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### STUDIES IN RUMINANT NUTRITION WITH PARTICULAR REFERENCE TO

#### NON-PROTEIN MITROGEN AND PHOSPHORUS

A Thesis submitted to the University of Glasgow for the degree of

# DCCTOR OF PHILOSOPHY

In the Faculty of Veterinary Medicine

by

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May 1974.

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#### Summary.

The work described is concerned with investigations into the fortification of dried molassed sugar beet pulp with non-protein nitrogen, phosphorus, trace elements and vitamins. Materials containing between 3 and 11 % of added urea and with up to 17 and 40 % crude protein and 0.55 % phosphorus were manufactured. The cubed products were evaluated in a wide range of nutritional studies with ruminants.

The General Introduction gives an account of the metabolism of urea in the ruminant together with a review of the methods available for reducing the potential toxicity of dietary nonprotein nitrogen.

Section 1 describes investigations into the release rate properties of urea--containing molassed sugar beet pulp cubes. Experiments in vivo indicated that both the size and hardness of the cubes were important factors determining the rate and degree of ammonia production in the rumen. Whole hard molassed sugar beet pulp cubes were shown to have superior slow release properties to those of either a softer cube or a smaller barley based product containing similar quantities of urea.

In Section 2 urea phosphate, mono-ammonium phosphate and a granular ammonium polyphosphate were evaluated in balance trials with growing sheep as possible combined phosphorus and nitrogen supplements for inclusion in molassed sugar beet pulp. When mixed with additional urea and compared with equivalent amounts of phosphorus and nitrogen supplied as dicalcium phosphate and urea each material promoted similarly improved nitrogen and phosphorus retentions.

Section 3 describes the evaluation using growing sheep of a magnesium phosphate and a calcium magnesium phosphate as combined phosphorus and magnesium supplements. Comparisons were made with

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equivalent amounts of phosphorus and magnesium given as dicalcium phosphate and magnesium oxide. All three phosphorus sources promoted comparable daily retentions of phosphorus. The magnesium of both combined phosphorus and magnesium supplements was as well utilised as that of magnesium oxide. Magnesium phosphate was shown to be an effective prophylactic agent for the prevention of hypomagnesaemic tetany in lactating beef cows.

In Section 4 the effect of urea and phosphorus contained in molassed sugar beet pulp on the voluntary consumption of lowprotein roughage feeds was studied. Inclusion of either 3.0 or 7.8 % urea in 1 kg per day of molassed sugar beet pulp containing 0.55 % phosphorus similarly increased the voluntary intake of oat straw by steers by about 20 %. Increased straw intake was accompanied by an improvement in the digestibility of the dry matter and crude fibre of the straw. The addition of 3.0 % urea to 2.7 kg per day of molassed sugar beet pulp increased the voluntary consumption of oat straw by pregnant beef heifers by about 21 %. Increasing the total phosphorus intake of the heifers from about 6 to 17 g P/day with dicalcium phosphate did not increase straw intake in either the presence or absence of additional urea. Supplementation with urea tended to increase the digestibility of the straw and the concentration of glucose in the blood plasma. The voluntary consumption of hay was reduced by about 20 % in late pregnancy when no supplementary urea was given. A continued low protein intake in late pregnancy did not influence the immune lactoglobulin concentration in the colostrum of the heifers or the birth weight or quantity of immune lactoglobulin absorbed by their newborn calves.

Section 5 describes experiments in which a molassed sugar beet pulp product containing 32 % crude protein was compared with a mixture of equal parts of decorticated groundnut and cottonseed meals as a protein source for rapidly growing 100 kg steers. Both forms of

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supplementation equally and significantly increased live-weight gains and improved food conversion ratios compared with those recorded for an unsupplemented diet. A high urea molassed sugar beet pulp product was somewhat less effective than a conventional protein source as a supplement for rapidly growing 20 kg sheep.

In Section 6 molassed sugar beet pulp materials containing either 2.7 or 7.8 % of added urea were used to replace about onehalf of the additional protein derived from vegetable protein sources in a milk production concentrate given to dairy cows. The concentrate mixture containing the high urea molassed sugar beet pulp material presented considerable palatability problems and was associated with a marked decline in milk yield. Fully satisfactory milk yields were obtained with the milk production concentrate which included 50 % of the molassed sugar beet pulp material supplemented with 2.7 % urea.

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#### GENERAL INTRODUCTION

Ruminant animals have the ability to convert non-protein nitrogen to microbial protein in the rumen. The microbial protein synthesised is then available to the animal as a source of dietary protein.

Apart from the small amounts of non-protein nitrogen naturally present in normal feedstuffs, comparatively large amounts of nonprotein nitrogen are continuously presented to the ruminant digestive tract in the form of urea, which enters endogenously via the saliva, or by diffusion across the rumen wall. (Houpt and Houpt, 1968).

Non-protein nitrogen utilisation is an integral part of normal digestion in the ruminant. Much research effort has been devoted to the evaluation of added non-protein nitrogen materials as partial replacements for the natural proteins present in diets for ruminants. To date, urea is the only material readily and cheaply available.

It has been clearly demonstrated in the experiments of Virtanen (1966) and Oltjen (1969) that reasonably satisfactory growth, reproduction and lactation could be achieved when urea formed as much as 97 % of the total nitrogen of largely synthetic diets fed to ruminants over extended periods of time. Animal productivity was however less than would normally be considered desirable when entirely natural protein diets are fed.

In many practical animal husbandry systems it is possible that the somewhat reduced productivity which might result from feeding non-protein nitrogen may be offset by the economics and commercial availability of compounds such as urea, compared with plant or animal proteins.

In terms of present day (1974) prices urea is a substantially cheaper nitrogen supplement than vegetable protein. Decorticated

groundnut containing 45 % crude protein is commercially available at about £120/tonne. When an allowance of £50/tonne is made for the energy content of groundnut, it can be calculated that 1 kg of crude protein costs 15 p. In marked contrast, 1 kg of crude protein equivalent supplied as urea (£40/tonne) costs 1.4 p.

In the introduction to this Thesis an account of the properties, general metabolism and methods of reducing the potential toxicity of urea will be given. The scope of the experimental investigations undertaken is also outlined. Those parts of the very extensive literature on urea feeding and metabolism in the ruminant which bear most directly on the work undertaken and its subsequent interpretation will be reviewed. Detailed literature reviews involving specific aspects of urea and non-protein feeding are included in the introductions to the appropriate experimental sections.

#### Composition and general properties of urea.

Urea is manufactured by the reaction of carbon dioxide and ammonia under high temperature and pressure. It is available in solid form as off-white or tan coloured prills and crystals. It has no odour, but has a cool, saline taste which makes it somewhat unpalatable to livestock. Urea has a very low (c. 0.2 %) moisture content and whilst not very hygroscopic it should be stored in a dry, cool place. It is very soluble in water (40 and 87 g/100 g at 0°C and 100°C respectively). In the solid form urea is stable up to its melting point (132°C) when it begins to decompose to form biuret and other related compounds before final decomposition to carbon dioxide and ammonia. A saturated solution of urea does not begin to decompose appreciably below 70°C when hydrolysis produces detectable quantities of carbon dioxide and ammonia.

Urea  $(H_2N-CO-NH_2)$  contains 46.6 % N which is theoretically equivalent (% N x 6.25) to 291 % crude protein. Commercially available urea usually contains between 42 and 45 % N ( $\equiv$  262-281 % crude protein) because of the inclusion of small amounts of inert materials such as kaolin added to reduce its natural hygroscopicity and improve its handling and mixing properties. Under normal storage conditions (i.e. not unduly damp or alkaline) urea is unlikely to react with cereal products and oil-seed residues present in animal feedingstuffs.

#### Urea metabolism in the ruminant.

Dietary urea entering the rumen is rapidly hydrolysed by the bacterial enzyme urease to ammonia and carbon dioxide (Pearson and Smith, 1943). The degradation of true protein proceeds more slowly, via the intermediate compounds of peptides and amino acids, which may be further altered to ammonia and volatile fatty acids. The quantity of nitrogen present as free amino acids in the rumen is relatively small (Annison, 1956) as rumen fluid has high deaminase activity (Warner, 1956). The extent to which dietary proteins are hydrolysed in the rumen depends on their solubility. (McDonald, 1954; McDonald and Hall, 1957).

When urea and other non-protein nitrogen materials replace part of the natural protein in diets for ruminants, ammonia is an important end-product of nitrogen metabolism in the rumen. The actual concentration of anmonia in rumen does however reflect a complex dynamic situation.

Ammonia is continually resynthesised into amino acids, which are subsequently incorporated into microbial protein. The energy and carbon required for these syntheses are derived from dietary carbohydrate fermentation products. To a limited extent free amino acids and peptides may be incorporated directly into microbial protein. (Wright and Hungate, 1967; Nolan and Leng, 1972).

The microbial protein synthesised in the rumen together with unchanged dietary material then passes to the abomasum and small intestine. Some ammonia may not be elaborated in the rumen. Mclaren, Anderson, Martin and Cooper (1961) and Hoshino, Saramaru and Morimoto (1966) have demonstrated that ammonia may be utilised by the rumen mucosa in synthesising L-glutamate. Hoshino et al. (1966) also observed the ability of rumen mucosa to synthesise and hydrolyse glutamine. They postulated that

glutamine serves as a storage form of ammonia in rumen mucosa. Small amounts of ammonia may be carried with the ruminal digesta to the abomasum and small intestine where absorption into the portal blood may occur. (McDonald 1948).

Quantitatively larger amounts of ammonia pass directly across the rumen wall to the blood. McDonald (1948) estimated that the quantity of ammonia N thus absorbed from the rumen may be 4 to 5 g per day in sheep. Later studies (Lewis, Hill and Annison, 1957) increased that estimate to as high as 14 g ammonia N in 24 hours. The extent to which ammonia absorption occurs in the rumen is influenced both by the concentration gradient (Lewis et al. 1957) and pH. (Bloomfield, Kearley, Creach and Muhrer, 1963). Ammonia is a weak base with the pka of about 8.80 to 9.15 (Bloomfield et al. 1963). An alkaline rumen pH causes the  $NH_{L}^{+}$  ion which is relatively tissue impermeable to be converted to the uncharged free ion which is rapidly absorbed. Ammonia entering the portal blood from the rumen and small intestine is carried to the liver where conversion to urea occurs. Lewis et al. (1957) reported that the liver was able to convert all the absorbed ammonia to urea until the concentration of ammonia in the portal blood reached a level of 0°8 mM/l, which corresponded to a rumen ammonia concentration of 55 to 60 mM/1. When the rumen ammonia concentration was increased from 60 to 100 mM/l a slow "leakage" of ammonia into peripheral circulation occurred indicating that both the capacity of the liver to convert ammonia to urea and the kidneys to excrete ammonia had been exceeded.

Urea entering the peripheral circulation is largely excreted in the urine. Endogenous urea also enters the rumen by salivary recirculation or by diffusion across the rumen wall, where it is again rapidly hydrolysed to ammonia. (Houpt, 1959; Somers, 1961a, b, c, d; Packet and Groves, 1965; Cocimano and Leng, 1967; Houpt

and Houpt, 1968). Diffusion through rumen epithelium seems to be the main route of entry. Elood urea may also diffuse into the abomasum and intestines, (Le Bars, 1967; Cocimano and Leng, 1967) where it may be hydrolysed by intestinal urease enzymes (Sidhu, Jones and Tillman, 1968) and the resultant ammonia absorbed by the tissues of the intestinal tract (McDonald, 1948).

Efficient utilisation of dietary non-protein nitrogen, particularly urea, is dependent on the maximum and rapid incorporation of runen ammonia into microbial protein. This will in turn limit the amount of ammonia available for absorption into the portal circulation and minimise a potentially toxic situation. The rate at which ammonia is produced in the runen should ideally closely parallel its rate of utilisation by runen micro-organisms. In the absence of rapid ammonia utilisation, toxicity may arise.

Symptoms of ammonia toxicity in ruminants include muscular twitching, excessive salivation, bloat, ataxia and laboured breathing followed by tetany, complete collapse and death (Armstrong and Trinder, 1966). Such signs generally occur within 30-60 minutes of feeding urea in excessive quantities. A single toxic oral dose of urea in solution is in the order of 0.45 g urea/kg liveweight which is equivalent to 31 g urea for an adult 70 kg ewe or 225 g urea for a mature 500 kg cow. Acute toxicity is generally reported to occur when peripheral blood ammonia concentrations exceed about 1000  $\mu$ g NH<sub>3</sub>-N/100 ml (eg. Austin, 1967; Chalupu, 1968). Such high values have been reported to occur when the runen ammonia concentrations exceed 80 mg NH<sub>3</sub>-N/100 ml (Lewis et al. 1957; Coombe and Tribe, 1958).

Factors which would seem to be of importance in reducing the risk of ammonia toxicity and which may perhaps increase the efficiency of utilisation of dietary nitrogen resultant upon urea feeding include;

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(a) Urea-containing feeds should be introduced gradually to the diet, over a period of two to three weeks, so that appropriate adaption of the rumen microflora may take place.

(b) Urea should preferably be presented to the rumen when active fermentation of carbohydrate material is possible. The presence of readily fermentable carbohydrate in the rumen at the time of urea ingestion ensures that carbon and energy essential for the rapid incorporation of ammonia into microbial protein is immediately available. Numerous experiments have shown that starch is more effective in promoting microbial protein synthesis and reducing the concentration of ammonia in the rumen than molasses or simple sugars (e.g. Mills, Booth, Bohstedt and Hart, 1942; Mills, Lardinois, Rupel and Hart, 1944; Bell, Gallup and Whitehair, 1953). In this context urea would seem to be well suited for use as a protein replacer in largely cereal-based diets.

(c) Urea is best accepted when fed frequently and in small amounts during a 24-hour period. There is wide appreciation of the value of urea in feedlot systems in America. The undoubted efficiency of urea as a nitrogen source relative to intact vegetable proteins in barley beef systems has frequently been demonstrated (e.g. Kay and Macdearmid, 1972). The fact that animals are allowed unlimited access to concentrate feeds in such systems would seem to be ideal, for it has been demonstrated (Campbell, Howe, Martz and Merilan, 1963) that frequency of feeding is an important factor from the view point of maximum urea nitrogen utilisation. In one experiment the live-weight gains of beef cattle fed supplementary nitrogen in the form of soya protein offered twice per day were 0'71 kg/day compared with 0.55 kg/day when urea was given twice per day. When the same total amounts of concentrate were offered in six feeds per day the daily

live-weight gain of the cattle fed soya protein remained virtually the same (0.75 kg/day), but that of the cattle receiving the urea supplement increased to 0.69 kg/day.

