.93
7	11.50	27.2	5.08
8	12.25	25.6	5.11
9	13.48	25.0	4.97
10	11.22	28.5	5.09
11	12.60	29.3	5.12
12	13.80	24.1	5.07
13	7.84	37.3	4.84
14	8.96	37.5	5.03
15	10.92	31.4	4.95
16	9.81	33.0	4.99
17	10.15	33.6	4.83
18	11.27	30.7	4.87
19	13.58	22.3	4.85
20	13.95	21.2	4.90
21	13.57	23.2	4.81
22	10.16	28.8	5.06
23	12.60	29.4	5.16
24	12.02	27.3	4.92

APPENDIX TABLE V - Effect of Maturity on Percentage of Total and Digestible Nutrients Supplied by Timothy Hay (Dry matter basis).

Hay fraction	Maturity	Dry Matter		Energy		Crude Protein		Crude Fibre	
		Total	Digestible	Total	Digestible	Total	Digestible	Total	Digestible
Leaf	Early bloom	39	41	40	42	45	49	36	36
	Seed almost mature	33	37	33	35	28	27	35	47
Stem	Early bloom	41	39	40	37	24	10	48	47
	Seed almost mature	40	33	40	34	19	5	52	37
Head	Early bloom	20	20	20	20	31	40	16	17
	Seed almost mature	27	30	27	30	53	68	13	16