The fact that dairy cattle normally receive concentrates only twice per day may be of some significance in the appreciation of why urea feeding is not presently widely accepted for this class of livestock. Armstrong and Trinder (1966) from a review of the results of over 30 trials with lactating cows conducted between 1940 and 1965, where urea was generally added at rather more than 2 % to supply all of the additional protein required in the cereal concentrate fed for milk production, concluded that urea was less effective than vegetable protein. The overall mean yield of about 14 kg milk/day was reduced by about 0.8 kg/day when urea-containing diets were fed. Bartlett and Blaxter (1947) have shown in one experiment that the fall in potential milk yield resulting from the use of a concentrate mixture containing 2 % urea was greatest at high milk yields. High yielding dairy cows fed urea-containing concentrates naturally receive a greater proportion of their total daily intake of nitrogen in the form of The ingestion of comparatively large amounts of urea in urea. only two feeds per day, combined with the rapid solubility of urea in the rumen may result in a sharp rise in blood and rumen ammonia concentrations after feeding. This may be closely paralleled by both an increased blood urea concentration, and an elevated excretion of urea in the urine. Thus it would seem that the depression in milk yield often associated with feeding ureacontaining diets to dairy cows (particularly high yielding animals) may in part be attributable to the feeding practice adopted. A more sensible but perhaps impractical approach may be to feed such diets several times per day.

(d) An acid rumen pH favours the formation of the  $NH_4^+$  ion within the rumen and hence reduces both the amount, and rate at which free ammonia enters the portal circulation. The increased efficiency of utilisation of urea, often noted when it is fed in high cereal diets, may be associated with a generally lowered rumen pH (4.5 to 5.6) compared with a pH of 6.0 to 7.0 normally observed when a mixed diet of concentrate and roughage are given.

(e) A non-dietary factor which may be of significance in maintaining low concentrations of circulating ammonia in the blood is the efficiency of liver function. Parkins, Hemingway and Brown (1973) have demonstrated that when the liver function of sheep was progressively impaired by either copper sulphate consumption or administration of carbon tetrachloride the susceptability to ammonia toxicity resulting from the administration of urea increased. The elevated blood ammonia concentrations that were recorded were associated with progressive liver dysfunction. The possibility of pre-existing liver damage may be an important factor to take into account when assessing the potential toxic hazard of feeding urea-containing diets to ruminants.

# Methods of reducing the potential toxicity of dietary non-protein nitrogen.

The use of urea in practical farm diets for ruminants is increasing rapidly. When sensible feeding practices are adopted (i.e. with the gradual introduction of urea-containing diets in appropriate amounts) there is no appreciable risk of toxicity. However many feeding systems involve the use of materials containing much more than the 1.25 to 2.50 % urea needed to raise the crude protein (CP) content of cereals from about 10 % to 13.5 % or 17.5 % CP. For example, many protein/vitamin/mineral concentrate products on the market containing 34 % CP and intended to be mixed with about three to six times their own weight of cereals, may themselves have as much as 28 of the 34 % CP as urea and contain up to 10 % urea. Some special mineral/vitamin preparations may contain as much as 100 to 140 % CP based on the inclusion of 33 to 50 % urea and be intended for use at only about 2.5 % to 4.0 % in the concentrate part of the diet. Urea may also be included at about 10 % in urea/molasses liquid feeds. The risk of accidental ammonia toxicity occurring could increase when such products are used carelessly on the farm due to inadequate mixing or to over-consumption.

There have been many approaches (other than the adoption of sensible feeding practices) to reduce the potential toxicity of dietary urea, and to improve urea nitrogen utilisation. In this context, much emphasis has been directed towards slowing down the rate of dissolution of urea in the rumen. One method of approach has been the investigation of non-protein nitrogen compounds which are less soluble in the rumen than urea.

Biuret  $(H_2N-CO-NH-CO-NH_2)$  contains 39 % N and because of its relative insolubility in water (2 g/100 ml at 25°C) there is a reduced rate of release of ammonia in the rumen occurring after hydrolysis with the bacterial enzyme biuretase. Several workers have reported biuret to be as efficient a source of non-protein nitrogen for ruminants as urea (Gaither, Garrigus, Forbes and Hatfield, 1955; Meiske, van Arsdell, Luecke and Hoefer, 1955). It is generally accepted however that a considerable period (4 - 6 weeks) of adaption to biuret feeding is necessary before maximum utilisation occurs (Mclaren, Anderson, Welch, Campbell and Smith, 1959). The principal advantage of biuret over urea appears to lie in its markedly less potential toxicity relative to urea (Hatfield, Garrigus, Forbes, Neumann and Gaither, 1959). Sheep that were drenched with up to 1.37 g biuret/kg live weight (= 1.16 Urea/kg live weight) exhibited no abnormal reactions or symptoms of toxicity. About 0.45 g urea/kg liveweight is generally considered to be a dangerous amount. Furthermore, biuret is tasteless and there are no palatability problems.

Isobutylidene diurea (IB DU) (CH<sub>3</sub>)<sub>2</sub> CH.CH.(NH CO NH<sub>2</sub>)<sub>2</sub> a condensation product of urea and isobutyraldehyde is a nonhygroscopic, sparingly water-soluble material containing 32 % N. The slow release properties and negligible toxicity of IB DU relative to urea have been clearly demonstrated in the experiments of Parkins, Ritchie and Hemingway, (1971a). A single dose of 100 g IB DU (equivalent to 70 g of urea) administered as a drench to a 25 kg sheep produced no signs of toxicity in circumstances where about 15 g urea would normally have been fatal. Inclusion of IB DU in all cereal diets fed ad-libitum to growing lambs and calves resulted in fully satisfactory live-weight gains and food conversion ratios relative to soya bean meal (Parkins, Ritchie and Hemingway, 1971b; Hemingway, Parkins and Ritchie, 1972a). The value of IB DU as a replacement for the natural protein supplement in production diets for lactating dairy cows has still to be evaluated.

A wide range of other urea-derivatives have also been tested. In most experiments in vitro culture techniques have been used. Cellulose digestion and ammonia production were often the criteria for evaluation. Brent, Newland, Ullrey and Bradley (1966) compared in vitro the metabolism of urea, 1-3 diamethyl-urea, biuret, biurea, guanidine hydrochloride, guanyl-urea sulphate and thiocarbanalide. Only urea appeared to be hydrolysed to a useful degree. Simpson and Jones (1967) indicated that when urea derivatives, (acetylurea, methylurea, glycourea, biuret and others) were compared with crystalline urea, hydrolysis proceeded more slowly. Crystalline urea, methylurea and glycourea promoted the greatest increase in cellulose digestion. To date however none of these various materials seem to have reached commercial production.

Several attempts have been made to reduce the rate of ammonia release from usea in the rumen by coating it with water insoluble materials. Although coating usea prills with fat and waxy-type materials (Johnson, Bentley and Hershberger, 1962), clay-like materials and sodium proprionate, or ethyl cellulose (Ward and Cullison, 1970) have been claimed to reduce the rate of solution and dispersion of usea in the rumen, there have been no indications from feeding trials that these materials are any more efficiently utilised than untreated usea.

In America a gelatinized starch-urea product with slow release properties has been developed (Deyoe, Bartley, Pfost, Boren, Berry, Anstaett, Helmer, Stiles, Snug and Meyer, 1968). The gelatinized starch component of this material acts as a protective matrix to the contained urea, thereby reducing its rate of solubility in the rumen. At the same time it provides a readily available energy substrate for rumen micro-organisms. This product (Starea) is now commercially available in America as a

protein supplement for ruminants.

The production of ammonia in the rumen depends fundamentally on ruminal urease activity. Pearson and Smith (1943) have demonstrated using in-vitro techniques that rumen fluid has a high urease activity at all times. Consequently there have been several attempts to improve urea nitrogen utilisation by inhibiting ruminal urease activity. Tillman and Sidhu (1969) have reviewed the use of suitable chemicals for the inhibition of rumen ureolysis and proteolysis. Of the products tested only acetohydroxamic acid would seem to be of any practical use.

A more direct approach to decreasing rumen urease activity has been to produce circulating antibodies to urea (Harbers, Tillman, Visek and Glimp, 1965; Glimp and Tillman 1965; Sidhu, Jones and Tillman, 1968). Although in these studies immunised animals had reduced plasma ammonia concentrations following administration of urea, indicating that the ureolytic activity of the rumen fluid was decreased, blood urea concentrations remained unaltered (Sidhu et al. 1968) or even increased (Glimp and Tillman 1965). Any increase in performance noted in immunised animals would therefore seem to depend on the efficiency of the urea recycling process or factors governing the synthesis of bacterial protein from ammonia in the rumen.

Another alternative method of reducing urease activity has been to increase the acidity of the rumen at the time of urea ingestion, particularly over the first 1-2 hours. Perez, Warner and Loosli (1967), Kitchie, Parkins and Hemingway (1972) and Hemingway, Parkins and Kitchie (1972b) have described the effects of urea phosphate (a readily soluble material containing 17 % N and 19 % P) relative to urea on the concentrations of ammonia in the blood of calves and sheep. Administration of urea (0.3 g/kg live weight) in solution by oral drench increased the concentration of ammonia in blood one hour later to 230  $\mu$ g/100 ml

compared to about 10  $\mu$ g/100 ml when urea phosphate supplying the same amount of total nitrogen was given to over-night starved sheep (Ritchie et al. 1972). That this effect was due to a temporary reduction in rumen pH was shown by reproducing the effect by adding equivalent amounts of either phosphoric acid or sulphuric acid to the rumen with the urea. It was subsequently demonstrated (Hemingway et al. 1972b) that the acidity of urea phosphate was also adequate to significantly reduce the tendency of further added urea to increase blood ammonia concentrations.

Various ammonium phosphates have also been considered as alternatives to urea partly because of the possibility that the phosphate moiety of such materials might exert some buffering effect on rumen pH. Russel, Hale and Hubert (1962) recorded a lower maximum blood ammonia concentration when diammonium (DAP) was administered to lambs, than when equivalent amounts of nitrogen were given in the form of urea. Administration of up to 2.42 g DAP/kg liveweight (equivalent to 1.10 g urea/kg liveweight) produced no visible signs of toxicity. Under comparable conditions 0.44 g urea/kg liveweight proved fatally toxic. The reduced toxicity of DAP relative to urea was attributed to the smaller increase in rumen pH which occurred after the administration of DAP. It is generally accepted however that DAP is a less effective nitrogen source for ruminants than urea (Oltjen, Waller, Nelson and Tillman, 1963). A principal disadvantage of including non-protein nitrogen materials such as di- and triammonium phosphates in diets for ruminants is that these materials may release ammonia when in contact with damp feed troughs and ruminant saliva, resulting in a reduction in food intake (Oltjen et al. 1963; Reaves, Bush and Stout, 1966). The unpalatable nature of DAP-containing diets may be overcome by using monoammonium phosphate (10 % N, 23 % P) or a mixture of mono-ammonium

phosphate and diammonium phosphate (Reaves et al. 1963). The use of mono-ammonium phosphate as a source of supplementary non-protein nitrogen is somewhat restricted because of its low nitrogen content.

Repp, Hale, Cheng and Burroughs(1955) have determined the toxicity of the ammonium salts of organic acids. When compared on an isonitrogenous basis with urea only ammonium succinate proved to be of low toxicity. In comparative feeding trials with lambs where ammonium acetate, ammonium propionate, ammonium formate or urea replaced 50 % of the protein nitrogen of the ration, all forms of non-protein nitrogen supported growth equally well. Growth rates were however lower than those obtained on the completely vegetable protein diet. (Repp, Hale and Burroughs, 1955). Although ammonium salts of organic acids would seem to be effective sources of non-protein nitrogen they hold no special merit over urea as possible alternative non-protein nitrogen sources as they are of equally potential toxicity.

Included in the numerous sources of non-protein nitrogen investigated are ammoniated products, especially molassed sugar beet pulp materials. Broster, Balch, Bartlett and Campling (1960) reported that the treatment of sugar beet pulp with ammonia to increase the crude protein concentration from 10.4 to 20.4 % was almost as effective as decorticated groundnut cake in increasing the yields and composition of milk. Similarly Chomysyn, Bielinski and Slabon (1962) concluded that two-thirds of the milk production concentrate of dairy cows may be replaced by ammoniated or urea-containing sugar beet pulp without loss of milk yield or reduction in milk fat. Provided the ammonia treatment is such that the total nitrogen content of the beet pulp does not exceed 6 % (i.e. about 37 % CP) there are nc

palatability problems. (Millar, 1941).

Ammoniated sugar beet pulp would seem to be a promising source of non-protein nitrogen and warrant further investigation.

Many of the non-protein nitrogen materials discussed in the preceding paragraphs have additional and often complicated manufacturing processes and necessarily their cost per unit of nitrogen exceeds that of urea.

At the present time the commercial availability of such materials as IB DU and urea phosphate in Great Britain is very limited. Nevertheless these materials are still considerably cheaper than oil seed residue meals. In Great Britain the amount of crude protein in a compound feedingstuff is defined as the total nitrogen content multiplied by 6.25. The amount of the crude protein equivalent present as urea may be included in the total crude protein figure but the amount present as urea must be declared separately. Nitrogen present as ammonium or nitrate containing materials may specifically not be included as crude protein. (Fertiliser and Feedingstuffs Regulations, 1973.)

Under this legislation it would appear, at least for the present, that materials such as IB DU, biuret and urea phosphate (which are essentially very similar to urea) because they are not named specifically in the legislation can be equated with crude protein and may be declared as % N x 6.25 without any other disclosure. There could thus be some incentive for their practical utilisation in diets, if only on a short-term basis.

#### The Scope of the Present Experimental Investigations.

The experimental programme described in this Thesis has been concerned essentially with investigations concerning the supplementation of dried molassed sugar beet pulp with nonprotein nitrogen and appropriate minerals and vitamins.

Some 500,000 tonnes of molassed sugar beet pulp are available in Great Britain each year for livestock feeding and an increasingly large proportion of this output is now cubed (normally 1.27 cm diameter x2.5 cm length and some lesser production is of 0.8 cm diameter material).

Molassed sugar beet pulp as ordinarily produced has the following approximate composition; 40 % molasses, 10.0 % crude protein, 12.5 % crude fibre and 58 % soluble carbohydrate. The crude protein is about 60 % digestible but some 90 % of both the crude fibre and soluble carbohydrate are digested by ruminants. The metabolisable energy value (Ministry of Agriculture, Fisheries and Food. A.D.A.S. Advisory Paper No.11, 1971) is generally taken as 2.59 Mcal/kg. . The approximate mineral composition is; 0.8 % Ca, 0.08 % P, 0.4 % Na, 0.4 % Cl, 0.6 ppm Co, 15 ppm Cu, and 50 ppm Mn. Molassed sugar beet pulp is generally used as an energy source at about 1.5 to 2.0 kg per day for adult and growing cattle. Compared with feeding cereals for an equivalent energy intake it supplies appreciably more calcium but markedly less phosphorus. Appropriate additions of non-protein nitrogen and phosphorus (in particular) to molassed sugar beet pulp may produce a more nutritionally valuable product. Parkins (1972) has described the formation of a molassed sugar beet cube supplemented with 2.8 %urea, 3.0 % dicalcium phosphate, trace minerals, and Vitamins A and D giving a product containing about 17 % crude protein and O•7 % P which would be suitable for a wide range of nutritional applications e.g. to be fed for milk production at 1.0 kg per 2.5 kg milk, or

to be mixed with an equal weight of cereal to give a product with about 13.5~% crude protein for other growing and adult stock.

The procedure adopted by the British Sugar Corporation Ltd. for the production of this product (Triple Nuts) uses normally available raw materials and existing large scale production processes. After the extraction of the beet juice in large continuous diffusers, the residual shredded beet pulp is pressed in twin screw presses which yield the pressed pulp at about 22 % dry substance. The conventional procedure provides for continuous addition of molasses to the pressed pulp and the mixture is then dried in direct fixed rotary driers to produce molassed dried pulp in loose form. In some factories this material is then cubed.

In the plant for production of Triple Nuts, urea is added continuously to the molasses stream in a proportioning mixer. The urea dissolves readily in the molasses and this addition takes place in the same plant as is used at other times during the processing season for adding insoluble magnesite in the production of molassed sugar beet pulp with added magnesite. The automatic control of the proportioning between molasses and urea is adjusted so that the final cubed product contains about 2.8 % urea.

The urea-containing molasses is mixed with the pressed pulp and dried as in the conventional process and then the dicalcium phosphate, trace minerals and vitamins are added as a powder to the dried molassed pulp in a proportioning mixer and this product is then fed to the cubing machines. The proportioning between the dried molassed pulp and the phosphate/mineral/vitamin mixture is adjusted so that the final product contains up to 0.7 % P together with the desired content of trace minerals and vitamins A and D. A typical analysis of this product is as follows; 1.7 % CP, 1.25 % Ca, 0.55 % P, 0.3 % Mg, 1.0 % Na Cl, 40 ppm Fe, 15 ppm Cu,

2 ppm Co, 60 ppm Zn, 13 ppm I, 120 ppm Mn, and with 20 m i.u.s. Vitamin A and 5 m i.u.s. Vitamin D added per tonne.

An account of the successful results obtained in a large number of nutritional trials involving the feeding of Triple Nuts in quantity to lactating cows, pregnant and lactating ewes and rapidly growing beef cattle has been published by Hemingway and Parkins (1972). In this Thesis further nutritional studies have been conducted with Triple Nuts and with a comparable product containing between 7 and 11 % of added urea, which increased the total crude protein content of the molassed sugar beet pulp from 10 % to 30-40 %. It was intended that this material would be used with several times its own weight of cereals as a protein concentrate. This latter product was prepared by adding additional urea in aqueous solution to the existing mixture intended for the production of Triple Nuts just prior to passage into the cubing machines.

The inclusion of some 40 % molasses, and the hard physical nature of molassed sugar beet pulp cubes and the consequent time taken for them to absorb water and disintegrate in the rumen, would seem to make this an ideal material for the inclusion of urea. The high content of readily soluble carbohydrate and physical nature of this product might combine to make urea inclusion both a safe and at the same time a possibly more efficient nitrogen source to the ruminant and with a measure of slow release.

Section I of this Thesis is concerned with investigations into the possibility that the size and hardness of a molassed sugar beet pulp cube may reduce the rate of release of urea into the rumen.

Molassed sugar beet pulp is frequently fed as an energy supplement to ruminants receiving poor quality hay, straw or silage.

Poor quality roughages although well supplied with calcium. invariably contain inadequate amounts of phosphorus. An adult 500 kg dairy cow for example, requires for maintenance and the production of 10 kg of milk per day 0.41 % calcium and 0.38 % phosphorus in the dry matter. (Agricultural Research Council, 1965). Hemingway, MacPherson, Duthie and Brown (1968) have shown that 88 %of silages and 66 % of hays grown in Scotland fed with unsupplemented barley/vegetable protein mixtures would supply adequate amounts of dietary calcium. In marked contrast a very high proportion of both silages and hays (98 % and 99 % respectively) contain less than the recommended intake of 0.38 🖇 phosphorus. Inclusion of molassed sugar beet pulp (c.0.8 % Ca, c.0.08 % P) as an energy supplement in poor quality roughage diets for ruminants would tend to increase dietary calcium intakes as hays and silages generally contain about 0.4 % and 0.6 % Ca respectively. Phosphorus intakes however would not be improved as hays and silages commonly contain only about 0.1 - 0.2 % P.

Section 2 of this Thesis is concerned with the investigation of various compounds containing both nitrogen and phosphorus which could be used (with additional urea) as combined non-protein nitrogen and phosphorus supplements in molassed sugar beet pulp. The materials investigated include urea phosphate (16.5 % N, 19.5 % P), mono-ammonium phosphate (10.1 % N, 23.5 % P) and a granular ammonium polyphosphate (12.5 % N, 28.0 % P).

Molassed sugar beet pulp has been used as a carrier for calcined magnesite, a material used as a prophylactic agent for the prevention of hypomagnesaemic tetany in cattle. Calcined

magnesite is generally added at about 3.3 % to molassed sugar beet pulp so that the supplemented product when fed at about 1.5 to 2.0 kg/day would provide an additional dietary intake of about 30 to 35 g Mg/day. The high molasses content of sugar beet pulp would seem to be very effective in reducing the normally unpalatable nature of calcined magnesite. The Agricultural Research Council (1965) has recommended that a lactating Friesian cow requires 7.5 g Mg/day for maintenance plus 0.63 g Mg/kg milk produced. It was also recommended that these minimal allowances might be increased by 2.0 g/day "to take into account the variation in need from individual to individual." These quantities are based on the assumption that 20 % of dietary magnesium is available.

A lactating cow at spring grass yielding 30 kg milk/day would therefore require about 28 g Mg/day. Current recommendations for the prevention of clinical hypomagnesaemia are that 2 ozs of supplementary calcined magnesite (about 30 g Mg) should be fed per day. This in itself offers rather more magnesium than the ARC (1965) recommends, and is in addition to that which is already supplied in the herbage. Much smaller amounts of magnesium in the form of magnesium alloy 'bullets' supplying 2 - 4 g Mg/day have been shown by Ritchie and Hemingway (1968) to be effective in controlling clinical hypomagnesaemic tetany in dairy cows at spring grass. Whilst it is well recognised that some form of magnesium supplementation is necessary where cattle graze lush spring grass there seems to be lack of acceptance regarding the amounts of additional magnesium required. It is possible that the dietary availability of magnesium may vary considerably depending on the form of supplementation.

Magnesium supplementation may also be necessary where outwintered stock or housed animals are fed poor quality hay or silage

for maintenance purposes. Based on daily intakes of 9 kg of hay dry matter or 7 kg of silage dry matter, Hemingway et al. (1968) have shown that 20 % of a large number of both hays and silages grown in Scotland would supply less than the recommended (ARC, 1965) intake of 7.5 g Mg/day. If the desirable intake were increased to 9.5 g Mg/day, then 78 % of hays and 68 % of silages would be inadequate.

The general deficiency of phosphorus, but not necessarily calcium, in poor quality hays and silages fed in such situations has previously been described. Phosphorus supplementation would also be necessary where cattle graze lush spring and autumn grass.

Section 3 of this Thesis is concerned with the evaluation of two novel magnesium phosphates, a calcium magnesium phosphate (16.1 % Ca, 6.0 % Mg, 18.5 % P) and a magnesium phosphate (20.0 % Mg, 18.5 % P) which might be of use as combined magnesium and phosphorus supplements for both sheep and cattle.

Low-protein roughages such as cereal straws and poor quality hays are frequently used in low-cost maintenance diets for ruminants. Such materials are not readily eaten in quantity and are poorly digested by ruminants. One of the principal uses of urea is as a supplement to low-protein roughage feeds. Many experiments have shown that the addition of urea to low-protein roughage feeds markedly increases their voluntary consumption by ruminants. For example, Campling, Freer and Balch (1962) recorded that the mean intake of low-protein catstraw (c.3.0 % CP) fed ad libitum to adult cows was increased by up to 43 % when 25 - 150 g urea/day was given by continuous intraruminal infusion. There were also associated increases in the apparent digestibility of the crude

fibre and N-free extract fraction of the straw. The major role of urea in improving the nutritive value of low-protein roughage feeds would seem to be the provision in the rumen of ammonia in sufficient concentrations to promote active growth and multiplication of cellulolytic micro-organisms responsible for the digestion of roughage materials. Many poor-quality roughage feeds in addition to being deficient in energy and protein, contain inadequate (ARC, 1965) amounts of phosphorus. Hemingway et al. (1968) have recorded a significant correlation between the crude protein content and the concentration of phosphorus in many samples of hay and silages grown in Scotland. There is little published information on the quantitative effects of a low dietary intake of phosphorus on appetite. This may be of particular significance when mixed diets of cereal straw and/or poor quality hay (c.1.5 - 4.0% CP, c.0.1% P) and molassed sugar beet pulp (c.10.0 % CP, c.0.08 % P) are fed to ruminants for extended periods.

Section 4 of this Thesis is concerned with an assessment of the effect of supplementary urea and/or phosphorus contained in a molassed sugar beet cube on the voluntary intake of lowprotein oatstraw by British Friesian castrates (steers) and by pregnant beef cows. The effects of such additions on the digestibility of the roughage, the composition of the rumen liquor and the concentration of various blood constituents are also described. In one experiment where beef cows (in late pregnancy) were fed a low-protein hay and molassed sugar beet pulp variously supplemented with urea, the effects of the contrasting dictary treatments on the voluntary consumption of the hay and the  $\forall$ -globulin concentration of the colostrum were measured.

One of the main reasons for using urea in diets for ruminants is that in comparison with vegetable proteins it is a far cheaper source of supplementary nitrogen. For reasons described in a previous part of the general introduction to this Thesis urea can be used to particular advantage when included in mainly cereal-based diets and in situations where ruminants are allowed unrestricted access to concentrate feeds. Such ad libitum feeding systems are fairly common for fattening cattle and to a lesser extent sheep. Parkins (1972) has conducted a series of experiments in which a molassed sugar beet pulp cube containing 17 % crude protein (c.2.8 % urea) was used as a major component of diets for fattening cattle. When this material was mixed in equal parts with barley and given ad libitum to 3-month-old steers their growth rate was the same as when a barley/vegetable protein mixture with the same crude protein (13.5 %) was offered. However, inclusion of 50 % of the molassed sugar heet pulp product reduced the efficiency of food conversion by about 6 %.

Section 5 of this Thesis is concerned with the evaluation of a molassed sugar beet pulp product containing between 30 and 40 % crude protein as a protein/vitamin/mineral concentrate for rapidly growing cattle and sheep. It was intended that this material should be mixed with about 85% of cereals to give a final product containing about 13.5% crude protein.

There is considerable difference of opinion regarding the value of urea in diets for dairy cows. Some reduction in milk yield may occur when urea is fed, but this may be partially offset by a reduction in feed costs. One of the main practical problems associated with feeding urea to dairy cows seems to be the

acceptability of diets containing 2.0 to 2.5 % urea (i.e. where urea is used to supply all of the additional protein required in the cereal concentrate fed for milk production), particularly in early lactation when milk yields are in excess of 20 kg/day. At the present time, however, it is generally agreed that about 1.25 % urea can be satisfactorily included in concentrate mixtures for dairy cows, so that the crude protein contributed by urea does not exceed about one-third of the total crude protein in the concentrate. Molassed sugar beet pulp would appear to be a most suitable medium in which to feed urea to dairy cows because its high content of molasses (c.40 %) may overcome the unpalatable nature of urea. Recent evidence would also suggest that molassed sugar beet pulp may satisfactorily replace barley on an equal dry matter basis as an energy source in concentrate mixtures fed for milk production (Castle, 1972).

In section 6 of this Thesis an experiment designed to evaluate the effect of a molassed sugar beet pulp product containing 32 % crude protein (c.8 % urea) as a partial protein replacement in the production diet fed to lactating dairy cows is described.

#### Experimental Methods.

The experimental work described in this Thesis has involved a total of 18 separate experiments. Investigations have been undertaken with both cattle and sheep. All the experiments have been conducted according to standard and well recognised experimental procedures employing for example 4 x 4, replicate 4 x 4 and 5 x 5 Latin square designs. The randomised block type design has also been used. Information has been obtained in respect of the live-weight gain of growing cattle and sheep and the milk production of dairy cows. Many of the experiments have involved controlled feeding

over extended periods, the measurement of food intake and the quantitative collection of faeces and urine on a daily basis. The effect of various contrasting dietary treatments on the composition of the rumen liquor and the concentration of various blood parameters have also been determined. Necessarily, this has produced a large number (several thousands) of individual recordings and analyses. In the interest of conciseness and brevity only the mean data and associated standard errors appropriate to each experiment are presented in this Thesis. The whole collection of other individual, day-to-day, observations involving some nine hundred pages are lodged separately in the Animal Husbandry Department, Glasgow University Veterinary School.

In the presentation of the mean results from each experiment a standard statistical terminology has been adopted throughout and is as follows: \*=P < 0.05, \*\* = P < 0.01, \*\*\* = P < 0.001, N.S. = not significant, n.d. = value not determined.

The analyses of foodstuffs, faeces, urine, milk and rumen liquor were undertaken using standard analytical techniques. A brief outline of these methods is given in the Appendix.

#### SECTION 1

# THE EFFECTS OF THE SIZE AND HARDNESS OF UREA-CONTAINING MOLASSED SUGAR BEET PULP CUBES ON THE CONCENTRATIONS OF AMMONIA

# IN THE BLOOD AND RUMEN CONTENTS OF CATTLE.

# Introduction.

Molassed sugar beet pulp cubes as normally produced (c.1.27cm diameter, c.2.5cm length) are rather hard and take some time to soften and disintegrate when in contact with liquid media such as rumen fluid. Examination of the rumen contents of adult fistulated cows has shown that whole molassed sugar beet pulp cubes may still be present in the rumen 1 - 2 hours after being fed. It is presumed that a proportion of molassed sugar beet pulp cubes when fed to cattle, escape mastication and pass intact into the rumen.

Parkins (1972), has described the rate of release of urea from urea-containing molassed sugar beet pulp cubes (c.2.8% urea) suspended in water at room temperature. Materials of constant diameter (c.1.27cm) were sub-divided into three fractions; whole cubes (c.2.5cm length), halved cubes (c.1.3cm length) and quartered cubes (c.0.6cm length). A crumbled sample was prepared by pulling the cubed material apart to its original shredded form and fully ground material was also used. For the ground material all the urea appeared in solution after 0.25 hours and for the shredded material some 70% was in solution after 1 hour and 90% after 2 hours. In marked contrast, only 60% of the urea in the whole cube was dissolved after 3 hours and it took 6 hours for 90% to dissolve. Halved and quartered material gave intermediate results. In a subsequent experiment when whole or crumbled molassed sugar beet pulp materials containing 2.8% urea were given per rumen fistula to an adult cow, the concentration of ammonia produced in the rumen liquor one hour after administration of the

whole cubes was only about one-half of that recorded for the crumbled material. Accordingly, molassed sugar beet pulp cubes would seem to be a most suitable medium in which to include urea. The high molasses content (c.40%), and slow rate of disintegration of this material, appears to reduce the rate of solution of urea in the rumen. The urea inclusion is thus a safer and at the same time perhaps, a more efficient nitrogen source for ruminants.

Two factors which would seem to be of importance in determining the rate at which urea is released from molassed sugar beet pulp cubes are the size and hardness of the cubes. Both these factors may be of even greater significance when up to 11% of urea is included and where the final product is intended for use as a protein concentrate. Three of the experiments to be described have been concerned with assessing the effect of the size and hardness of a high urea molassed sugar beet pulp cube (containing 7.4% urea and with about 30% crude protein) on the blood and rumen ammonia concentrations of adult cattle. In the fourth experiment the effect of reducing the size of a whole molassed sugar beet pulp cube containing 2.7% urea, on the rate of solution of urea in the rumen has been determined.

#### Materials and Methods.

Two forms of high urea molassed sugar beet pulp cubes (SBPHU) were produced; hard cubes containing a total of about 7.2% of added urea and with 28.6% crude protein, and soft cubes containing a total of 7.6% added urea and with 31.1% crude protein. Both products were of normal production size (c.2.5cm length, c.1.27cm diameter).

The soft SBPHU cubes differed markedly from the hard SEPHU cubes in their physical characteristics. The soft SBPHU cubes were rather loosely formed. They contained a large number of transverse fissures on their outer surface in which considerable

amounts of crystalline urea could be seen. These soft SEPHU cubes were readily broken apart by hand. In contrast, the hard SEPHU cubes had a smoother exterior surface, could not be broken by hand and took much longer to soften and disintegrate in water and rumen fluid.

For experimental purposes the hard and soft SBPHU materials were each separated by hand into three fractions; whole cubes (c.2.5cm length, c.1.27cm diameter), halved cubes (c.1.3cm length, c.1.27 diameter) and quartered cubes (c.0.6cm length, c.1.27cm diameter). The various high urea molassed sugar beet pulp products were compared with a barley-based cube (c.1.5cm length, c.0.5cm diameter) which contained 9.4% of added urea and with 38.6% crude protein.

The three experiments conducted with high urea molassed sugar beet pulp cubes (Experiments 1.1, 1.2 and 1.3) each followed the same general pattern. Four adult non-lactating cows (mean liveweight 535kg) each fitted with a rumen fistula (Avon Rubber Co. Ltd.) were maintained on hay (8-9kg/day) and 1.5kg/day of normal production molassed sugar beet pulp cubes (8.8% crude protein). Hay and unsupplemented molassed sugar beet pulp were given at 8.00 hours and hay alone was fed at 16.00 hours. In each experiment there were four treatments which involved giving the three different sized fractions of high urea molassed sugar beet pulp cubes and the high urea barley cubes in a  $4 \times 4$  Latin Square design. In all three experiments each of the four cows received one of the four treatments in appropriate sequence at 7-day intervals at 9.00 hours. No hay was given at the evening feed prior to the day on which the experimental materials were administered.

Each product was given to the appropriate cow in amounts calculated to supply 0.2g urea/kg liveweight. In practice this

amounted to about 1.4kg of each product, thereby supplying about 100g of urea on each occasion. In Experiments 1.1 (hard cubes) and 1.2 (soft cubes) the materials were given per rumen fistula. In Experiment 1.3 the experimental products were fed to the cows and each animal was allowed 0.5 hours to consume the products. Thereafter, remaining food residues were collected and the actual fresh weight of material consumed recorded.

Rumen liquor samples were obtained just prior to and 0.5, 1, 2 and 4 hours after administration of the products. Blood samples were taken at the same times after administration but not beforehand. Analyses were undertaken to determine blood and rumen ammonia and blood urea concentrations.

On each occasion that rumen liquor samples were obtained, the rumen digesta was carefully examined and the physical appearance and rate of disintegration of the experimental products noted.

Experiments 1.1, 1.2 and 1.3 were conducted in sequence so that the whole period involved was 12 weeks.

In Experiment 1.1, the four treatments given per rumen fistula were; (A) whole hard SBPHU cubes (B) halved hard SBPHU cubes (C) quartered hard SBPHU cubes and (D) whole barley-urea cubes.

The four treatments given per rumen fistula in Experiment 1.2, were as for Experiment 1.1 except that the soft SBPHU cubes were used for treatments (A), (B) and (C).

In Experiments 1.1 and 1.2 the hard and soft SEPHU treatments were administered per rumen fistula so that the relative rates of release of urea into the rumen could be assessed without the possible complication that the cows might fracture the materials in part by mastication. To be more in accord with practical conditions, in Experiment 1.3 all the treatments were fed to the cows. The four treatments were; (A) whole hard SBPHU cubes, (B) whole soft SBPHU cubes, (C) quartered hard SBPHU cubes and (D) whole barley-urea cubes.

## Results.

# Experiment 1.1 (Table 1.)

There were obvious differences in the manner`in which the various hard SBPHU fractions disintegrated in the rumen. Whole hard cubes could still be found in samples of rumen digesta 1-2 hours after being administered. The rumen liquor in most instances had only penetrated a few millimetres below the surface of the cubes leaving a hard, dry, central core. The halved, hard cubes had a similar appearance after 0.5 hours in the rumen, but were completely dispersed within 1 hour. In contrast, the quartered hard SBPHU cubes and the barley-urea cubes disintegrated rapidly and within 0.25 to 0.5 hour had completely dispersed.

The mean concentrations of ammonia in the rumen liquor and the blood were generally associated with the rates at which the various hard SEPHU fractions and barley-urea cubes disintegrated in the rumen. The maximum rumen ammonia concentrations for all four products was generally attained about 2 hours after administration. Whole hard SEPHU cubes gave a significantly (P < 0.05) lower concentration of ammonia in the rumen liquor (43.5mg/100ml) than either the quartered hard cubes (64.1mg/100ml)or the barley-urea product (78.1mg/100ml) after 2 hours. Four hours after administration the rumen ammonia concentration for the whole, halved and quartered SEPHU materials had declined to 24.5, 33.9 and 36.0mg/100ml respectively. In comparison after 4 hours the mean rumen ammonia concentration recorded for the barley-urea cube was still as high as 50.9 mg/100ml.

The most marked differences were seen in respect of blood ammonia concentrations. The barley-urea cube produced blood Table 1. Experiment 1.1 Mean concentration of ammonia in the rumen liquor (mg/100ml) and blood (µg/100ml) and the increase in blood urea concentration (mg/100ml) of cows following the administration per rumen fistula of hard high-urea molassed sugar beet pulp cubes to supply 0.2g urea/kg liveweight.

TTAGMG	arguro.					
	Tir	ne after	r administ	ration (1	nours)	
Rumen NH3-N	0	0•5	1•0	2•0	4.0	
A Whole	3•0	26•7	41•8	43•5	24•5	• •
B Halved	2•7	34•6	53•1	61•0	33•9	
C Quartered	3•8	45•1	66•1	64•1	36•0	
D Barley	3•7	38•4	61•0	78•1	50+9	
S.E. of Mean $(+)$	0•45	4•10	7•65	5•58	2•81	
Significance	N.S.	N.S.	N.S.	D>A##	D>A,B,C*	¥
				C>A*	C>A*	
Blood NH3-N						
A Whole	n.d.	27•8	31•5	107•0	72•3	
B Halved	n.d.	38•5	81•0	115•0	42•0	
C Quartered	n.d.	70•3	94•0	140•0	84•3	
D Barley	n.d.	151•8	224•0	248•0	165•8	
S.E. of $Mean(\pm)$	_	<u>3</u> 2•08	12•88	16•02	23+38	
Significance	-	N.S.	D>A,B,C*∺+	D>A,B,C**	* D>A,B,C*	÷
			B <b>,</b> C>A*			
Blood Urea						Increase over 3.5 hours
A Whole	n.d.	9•2	10•4	12•8	17•5	8•3
B Halved	n.d.	9•4	10•9	13•8	18•6	9•2
C Quartered	n.d.	10•7	14•1	18.1	23•8	13•1
D Barley	n.d.	10.9	13•6	17•8	24•3	13•4
S.E. of Mean(±)		***	-	~	***	0.56
Significance	len.	-	-	· ·	495	C,D>A,B**

ammonia concentrations which were at least double those recorded for the halved or quartered SBPHU materials and about seven times as large as those for the whole hard SBPHU cube after 1 hour. By 2 hours after administration the barley-urea cube still showed a mean blood ammonia concentration (248µg/100ml) which was almost double that of any of the three hard SBPHU fractions. Marked differences in the blood ammonia concentrations produced by the barley-urea cube and the various hard SBPHU materials were still apparent after four hours.

In respect of blood urea concentrations all four products produced broadly linear increases over the 3.5 hours period during which blood samples were taken. At any one time after administration of the products the blood urea concentrations given by the whole hard SEPHU cubes were comparable with those of the halved hard SEPHU cubes and those of the quartered hard SEPHU cubes were closely similar to the blood urea concentrations produced by the barley-urea product. Both the whole hard SEPHU cubes and the halved hard SEPHU cubes gave rise to significantly (P < 0.01) lower increases in blood urea concentration than did either the quartered hard SEPHU cubes or the barley-urea material.

#### Experiment 1.2 (Table 2.)

In marked contrast to the hard SEPHU materials described in Experiment 1.1, all three soft SEPHU fractions (whole, halved and quartered) disintegrated rapidly in the rumen and within 0.25 to 0.5 hour were completely dispersed. The barley-urea cube which acted as a control treatment between Experiments 1.1 and 1.2, again completely disintegrated within 0.5 of an hour.

The soft SBPHU cubes generally gave rise to much higher concentrations of ammonia in the rumen liquor than did the hard SBPHU cubes (Experiment 1.1) especially after 1 hour. Subdivision of the soft SBPHU cubes did not affect rumen ammonia

Table 2. Experiment 1.2 Mean concentration of ammonia in the rumen liquor (mg/100ml) and blood (µg/100ml) and the increase in blood urea concentration (mg/100ml) of cows following the administration per rumen fistula of soft high-urea molassed sugar beet pulp cubes to supply 0.2g urea/kg liveweight.

Time after administration (hours)

Rumen NH3-N	0	0•5	1.0	2•0	4.0	
A Whole	2•9	57•9	71•8	67•3	41•9	
B Halved	4•2	51•7	71•5	65•7	40•3	
C Quartered	4•1	59•7	82•2	95+3	50•4	
D Barley	6•2	46•5	72•0	80•6	64•5	
S.E. of Mean( $\pm$ )	<b>1</b> *01	5•73	5•41	8•29	7•95	
Significance	N.S.	N.S.	N.S.	C>B*	N.S.	
Blood NH3-N						
A Whole	n.d.	90•0	164•0	131•3	91•0	
B Halved	n.d.	90•0	132•0	124•8	81•0	
C Quartered	n.d.	98•8	177•8	218•5	149•5	
D Barley	n.d.	135•0	185•8	213•0	175•3	
S.E. of Mean( $\pm$ )	-	30•28	32•46	22•77	17•99	
Significance	-	N.S.	N.S.	C,D>A,B*	D>A,B	¥•
. ,					C>B*	
Blood Urea						Increase over 3.5 hours
A Whole	n.d.	11•5	13•1	16•1	22•6	11•1
B Halved	n.d.	12•4	13•1	16•9	23•9	11•5
C Quartered	n.d.	13.1	14•6	17•9	23•7	10•6
D Barley	n.d.	13•9	15•7	19•3	26•7	12•8
S.E. of Mean( $\pm$ )	-	-	-	-	-	0.81

Significance

N.S.

concentrations after 1 hour but after 2 hours the quartered soft SEPHU cubes produced significantly (P<0.05) higher concentrations of ammonia in the rumen liquor than was the case for the halved soft SEPHU cubes. The difference between the concentration of ammonia produced in the rumen liquor by the quartered soft SEPHU cubes and the whole soft SEPHU cubes after 2 hours failed marginally to be significant.

All three soft SEPHU fractions produced blood ammonia concentrations which did not differ from the barley-urea product at both 0.5 and 1 hour after administration. After 2 hours, the mean blood ammonia concentration for both the whole and halved SEPHU cubes (132 and 125µg/100ml respectively) was significantly less than the mean blood ammonia concentration recorded for either the quartered soft SEPHU cube (219µg/100ml) or the barley-urea product (213µg/100ml). After 4 hours, the blood ammonia concentrations for both the barley-urea product and the quartered soft SEPHU cube were still almost twice as high as those for the whole and halved soft SEPHU fractions.

There were no significant differences between the various grades of soft SBPHU cubes and the barley-urea product in respect of the increase in blood urea concentration over 3.5 hours.

# Experiment 1.3 (Table 3.)

Considerable difficulties were experienced in the feeding of the four experimental products, particularly the barley-urea cube. One cow (of the four) on three separate occasions refused to eat this product. The three high-urea molassed sugar beet pulp materials were more readily consumed although not always completely. Residues which were left (8 to 9% of the total fresh weight) were usually small.

The appearance and rate of disintegration of various SBPHU materials and barley-urea product was similar to that recorded Table 3. Experiment 1.3 Mean concentration of ammonia in the rumen liquor (mg/100ml) and blood (µg/100ml) and the increase in blood urea concentration (mg/100ml) of cows following the consumption of various high-urea molassed sugar beet pulp cubes to supply 0.2g urea/kg liveweight.

Time after feeding (hours)

 $\mathcal{I}\mathcal{U}$ 

Rumen NH3-N	0	0•5	1•0	2•0	4•0	
A Whole Hard	3•2	44•9	52+2	43•5	21•1	
B Whole Soft	4•1	58•6	63•9	49•0	27•9	
C Quartered Hard	4.1	40•3	48•4	41•6	22•9	
D Barley	3•7	59•1	72•9	66•3	44•6	
S.E. of Mean( $\pm$ )	4•39	4•73	5•57	3•15	3•69	
Significance	N.S.	N.S.	D>A,C*	D>A,B,C	** D>B,*	A,C***
Blood NH3-N						
A Whole Hard	n.d.	62•4	117•0	153•8	114•6	
B Whole Soft	n.d.	137•5	248•0	225•3	127•1	
C Quartered Hard	n.d.	77•4	191•3	230•3	93•1	·
D Barley	n.d.	227•5	271•8	282•8	212 <b>•3</b>	
S.E. of $Mean(\frac{+}{-})$	-	42•79	51•95	<b>59•2</b> 8	32•91	
Significance	-	N.S.	N.S.	N.S.	N.S.	
Blood Urea						Increase over 4 hours
A Whole Hard	n.d.	14•3	15•4	19•3	24•3	10•0
B Whole Soft	n.d.	17•3	20•4	24•0	30•4	13•1
C Quartered Hard	n.d.	13•9	16•4	20•6	27•3	13•4
D Barley	n.d.	16•7	20•4	24•0	30•5	13•8
S.E. of Mean(+)	-	-	-	·_	-	1.14
Significance	-	-	-	-	-	N.S.

when the same materials were administered per rumen fistula. Whole soft SBPHU cubes and the barley-urea product were completely dispersed within 0.5 hour following consumption. Indeed, on one occasion when samples of rumen digesta were examined 10 minutes after the cows had been fed, no trace could be found of intact whole soft SBPHU cubes. The barley-urea cube and the quartered hard SBPHU cube were at this stage present almost entirely in their original form. It is assumed that little breakdown of these two materials took place in the mouth. the cubes being swallowed predominately in their original form. The quartered, hard SBPHU cubes generally took longer to disperse (0.5 to 1 hour) than did either the barley-urea cube or the whole soft SBPHU cube. In contrast, whole hard SBPHU cubes could still be found in the rumen 1 hour after consumption but a considerable amount of disintegrated material was also present. This is perhaps consistent with the fact that some at least of the whole hard SBPHU product was broken down in the mouth to material equivalent in size to quartered and halved hard SBPHU cubes.

For the one cow which would not eat the barley-urea cube the mean blood and rumen ammonia data recorded when this animal was given the barley-urea cube per rumen fistula in Experiment 1.1 and 1.2 has been utilised rather than a calculated "missing value."

One hour after administration of the barley-urea cubes gave a concentration of ammonia nitrogen in the rumen liquor of 72.9mg/100ml which was significantly (P<0.05) higher than for either the whole or quartered hard SBPHU cubes. After 2 hours the ammonia concentration in the rumen liquor for the barley-urea product was significantly higher (P<0.01) than for any of the SBPHU products.

Marked differences were recorded in the mean concentration

of ammonia nitrogen in the blood of the cows both 1 and 2 hours after feeding. The lowest concentrations (117-154µg NH<sub>3</sub>-N/100ml) resulted from the feeding of the whole hard SBPHU cube. In contrast, the mean blood ammonia concentration for whole soft SEPHU cube ranged from 248 to 225µg/100ml, and that of the barleyurea cube from 272 to 283µg/100ml. There was however, a very large standard error of 52-59µg/100ml associated with the mean blood ammonia concentrations after 1 and 2 hours. This was much greater than the error of 12 to 32µg/100ml obtained when these various materials were administered per rumen fistula. In consequence, none of the differences in respect of blood ammonia concentrations after 1 and 2 hours proved to be significant, although there was an obvious trend for the highest concentrations to be associated with the feeding of the barley-urea cube and the whole soft SBPHU cube which also produced the highest concentrations of ammonia nitrogen in the rumen fluid.

There were no significant differences in the concentrations of urea in the blood but the smallest increase was found when the whole hard SBPHU cube was fed.

#### Experiment 1.4

The effect of reducing the size of a urea-containing molassed sugar beet pulp cube on the rate of release of urea in the rumen.

#### Materials and Methods.

The three experiments previously described (Experiments 1.1, 1.2 and 1.3) have shown that both the length and hardness of a high urea (c.74% urea) molassed sugar beet pulp cube of constant diameter (c.1.27cm) had an important influence on the rate at which the contained urea appeared in solution in the rumen and subsequently on the blood ammonia concentration. After the completion of this particular series of experiments a further

experiment was conducted with a smaller diameter cube.

Two materials containing 2.7 % of added urea and with 15.4 % crude protein were prepared; (A) whole molassed sugar beet pulp cubes (SBPU; c.2.5 cm length, c.0.8 cm diameter) of a hardness intermediate between the materials used in Experiments 1.1, 1.2 and 1.3 and (B) a ground product prepared by grinding these whole SBPU cubes through a 3 mm screen using a laboratory hammer mill.

Four adult fistulated cows (mean liveweight 551 kg) were used in the experiment. Two of the cows were fed the whole SBPU cubes and two the ground material. The treatment sequence was reversed on three occasions at weekly intervals, so that a total of six separate investigations per treatment were made.

During the one week interval between each experimental feeding period the cows were maintained on a diet of 8-9 kg/day of hay together with about 2.0 kg of unsupplemented 2.5 cm length x 0.8 cm diameter molassed sugar beet pulp cubes (9.56 % crude protein). Hay and unsupplemented molassed sugar beet pulp were given at 8.00 hours and hay alone was fed at 16.00 hours. No food was given at the evening feed or on the morning prior to the administration of the experimental materials.

Each material containing 100 g of urea (i.e. 3.76 kg) was fed to the appropriate cow on each occasion. Each cow was allowed 0.25 hour to consume the products, thereafter any remaining food residues were collected and administered per rumen fistula.

Rumen liquor samples were obtained just prior to and 1 and 2 hours after consumption of the products. Blood samples were taken at the same times after feeding but not beforehand. Analyses were undertaken to determine blood and rumen ammonia and blood urea concentrations.

#### Results (Table 4.)

The small SBPU cubes were readily consumed by each of the four cows. The ground material was less palatable. Two of the four

<u>Table 4.</u> Experiment 1.4 Mean concentration of ammonia in the rumen liquor (mg/100ml) and blood (µg/100ml) and the increase in blood urea concentration (mg/100ml) of cows following the consumption of whole (2.5cm length× 0.8cm diameter) and ground molassed sugar beet pulp cubes to supply 100g of urea.

	Time after	administration	(hours)	
Rumen NH <sub>3</sub> -N	0	1	2	
A Whole	4•8	49•0	37•9	
B Ground	3•7	52•7	36•9	
S.E. of Mean( <sup>+</sup> )	0•32	4•44	3•83	
Significance	N.S.	N.S.	N.S.	
Blood NH3-N				
A Whole	n.d.	250•5	219•5	· ·
B Ground	n.d.	198•5	179•5	
S.E. of Mean(±)	-	44•01	22•56	
Significance	-	N.S.	N.S.	
Blood Urea				Increase over
				1 hour
Λ Whole	n.d.	15•2	19•0	3•8
B Ground	n.d.	14•7	17•0	2•3
S.E. of Mean( $\pm$ )	-	-	-	0.67

Significance

40

N.S.

cows left 10 to 15 % of the total fresh weight of ground material on two occasions. Food residues were however re-administered to the appropriate cow per rumen fistula within 15 to 20 minutes of being fed.

Both the whole SEPU product and the ground material produced a concentration of ammonia in the rumen liquor of about 50 mg/100 ml 1 hour after being fed. After 2 hours the rumen ammonia concentration for both products had fallen to about 37 mg/100 ml.

The mean blood ammonia concentration resulting from feeding the whole SBPU cube ranged from 251 to 220  $\mu$ g/100 ml 1 to 2 hours after feeding and that of the ground material from 199 to 180  $\mu$ g/100 ml. There were no significant differences between the two treatments in respect of either blood ammonia or blood urea concentrations.

It is concluded that the inclusion of urea in a smaller sized sugar beet cube (c.2.5 cm length, c.0.8 cm diameter) was not effective in reducing the rate of release of urea. <u>Discussion and Conclusions (Experiments 1.1, 1.2, 1.3 and 1.4)</u>

The results of this series of experiments indicate that the rate of release of urea from a molassed sugar beet pulp cube is related both to the size and physical characteristics of the cube. A cube which is rather soft and loosely formed is readily broken down in the mouth, and when in contact with liquid media in the rumen rapidly disperses into its original shredded form. A more ideal product, which affords a measure of slow release, is a hard, compact cube. The hard SEPHU cubes used in Experiments 1.1, and 1.3 had a smooth outer surface which contained very few cracks or fissures. Both the hardness and the nature of the external surface of the cube reduce the rate at which rumen liquor penetrates the interior of the cube, with the result that the structure of the cube is maintained for a much longer time in the rumen. The fact that whole hard SEPHU cubes are only partially

disintegrated in the mouth and that a proportion may pass intact into the rumen would seem to be of importance in delaying their rate of dispersion.

In all four experiments the greatest differences in respect of the concentrations of ammonia produced in the rumen and in the blood were recorded 1 hour after the administration per rumen fistula or following consumption of the various whole SBPHU and SBPU materials and the barley-urea cube. Table 5 summarises these results. The effectiveness of the whole hard SBPHU cube compared with the whole soft SBPHU cube in reducing the rate of solution of urea in the rumen is clearly apparent when direct comparisons are made between Experiments 1.1 and 1.2. One hour after administration per rumen fistula of the whole soft SBPHU product the concentration of ammonia nitrogen in the rumen liquor was about 72mg NH<sub>3</sub>N/100ml, compared with a concentration of 42mg NH3-N/100ml when the whole hard SBPHU product was given. The effects of the two contrasting treatments on the concentrations of ammonia in the blood after 1 hour were even more striking, the whole soft SBPHU material producing a blood ammonia concentration which was about five times as great as that recorded when the whole hard SEPHU product was given. Similar differences in rumen and blood ammonia concentration (but of a lesser degree) were also apparent when the soft and hard SBPHU products were fed by mouth (Experiment 1.3). The slightly elevated rumen ammonia concentration recorded 1 hour after feeding the hard SBPHU product compared with the concentration recorded when the same product was administered per rumen fistula is perhaps consistent with the fact that some of this product was broken down in the mouth, and in consequence there was a more rapid rate of solution of the contained urea in the rumen. There would seem to be little benefit, in terms of a slow release of urea into the

Table 5. The mean concentrations of ammonia in the rumen liquor (mg/100ml) and the blood (µg/100ml) 1 hour after the administration per rumen fistula or consumption of whole hard SEPHU cubes, whole soft SEPHU cubes, barleyurea cubes or whole SEPU cubes in Experiments 1.1, 1.2, 1.3 and 1.4.

Product	Whole hard	Whole soft	Barley	Whole
	SBPHU	SBPHU	urea	SBPU
Rumen NH3-N				
Experiment 1.1	41.8	-	61•0	-
Experiment 1.2	-	71•8	72•0	-
Experiment 1.3	52•2	63•9	72•9	-
Experiment 1.4	-	-	-	49•0
Blood NH3-N				
Experiment 1.1	31•5	-	224•0	-
Experiment 1.2	-	164.0	185•8	-
Experiment 1.3	117•0	248+0	271.8	-
Experiment 1.4		-	-	250•5

rumen when the size (both the length and diameter) of a urea containing molassed sugar beet pulp cube is reduced. In Experiment 1.4 for example, where the small (c.2.5cm length, c.0.8cm diameter) SBPU cubes were fed, a blood ammonia concentration of 251µg/100ml was recorded after 1 hour and was closely similar to the concentration of 248µg/100ml produced 1 hour after the consumption of whole soft SEPHU cubes (c.2.5cm length, c.1.27cm diameter) in Experiment 1.3.

The inclusion of about 9.4% of added urea in a barley-based cube had little effect on reducing the rate of release of urea into the rumen, compared with the inclusion of about 7.2% urea in a whole hard SBPHU cube. The slow release properties of the barley-urea cube were very comparable with those of the soft SBPHU cube and may be attributable to the very rapid rate of dispersion of this product in the rumen.

There have been many attempts to formulate non-protein nitrogen containing materials which have a reduced tendency to elevate blood and rumen ammonia concentrations than urea and which may therefore be less toxic and perhaps more efficiently utilised by ruminants. Table 6. lists a series of experiments in which a number of these products have been investigated. In general the amount of urea and urea equivalent supplied as one or other of the various products was about one-half that normally considered to be toxic when administered in a single dose to a hungry animal. Wherever possible the effects on the concentration of ammonia in the rumen liquor and in the blood 1 hour after administration have been tabulated. In comparison with insoluble non-protein nitrogen compounds of quite a different nature to urea such as biuret (Broome, 1968) and isobutylidene diurea (Parkins et al. 1971a) inclusion of urea in a hard rather than a soft SBPHU cube has only a marginal effect on reducing rumen ammonia concentration.

Table 6. The comparative effects of urea-containing molassed

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sugar beet pulp cubes and a number of alternative nonprotein nitrogen compounds on the concentration of ammonia in the rumen liquor (mg/100ml) and in the blood ( $\mu$ g/100ml) of cattle 1 hour after administration.

Reference	Product c	g urea equivalent/kg	Rumen	Blood
		L.W.	инз-и	<sup>NH</sup> 3 <sup>-N</sup>
Experiment 1.3	Whole hard SBPHU cubes	0•20	52	117
Experiment 1.3	Whole soft SBPHU cubes	3 0∙20	64	248
Experiment 1.3	Barley-urea cubes	0+20	73	271
Experiment 1.4	Whole SBPU cubes	0•18	49	251
Broome (1968)	Biuret	0.14	10	-
	Urea	0•10	55	-
Parkins et al.(1971a)	IsobutyLidenediurea	0•22	18	not detected
	Urea	0.22	80	100
				200
Ritchie et al.(1972)	Urea phosphate	0•30	62	35
	Urea	0•30	74	215
Ritchie et al*(1972)	Urea phosphate	0•30	••	10
	Urea	0•30	-	220
		-		
Russel et al.(1962)	Diammonium phosphate	0•24	-	217
	Urea	0•24	-	664
Hemingway (1969)	Starea	0•22	82	135
	Urea and ground barley	0.22	100	145
Vender and (1000)		0.00	1 ~	
Hemingway (1970)	Starea	0.22	65	
	Urea + maize meal	0•22	75	
Stiles et al.(1970)	Starea	0•36	37	-
	Urea + unprocessed gra	in 0.36	53	-

\* Experiment conducted with sheep.

The reduction in blood ammonia concentration from about 250 to 117µg/100ml achieved by increasing the hardness of the SBPHU was not as great as that recorded by Ritchie et al.(1972) for the comparison between urea and acidic urea phosphate, when administered to both cattle and sheep. On the other hand the reduction in blood ammonia concentration was of the same order as that recorded by Russel et al.(1962) in a comparison between urea and the less acidic diammonium phosphate. There is some difference of opinion regarding the merits of "Starea" as a slow release product. Stiles, Bartley, Meyer, Devoe and Pfost (1970) reported that the feeding of "Starea" to cattle lowered the concentration of ammonia nitrogen in the rumen liquor more than did feeding equivalent quantities of urea and unprocessed grain. "Starea" was also less toxic. Hemingway (1969, 1970) compared "Starea" with a mixture of either urea and ground barley or urea and maize meal. In both the latter experiments the concentrations of ammonia nitrogen in the rumen liquor were less when "Starea" was given. However, in the comparison between "Starea" and urea plus ground barley (Hemingway 1969) there was no reduction in the concentration of ammonia in the blood associated with the administration of "Starea." This is not consistent with a slow rate of hydrolysis of "Starea" in the rumen. In many respects the concentrations of ammonia produced in the rumen liquor following the administration of "Starea" in the experiments of Hemingway (1969, 1970) were comparable with those when very similar quantities of urea were supplied in either the whole soft SBPHU cubes or the barleyurea cubes given in Experiment 1.3.

It is concluded that although the slow release properties of a hard molassed sugar beet pulp cube containing about 7.2 % of added urea were not as marked as those of a number of more

expensive and sophisticated non-protein nitrogen containing materials, there would seem to be some advantage in producing a hard, rather than a soft, more loosely formed urea-containing product. It must be pointed out however that in the three experiments where the high urea molassed sugar beet pulp cubes were investigated about 1.4kg of the various products were either fed or administered per runen fistula. In practice this amounted to about 100g of urea being supplied by each of the products. Even so, the highest blood ammonia concentration was only about 248µg NH3-N/100ml and was recorded 1 hour after the whole soft SBPHU product was fed in Experiment 1.3. It is generally considered (Repp et al. 1955) that toxicity signs do not develop below about 600µg NH<sub>3</sub>-N/100ml. The high urea supplemented molassed sugar beet pulp products were intended for use as a protein concentrate and to be mixed with several times their own weight of cereals and it is unlikely that an intake in excess of 100g urea at one particular meal would be found in practice. Accordingly there appears to be an adequate safety margin when these particular materials are included in diets for ruminants.

#### SECTION 2

NON-PROTEIN NITROGEN AND PHOSPHORUS CONTAINING COMPOUNDS FOR USE IN MOLASSED SUGAR BEET PULP.

# Introduction.

Molassed sugar beet pulp (SEP) typically contains 0.8 % Ca and 0.08 % P. It is frequently used to replace cereals as an energy source for growing cattle or to supplement poor quality roughage feeds given to ruminants for maintenance purposes. In comparison with cereals (c.0.07 % Ca, c.0.35 % P) for an equivalent energy intake, molassed sugar beet pulp supplies appreciably more calcium but markedly less phosphorus. Phosphorus, but not always additional calcium, may also be necessary when SEP is used to supplement poor quality hays, straws and silages many of which, although well supplied with calcium (c.0.5 %), commonly contain only about 0.10 to 0.25 % P. The material most frequently used to correct deficiencies of phosphorus in ruminant diets is dicalcium phosphate (c.25 % Ca, c.17 % P).

Parkins (1972) has described the production of a supplemented molassed sugar beet pulp cube containing 3.0 % of added dicalcium phosphate, 2.8 % urea, trace minerals and vitamins A and D to give a product with up to 0.6 % P, 1.25 % Ca and 17 % crude protein. This product (Triple Nuts, British Sugar Corporation Ltd.) could be used as a milk production concentrate when fed at 1.8 kg/5 kg of milk and would supply sufficient calcium and phosphorus to meet the suggested (ARC, 1965) requirements without the need for further dietary supplementation. A 500 kg Friesian dairy cow for example, producing 20 kg of milk/ day and receiving 30 kg of silage (0.6 % Ca, 0.25 % P in DM) for maintenance and Triple Nuts (1.4 % Ca, 0.7 % P in DM) for all the milk produced, would have a total daily intake of 131 g Ca and 62 g P/day compared with a recommended (ARC, 1965) intake of

74 g Ca and 60 g P/day. The provision of some 57 g Ca/day in excess of the recommended (ARC, 1965) requirement may be not only unnecessary but also possibly undesirable.

Hignett and Hignett (1951) and Hignett and Hignett (1953) in a survey of 802 cows in 39 herds reported that a high ratio of calcium to phosphorus particularly in a low phosphorus diet was detrimental to the breeding efficiency of dairy cows, especially in late winter when the vitamin D status of cows may be low. In further controlled experiments (Hignett, 1956; Littlejohn and Lewis, 1960) however, where many considerable alterations were made in both the calcium and phosphorus intakes of heifers a high ratio of calcium to phosphorus in the diet was not associated with any conclusive detrimental effects on fertility. Nevertheless an intake of 57 g Ca in excess of the recommended daily requirement for calcium may not be needed. Triple Nuts supplemented with urea, vitamins and phosphorus alone would contain about 0.9 % Ca in the dry matter and would provide more than adequate amounts of calcium for milk production. Similarly, when 3.5 kg/day of Triple Nuts are fed as a straw balancer to 400 kg pregnant cows receiving about 5 kg/day of straw (0.3 % Ca, 0.1 % P in DM) the total daily intake of about 57 g Ca would greatly exceed the ARC (1965) recommended intake of 37 g Ca/day for cows in late pregnancy. The daily phosphorus intake of about 26 g P/day would be very close to the recommended intake of 30 g P/day.

The inclusion of dicalcium phosphate in molassed sugar beet pulp often provides unnecessary amounts of additional dietary calcium. A number of phosphorus sources which contain nitrogen instead of the more normal calcium are available. These include urea phosphate, diammonium phosphate, mono-ammonium phosphate and ammonium polyphosphate. Magnesium phosphates are also available in experimental quantities and some work with these

materials will be described in Section 3 of this Thesis.

Urea phosphate (c.17 % N, c.19 % P) has been reported (Russoff, Lovell and Waters, 1962) to be a satisfactory source of both nitrogen and phosphorus for growing heifers. Perez, Warner and Loosli, (1967) have also demonstrated urea phosphate to be an effective means of dietary nitrogen supplementation for both sheep and calves. They further showed that dietary urea present as urea phosphate was less potentially toxic than urea alone, probably because of the effect which urea phosphate has in depressing the pH of rumen contents and so limiting any potential increase in the concentration of ammonia in the rumen liquor and blood. Hitchie, Parkins and Hemingway (1972) and Hemingway, Parkins and Ritchie, (1972b) have also shown that the concentrations of ammonia in the blood of sheep following oral dosage with urea phosphate were less than where equivalent amounts of nitrogen and phosphorus were given as urea plus dicalcium phosphate.

Diammonium phosphate has been demonstrated by a number of workers (e.g. Oltjen, Waller, Nelson and Tillman, 1963; Johnson and McClure, 1964; Schaadt, Johnson and McClure, 1966) to be a well utilised source of dietary phosphorus but to be a generally less effective nitrogen source than urea for ruminants. Diets containing diammonium phosphate were often incompletely consumed; some of the nitrogen was frequently lost during any cubing process involving heat treatment and ammonia was found to be released when in contact with damp troughs or ruminant saliva. No such difficulties were encountered with mono-ammonium phosphate (Oltjen et al. 1963) but no experimental details were given. Reaves, Bush and Stout, (1966) were able to overcome some of the disadvantages of diammonium phosphate by using a mixture of monoanmonium phosphate and diammonium phosphate plus urea. Ammonia

was released less rapidly from the mixture than from diammonium phosphate, and lactating dairy cows of both average and high producing ability readily consumed rations containing 1.25 and 2.50 % of the mixture. Richardson, Perry, Dunn, Smith and Harbers, (1966) fed mono-ammonium phosphate at levels up to 204 g/day to steers and 34 g/day to sheep and reported no obvious symptoms to toxicity.

Ammonium polyphosphate, a material containing a mixture of ammonium ortho-phosphate and short chain ammonium polyphosphates (generally not above tetrapolyphosphates) is normally available in liquid form. Collenbrander, Muller, Wasson and Cunningham (1971) added either 0.58 % of ammonium polyphosphate (10.0 % N, 14.8 % P) or 0.50 % urea to corn stover silage at the time of ensiling. The silages were fed with a dry protein supplement so that the rations were isonitrogenous at 12 % crude protein. Heifers receiving ammonium polyphosphate treated silage gained significantly more weight than did heifers fed the control or urea treated silage. There was also evidence that ammonium polyphosphate provided a readily available source of phosphorus and is in agreement with the results previously reported by Noller, Collenbrander, Lane, Cummings and Muller (1967). Johnson and McClure, (1967) have demonstrated that liquid ammonium polyphosphate is as well utilised as dicalcium phosphate, as a source of dietary phosphorus for growing (230 kg) steer calves and adult sheep.

Urea phosphate, mono-ammonium phosphate and ammonium polyphosphate each contain low concentrations of fluorine (UP, 170 ppm F; MAP, 2000 ppm F; APP, 60 ppm F) and would seem to be materials of potential value as both phosphorus and non-protein nitrogen sources for ruminants without at the same time increasing a perhaps already adequate calcium intake.

All three materials are also very soluble in water and might therefore be dissolved in molasses prior to incorporation into molassed sugar beet pulp.

The present experiments (Experiments 2.1 and 2.2) describe two nutritional balance trials where growing wether sheep were fed a basal low phosphorus diet supplemented with either urea phosphate (Urea P; 16.5 % N, 18.5 % P), mono-ammonium phosphate (MAP; 10.1 % N, 23.5 % P) or a granular ammonium polyphosphate (APP; 12.5 % N, 28.0 % P). In Experiment 2.1 where Urea P and MAP were given as phosphorus supplements comparisons were made with equivalent amounts of nitrogen and phosphorus supplied as urea (46.0 % N) and dicalcium phosphate (DCP) containing 16.0 % P.

#### Techniques used in nutritional balance trials.

Three nutritional balance experiments are described in this Thesis. Experiments 2.1 and 2.2 have been concerned with the evaluation of non-protein nitrogen and phosphorus supplements; Experiment 3.1 (Section 3) involved comparisons between various calcium and magnesium phosphates. The three experiments were conducted in a 12 metres length x 7 metres width Nissen hut which had been converted into a metabolism house. The Nissen hut had a concrete floor, a suitable supply of water and was large enough to house 12 metabolism cages. There was also adequate space in which to weigh out and store food materials. Two types of metabolism cage were used in the experiments. In Experiments 2.1 and 2.2 they were of the design described by Duthie (1959), In Experiment 3.1 modified metabolism cages which had solid, non-adjustable (but removable) sides, and food troughs fitted with a protective meshed flap were used. Both types of cage had a wire mesh floor, below which was attached a sloping tray from which urine could be collected.

In each balance trial (Experiments 2.1, 2.2 and 3.1) growing wether lambs received a basal low phosphorus diet consisting of shredded molassed sugar beet pulp (SBP) and chopped hay (about 2 - 3 cm length). The actual mineral supplements and /or urea fed to each individual sheep were changed every 14 days and, in consequence, a 7-day balance period alternated with a 7-day change-over period when collections of urine and faeces were not required.

The most important features of nutritional balance trials are controlled feeding and the quantitative collection of both faeces and urine. In Experiments 2.1, 2.2 and 3.1 a standard procedure was adopted for the daily routine of feeding fixed amounts of materials and for the collection and sub-sampling of urine and faeces.

#### Weighing of foods.

The most efficient and convenient procedure was to weigh the total amounts of both hay and SEP required by each sheep per 7 days on one occasion each week. Separate portions of chopped hay  $(\pm 1 \text{ g})$  were weighed into polythene bags using a Sauter 5 kg Self-indicating scale balance. The daily rations of SEP  $(\pm 1 \text{ g})$  were similarly weighed directly into paper bags. At the same time representative samples of chopped hay and SEP were obtained for laboratory analysis. The mineral supplements and urea additions were each weighed  $(\pm 0.01 \text{ g})$  into sealed polythene containers using a Torbal 400 Balance. Thereafter the appropriate mixture of minerals and urea were individually mixed by hand through the daily allowance of SEP contained in each paper bag.

### Feeding and watering arrangements.

The SBP was fed in two approximately equal parts at 8.00 hours and 16.00 hours each day. Hay was given in one feed per day at 10.00 hours. Food residues were removed where necessary on alternate days and stored in sealed polythene bags until the end of each collection period. Water was provided in non-spill galvanised containers which were replenished twice per day. Collection and sub-sampling of faeces.

The inclusion of a high proportion of SEP (or other concentrate feeds) in diets for sheep tends to produce rather soft nonpelleted and perhaps rather fluid faeces which are often difficult to collect completely and efficiently. Two of the most common devices available for the collection of faeces from sheep are (a) a semi-rigid rubberised bag (Avon Rubber Co. Ltd.) or (b) a canvas bag which may, for convenience, be fitted with a zipfastener to facilitate faeces removal. Both the rubberised and canvas faecal bags are difficult to clean out properly particularly when the faeces are not well pelleted. Where the rubberised bag is used a more suitable procedure is to place a

polythene bag inside the rubberised bag to act as an internal liner. This particular type of faecal bag is somewhat expensive, rather bulky and not entirely suitable for sheep of liveweight below about 25 kg.

In a series of preliminary experiments with sheep maintained in metabolism cages it was found that a polythene bag alone could be used quite satisfactorily to collect rather soft faeces. Polythene bags of various size and gauge, together with a number of different methods of attachment to the hind quarters of sheep were investigated. The most suitable procedure with wether sheep was to clip them free of wool around the hind quarters and to fit a standard type of harness which preferably included a chest strap. One strap encircled the sheep behind the fore-legs and another was placed in front of the rear quarters. A 55 cm length of 7 mm polythene tubing was formed into a circle about 17.5 cm in diameter and the ends joined with a suitable solid plug about 5 cm in length. A polythene bag (200 gauge) measuring 30 cm width x 40 cm length was passed through the centre and turned over the outside of the circular polytheme tube to allow an overlap of about 8 cm. The polythene bag was held in place with four spring-loaded cast scissor-grip hooks (F. Martin & Son. Bridgeman Street, Walsall) placed at approximately equal intervals round the circumference of the tubing (Figure 1). The polythene bag was held securely to the tubing by the two semicircular jaws of the scissor hooks.

Each of the four scissor hooks were fastened by means of separate short lengths of nylon cord through holes made at appropriate points in the leather harness strap which passed in front of the hind quarters of the sheep. The two lowest cords were led beneath the body of the sheep, rather than round the hind quarters, prior to attachment. The length of the cords was such as to effect a close fitting of the polythene bag with



- 30cm
- Figure 1. Polythene faecal bag and supporting frame. The polythene bag is held securely to the supporting frame by the semi-circular jaws of the spring-loaded scissorgrip hooks.

the rear of the sheep.

The polythene bag and supporting frame are easily removed by appropriate pressure to release the scissor hooks. Replacement with a new polythene bag takes only a few seconds.

The total capacity of the bag is similar to that of the commonly used rubberised bag. It will conveniently hold about 1 kg of fresh faeces. The bag has been found not to be torn or dislodged by close and frequent contact with the metal sides and wire mesh floor of metabolism cages. During Experiments 2.1, 2.2 and 3.1 a total of 770 faecal collections were made over a 12week period from 15 sheep (liveweight 20 to 45 kg) and the bag was partially dislodged on only two instances.

The particular advantages of the technique described are those of availability, cheapness and simplicity and the ability to handle soft, non-pelleted faeces. The polythene bag is easily numbered for identification purposes and after sealing the faecal material may be conveniently handled and transported.

Throughout both Experiments 2.1, 2.2 and 3.1 the polythene bag was changed each day during both the collection and changeover periods. It was determined that this procedure was both more simple and rapid than the daily removal, emptying and refitting of either a canvas bag or rubberised bag. During the collection periods the daily output of fresh faeces was weighed  $(\pm 1 \text{ g})$  in the polythene bag, spread on a polythene tray and a weighed 10 % sub-sample was transferred to an appropriately labelled polythene bag intended to hold the combined 7-day subsample collection. The bulked 7-day sub-sample was retained under refrigeration until subsequent analysis. During the change-over periods, the faeces collected each day were discarded. <u>Collection and sub-sampling of urine</u>.

Urine was collected into a numbered polythene bottle (4.5 litre) fitted with a funnel holding glass wool and containing 25 ml 12 N sulphuric acid (Martin, 1966) placed directly beneath the exit point in the centre of the collection tray situated beneath the floor of the metabolism cage. The metal tray was rinsed with the minimum quantity of water each morning (at the same time as the faecal bags were changed) and the washings were allowed to pass into the polythene collection bottle. The total volume of urine + washings was measured ( $\pm$  5 ml) in measuring cylinders and 10% was transferred to a Winchester bottle intended to hold the complete 7-day sample. The bulked urine sample was retained under refigeration for analysis. It was found convenient to have a duplicate set of bottles (containing the added acid) available to place directly under the collection point each day. The urine and washings were allowed to run to waste during the change-over weeks.

The time cost of the routine procedures involved in a nutritional balance trial with wether sheep.

# Introduction.

Experiments 2.1 and 3.1 (Section 3) were conducted simultaneously in the form of two 5 x 5 Latin square designs involving a total of 10 growing wether sheep. In each balance trial there were 5 experimental periods each of 2 weeks, the first 7 days of each period being treated as change-over weeks. Complete collections of urine and faeces were made during the second 7 days for input-output balance purposes. The total experimental period involved was therefore 10 weeks. There seems to be no published information on the time-cost of the separate procedures involved in a nutritional balance trial. This may be of considerable interest where large numbers of animals (10 - 20) are employed in experiments of this type. During the course of Experiments 2.1 and 3.1 a record was kept of the time involved in the routine of feeding fixed amounts of hay and supplemented SBP and the daily collection and sub-sampling of faeces and urine from 10 sheep.

#### Results.

The timings of the individual operations described for the weighing of foods and the collection of urine and faeces samples are given in Table 7. These represent the mean of three separate assessments.

The total operational time involved in each 7-day collection period for the 10 sheep was 22 hours 17 minutes. The essential daily procedures (excluding the weekly weighing and mixture of foods) totalled 2 hours 24 minutes. This was reduced to 1 hour 28 minutes during each change-over week when recordings of urine and faecal outputs were not made (but where the faecal bags were changed each day to minimise the time taken to clean the cages). It can be calculated for example, that the total operational time involved in a single  $5 \times 5$  Latin square design involving 5 sheep for a total period of 10 weeks would be 109 hours 5 minutes.

The single most time-consuming item involving 33% of the total was that involved in the daily routine of removing and replacing the faecal bag, washing down the cages and changing the urine collection vessel. Where the experiment is such that caged sheep discrete, well-pelleted faeces (as was not the case in this particular trial) some saving in time might be effected by using the stainless steel urine/faeces separator described by Duthie (1959) or by employing a stretched nylon mesh (beneath the cage floor) which allows the selective passage of urine. A further total of 29.4 % of the total time was concerned with the measurement and sub-sampling of the daily faecal and urine outputs.

The daily feeding routine took only 2 minutes per sheep per day and represented some 10.6 % of the total time. The removal of food residues (2.6 % of the total time) was only a small item in these particular trials but could assume much greater importance in certain circumstances.

				00
Tab	le 7. The times (mins) taken in th	ie sepa	rate pro	ocedures
	of a 7-day nutritional balar	ice tri	al with	10 sheep.
		Each day	7-day total	% of total time
1.	Weighing of food intake for 7 days			
	(a) Hay	-	54	3•9
	(b) Concentrate		75	5•6
	(c) Concentrate additives averaging			
	2/day/sheep	<del></del>	150	11.2
	(d) Intermixture of additives with		•	
,	concentrate	-	50	3•7
2.	Provision of food and water			
	(a) Feeding, filling water bowls	20	140	10•6
	(b) Removal of food residues	5	35	2•6
3.	Urine and faecal collection			
	(a) Remove and replace faecal bag;	63	441	33+0
	wash down cages; remove and			
	replace urine collection vessel			
	(b) Faeces. Weigh; sub-sample 10 %	31	217	16•3
	to cumulative total			
	(c) Urine. Measure volume;	25	175	13•1

sub-sample 10 % to cumulative total

Total (7 days for 10 sheep) 22 hours 17 mins 100.0

The once-weekly weighing of food samples took 5 hours 29 minutes for the 10 sheep. (i.e.  $24 \cdot 4 \%$  of the total). A surprisingly large proportion of this time was concerned with the weighing and admixture of the mineral and urea additives for each sheep involved in these particular trials.

The operations involved were conducted expeditiously and with good facilities by two well-motivated individuals and represent perhaps the minimum time which can be attained for a trial of this type. No allowance has been included for miscellaneous items such as; assembly, repair and renewal of metabolism cages, necessary adjustment of harnesses, chopping of hay and preparation of concentrates, prior acclimatisation of sheep in cages, weighing of livestock and other animal treatments such as blood sampling and collection of rumen liquor.

The total experimental time-cost necessarily includes the associated analytical work which is not considered in detail here. In this particular trial, determinations were made on food, faeces, urine and blood by standard, non-automated methods of analysis in respect of phosphorus, magnesium, calcium and nitrogen. The operator-time involved in chemical analysis (including sample preparation) was 38 hours for each 7-day collection from the 10 sheep. This is in the same order as the total of almost 44 hours involved in the 7-day collection and 7-day change-over period for the 10 sheep.

Experiment 2.1

Urea phosphate and mono-ammonium phosphate as dietary phosphorus supplements for sheep

# Materials and Methods.

Five wether sheep (Suffolk x Greyface cross, aged about 3 months and initial liveweight 33 kg) were fed each of five diets in sequence in a 5 x 5 Latin square design. During an initial acclimatisation period of 14 days the sheep in metabolism cages (Duthie, 1959) were fed 900 g shredded molassed sugar beet pulp (SBP) supplemented with 11.25 g urea and 11.25 g DCP/day plus 100 g chopped hay.

During the subsequent 70-day experimental period there were 5 periods each of 14 days. Collections of urine and faeces were made in the last 7 days of each period. The amounts of basal diet (g fresh matter/day) given during each period were progressively

increased to ensure that the sheep would continue to make good growth, namely: Period 1, 900 g SBP + 100 g hay; Period 2, 1100 g SBP + 130 g hay; Periods 3 and 4, 1100 g SBP and 140 g hay; Period 5, 1300 g SBP + 160 g hay.

The SBP contained 1.68 % N, 0.06 % P and 0.68 % Ca (DM basis) and the hay contained 0.93 % N, 0.16 % P and 0.40 % Ca (DM basis). Accordingly, the total amounts of nitrogen, phosphorus and calcium in the basal diet increased from 14.46 g N to 21.02 g N, from 0.63 g P to 0.93 g P and from 5.87 g Ca to 8.55 g Ca in progression from Period 1 to Period 5.

The five supplements are detailed in Table 8. In Diet A, no supplement was given and the mean total phosphorus and nitrogen intakes were 0.71 g and 16.0 g respectively. Diets B, C and D provided an additional 1.75 g P and 4.6 g N/day. The additions were as DCP + urea (Diet B), as Urea P + urea (Diet C) and as MAP + urea (Diet D). In Diet E the supplement was 4.6 g N/day from urea but with no additional phosphorus. No attempt was made to equalise calcium intakes because of the already adequate calcium content of the basal diet (6.5 g Ca/day) but Diet B (DCP + urea) supplied 2.90 g Ca more than did the other diets.

Table 8. The amounts of each mineral supplement and urea provided (g/day) and the effective additions of nitrogen, phosphorus and calcium (g/day).

Diet	A	в	C	D	E
Urea	-	10.00	6•61	8•41	10.00
DCP	-	10•94	-	-	-
Urea P	-	- '	9•45	-	-
MAP	-		-	7•29	-
Effecti	ve l	Additions			
N	-	4•60	4•60	4•60	4•60
Р	-	1•75	1.75	1.75	
Ca		2.90	_		843

The feeding and watering arrangements and the daily procedures involved in the collection and sub-sampling of faeces and urine have been described previously. The sheep were weighed on the last day of each feeding period. Blood samples for the determination of phosphorus, calcium and urea were obtained prior to the morning feed on the 11th and 14th day of each feeding period. During Period 4 two sheep (Nos. 182 and 100) were bled on several occasions (Days 0, 2, 4, 7, 11 and 14) throughout the 14 day period, so that an assessment could be made of the rate at which the blood phosphorus concentration altered after the change from a nil to a phosphorus supplemented diet or viceversa. The respective treatment sequences were; sheep 182 Diet E to Diet B (nil to  $\pm 1.75$  g P/day); sheep 100 Diet D to Diet A ( $\pm 1.75$  g P/day to nil).

Nitrogen, phosphorus and calcium were determined in vrine, feeds and faeces. The appropriate analytical methods are described in the Appendix. The digestibility of the sugar beet pulp dry matter was determined from the feed input and faeces output data.

In calculating the amounts of faeces attributable to the sugar beet pulp alone, the digestibility of the DM in the small amount of chopped hay given was taken as 60 %.

#### Results (Table 9.)

# Live-weight gain.

The addition of both nitrogen and phosphorus resulted in fully satisfactory daily live-weight gains of 0.18 kg (Diet D containing MAP), 0.23 kg (Diet C containing Urea P) and 0.26 kg (Diet B containing DCP). These were all significantly greater than the mean gains of 0.09 kg/day recorded for both Diet A (no supplement) and Diet E which did not contain added phosphorus.

phosphorus and calcium balances (g/day) and mean plasma urea, phosphorus and calcium concentrations (mg/100 ml)Mean daily live-weight gain (kg), total DM intake (g/day), digestibility of the SBP DM (g), mean nitrogen, at the end of each 14-day feeding period. Table 9.

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	Significance	B≻A, E**; C, D≻A,	N.S.	A, B, C>E**	- Б, С, D, E>A** N.S. B, С, D>A, <u>E</u> **	- N.S. B, C, DZA, E** B, C, DZA, E**	- A, E>B, C, D** B, E>A, C, D** B>A, E**; B>C*;	N.S. B. C, D>A, E** A, E>B, C, D**
S. E.	of mean (+)	0.029	31.8	0.654	0.495 0.412 0.381	0.013 0.096 0.107	0.024 0.321 0.279	2.053 0.268 0.227
더	urea	60°0	1035	79.05	20.94 9.35 8.48 +3.11	0.72 0.01 0.93 -0.22	6.76 0.22 6.91 -0.37	28.88 2.67 11.98
Q	MAP+ urea	0.13	1011	81 <b>.</b> 03	22.28 9.10 8.31 +4.87	2.52 0.06 1.43 1.05	7.18 0.05 5.31 †1.82	30.33 6.07 10.37
U	UreaP+ urea	0.23	TTTT	82.37	22.38 9.35 8.14 +4.89	2.54 0.03 1.47 +1.04	7.22 0.06 6.12 +1.04	32.35 6.18 10.46
щ	DCP+ urea	0.26	LOIT	83.50	22.34 8.89 7.82 +5.63	2.53 0.04 1.35 +1.14	10.11 0.05 7.48 +2.58	31.25 6.21 10.37
A	LiN	0.09	1001	82.38	16.00 6.10 6.76 +3.14	0.71 0.01 0.82 -0.12	6.50 0.18 5.94 +0.38	24.53 2.60 11.75
		gain	take	DM	Input Urine Faeces Retention	Input Urine Faeces Retention	Input Urine Faeces Retention	Urea P Ca
Diet	Supplement	Live-weight gain	Total DM intake	Dig.of SBP	Nitrogen	Phosphorus	Calcium	Blood

#### Food consumption and DM digestibility.

Where both nitrogen and phosphorus supplements were provided the diets were invariably well consumed and there were no food residues of any account or any palatability problems. Where neither additional nitrogen or phosphorus (Diet A) or additional phosphorus alone (Diet E) were provided, three of the five sheep left appreciable amounts of hay and sugar beet. This generally occurred within about 7 - 10 days of a change from a fully supplemented diet (B, C or D) to one which was inadequate in either nitrogen or phosphorus. With a subsequent change to full supplementation in the next feeding period the intake of food was restored within 2 - 3 days. Addition of both nitrogen and phosphorus had no significant effect on the digestibility of the DM in the SEP. Where additional nitrogen alone was provided (Diet E) the DM digestibility tended to be depressed.

# Nitrogen retention.

Nitrogen retention was significantly increased from about 3.1 g N/day to about 4.87 - 5.63 g N/day when both nitrogen and phosphorus supplementation were given. For Diet A where no additional nitrogen was given there was a significantly reduced output of nitrogen in the urine (6.1 g/day compared with about 8.9 - 9.4 g/day). The mean concentrations of urea in the blood plasma (11 and 14 days after supplementation) showed that nitrogen and phosphorus additions produced values of about 30 - 32 mg urea/100 ml. compared with 24 mg/100 ml (Diet A, no additions) and 28 mg/100 ml (Diet E, with urea only).

# Phosphorus retention.

Supplementation with phosphorus increased the total daily phosphorus intake to about 2.5 g P/day (Diets B,C and D) compared with only about 0.7 g P/day for Diets A and E. In the absence of added phosphorus there were apparent negative retentions of -0.12

and -0.22 g P/day compared with positive and very similar retentions of 1.03 - 1.14 g P/day when all three forms of additional phosphorus were fed. There were also indications that supplementary dietary phosphorus increased urine phosphorus output.

# Calcium retention.

The largest apparent calcium retention (2.58 g/day) was recorded for Diet B which contained dicalcium phosphate and which accordingly gave the highest daily intake of 10.1 g Ca/day. Both Urea P and MAP resulted in calcium retentions of 1.04 and 1.82 g/ day respectively which were significantly greater than those of + 0.38 g Ca/day for Diet A, (no supplement) and -0.37 g Ca/day for Diet E (urea only). All forms of phosphorus supplementation markedly and significantly reduced the amounts of calcium in the urine (about 0.05 g ca/day) compared with those found with an inadequate phosphorus intake (0.18 and 0.22 g ca/day).

# Blood phosphorus and calcium concentrations.

Where no additional phosphorus was provided, the mean concentrations of phosphorus in the blood plasma were only about 2.6 mg P/100 ml compared with about 6.0 - 6.2 mg/100 ml withadequate supplementation. There were corresponding but reversed changes in the concentrations of calcium in the blood plasma. A value of about 10.4 mg Ca/100 ml in the presence of adequate dietary phosphorus was increased to about 11.8 mg Ca/100 ml some 11 - 14 days after removal of the phosphorus supplements. The data from the two sheep which were bled on alternate days during Period 4 (Table 10) indicated that changes in the concentrations of phosphorus in the blood plasma were rapidly initiated within 2 - 4 days of either the addition or withdrawal of supplementary dietary phosphorus.

Table 10. The blood phosphorus concentrations (mg/100 ml) of 2 sheep 0 - 14 days after either the addition or withdrawal of supplementary dietary phosphorus.

Dietary Sheep Day 0 2 change no. 9 11 14 4 7 nil to + 1.75 g 182 2.75 3.10 4.30 5.93 6.13 5.55 7.60 P/day

+ 1.75 g P/day 100 5.50 4.50 4.50 3.65 3.23 3.30 2.95 to nil

Experiment 2.2

Granular ammonium polyphosphate as a dietary phosphorus supplement for sheep.

# Materials and Methods.

On completion of Experiment 2.1 a further nutritional balance trial was conducted to evaluate a granular ammonium polyphosphate (APP; 28 % P, 12.5 % N) as a phosphorus source. The 5 wether sheep (mean liveweight 42 kg) which had been used in Experiment 2.1 were each fed a basal low phosphorus diet consisting of 1160 g of SBP (0.06 % P, 1.68 % N DM basis) and 140 g of chopped hay (0.15 % P, 1.05 % N DM basis) to which was added 6.25 g APP and 8.30 g urea per day. The basal diet contained 18.95 g N and 0.82 g P. The APP + urea supplement provided an additional 1.75 g P and 4.60 g N and accordingly the total daily intakes of nitrogen and phosphorus were increased to 23.55 g N and 2.57 g P/day. The supplemented diet was fed to each sheep for an experimental period of 14 days. The first 7 days were treated as a preliminary adaption period, and during the second 7 days complete collections of faeces and urine were made for inputoutput balance purposes. The daily feeding arrangements, and the

techniques used for the collection and sub-sampling of faeces and urine were as in Experiment 2.1. The sheep were weighed on the first and last day of 14 day feeding period. Blood samples for the determination of phosphorus, calcium and urea were obtained prior to the morning feed on day 11 and day 14.

Nitrogen and phosphorus were obtained in feeds, urine and faeces by the standard analytical methods described in the Appendix. Results (Table 11.)

There were no palatability problems and the five sheep readily consumed the APP supplemented diet. The mean daily live-weight gain of 0.20 kg/day recorded for the five sheep was considered entirely satisfactory in view of the very short term nature of the trial. Supplementation with APP resulted in a very similar phosphorus retention (+1.15 g P/day) as did the provision of an equivalent amount of additional phosphorus in the form of DCP in Experiment 2.1. Nitrogen and phosphorus supplementation as APP + urea in the present trial resulted in a rather lower mean daily retention of nitrogen (+ 4.34 g N/day) than where either form of NP supplementation were given in Experiment 2.1 (see Table 9) principally because of an increased loss of nitrogen in the urine. The mean daily nitrogen intake of sheep fed APP + urea was rather greater than that of the sheep fed either of the three nitrogen and phosphorus supplemented diets in the preceding experiment and this may explain the somewhat higher daily loss of N in the urine.

Table 11. Mean nitrogen and phosphorus balance (g/day) of the 5

sheep fed a supplement of APP + urea.

	Nitrogen	Phosphorus
Input	23.55	2.57
Urine	10.98	0.07
Faeces	8.23	1.35
Retention	+4.34	+1.15

The mean blood phosphorus, calcium and urea concentrations of the sheep fed APP + urea were 6.33, 10.50 and 31.62 mg/100 mlrespectively and the dry matter in the SBP was calculated to be about 82.7 % digestible.

It is concluded from the results of the present trial that granular APP is a satisfactory source of dietary phosphorus for ruminants.

# Discussion and Conclusions (Experiments 2.1 and 2.2).

Urea phosphate, mono-ammonium phosphate and ammonium polyphosphate (with appropriate additions of urea) when used as supplements to a low phosphorus diet resulted in very similar increases in phosphorus and nitrogen retention, urinary outputs and increments in the concentration of phosphorus and urea in the blood as did equivalent amounts of phosphorus and nitrogen supplied as dicalcium phosphate plus urea. All forms of phosphorus supplementation supported growth equally well. There were no palatability problems associated with the feeding of either Urea P, MAP or APP. The reduction in feed intake noted in Experiment 2.1 where neither additional nitrogen or phosphorus were given (Diet A) was a consistent finding throughout the experiment. Addition of nitrogen alone as urea (Diet E) did not relieve the depressive effect of a low phosphorus intake on appetite. It is possible that a low dietary intake of phosphorus adversely affects microbial activity in the rumen and this may lead to a reduced appetite even when there is an adequate intake of nitrogen. Experiments described in Section 4 have been designed to further evaluate the quantitative effects of a low phosphorus intake on voluntary food consumption and rumen metabolism.

Where supplementary phosphorus was provided as either DCP, Urea P, MAP (Experiment 2.1) or APP (Experiment 2.2), the sheep

had a mean total daily intake of 2.54 g P/day. At an overall mean liveweight of 39 kg and daily live-weight gain of 0.22 kg it has been suggested (ARC, 1965) that the minimum phosphorus intake should be about 3.60 g P/day. The overall mean phosphorus retention for the phosphorus-supplemented treatments of 1.09 gp/day implies a storage of about 4.9 g P/kg live-weight gain. This is within the range of 4.5 - 6.1 g P storage/kg live-weight gain described by the ARC (1965). The fact that normal rates of phosphorus storage were recorded in both Experiments 2.1 and 2.2 when the mean phosphorus intakes on the phosphorus supplemented treatments were about 2.54 g P/day which is considerably less than the ARC (1965) recommended intake of 3.60 g P/day may be because such a high proportion (1.75 g of the 2.53 g) was present as one or other of the mineral supplements. Alternatively the ARC (1965) recommendations would appear to be too high.

The granular ammonium polyphosphate used in Experiment 2.1 contained 63.5 % of the total phosphorus as non-orthophosphate and 36.5 % as orthophosphate. The major component of the nonorthophosphate was pyrophosphate. Ammerman, Forbes, Garrigus, Newmann, Norton and Hatfield, (1957) reported that gamma calcium pyrophosphate was a poor source of phosphorus for ruminants, whereas Tillman and Brethour, (1958) showed that acid sodium pyrophosphate was equally as available as mono-sodium phosphate. The present studies would indicate that the nonorthophosphates of ammonium polyphosphate are available.to the ruminant as a phosphorus source.

The suggested ARC (1965) requirement of calcium for 39 kg sheep gaining at about 0.22 kg/day is about 7.1 g Ca/day. The mean amount of calcium consumed in the basal diet when supplemented with either urea (Diet E), Urea P + urea (Diet C) and MAP + urea

(Diet D) in Experiment 2.1 was 7.05 g Ca/ day. Supplementation with DCP (Diet E) increased the mean calcium intake to 10.1 g Ca/day. This resulted in a greater apparent calcium retention of 2.58 g Ca/day, which was significantly (P< 0.05) greater than that of 1.04 g Ca/day recorded for the sheep when fed Urea P + urea. The data obtained for calcium and phosphorus retentions when sheep were fed the three different phosphorus supplements in Experiment 2.1 were also somewhat anomolous in that the ratios of calcium to phosphorus apparently retained were in the proportions 2.3, 1.0 and 1.7 for DCP, Urea P and MAP-containing diets respectively, which were not consistent with a regular pattern of deposition in the body.

In Experiment 2.1 Urea P and MAP and in Experiment 2.2 APP were given primarily as phosphorus sources. At the rates used (to add 1.75 g P/day to the basal diet) the amounts of non-protein nitrogen which each supplied were only a small proportion (about 15 - 30 %) of the total nitrogen supplement provided, the remainder being as urea. Accordingly, it is not possible from these present experiments to evaluate either urea phosphate, monoammonium phosphate or ammonium polyphosphate as nitrogen sources as the majority of additional nitrogen fed in each case was present as extra urea given to equalize nitrogen intakes.

#### SECTION 3

# PHOSPHORUS AND MAGNESIUM CONTAINING COMPOUNDS FOR USE IN MOLASSED SUGAR BEET PULP.

#### Introduction.

The experiments to be described have been concerned with the evaluation of two novel magnesium phosphates; (a) a magnesium phosphate (Mg P; 20 % Mg, 18.5% P) made by reacting calcined magnesite with phosphoric acid and composed principally of hydrated Mg HPO<sub>4</sub> with an additional amount (c. 10 %) of MgO, and (b) a calcium magnesium phosphate (Ca Mg P; 16.1 % Ca, 6.0 % Mg, 18.5% P) made by treating dolomite with phosphoric acid. Both materials are of low fluorine content (Mg P; 110 ppm F, Ca Mg P; 240 ppm F) and may therefore be of use as combined phosphorus and magnesium supplements for sheep and cattle when included in molassed sugar beet pulp.

There is little published information on the effectiveness of magnesium phosphates as mineral supplements for ruminants. Based on the plasma magnesium concentrations in calves Huffman, Conley, Lightfoot and Duncan (1941) indicated that the magnesium of magnesium phosphate was as well utilised as magnesium oxide, carbonate and chloride. Hemingway and Brown (1967) compared a magnesium ammonium phosphate (16 % Mg, 9.0 % N, 20 % P) with dicalcium phosphate (17.5 % P, 23.5 % Ca) as a source of supplementary phosphorus for growing (25 kg) lambs. Retentions of +1.33 and +1.16 g P/day were described for the dicalcium phosphate and magnesium ammonium phosphate treatments respectively. Normal blood phosphorus concentrations were maintained on each treatment. The basal diet supplemented with magnesium ammonium phosphate contained about 0.16 % more magnesium than did the corresponding diet containing dicalcium phosphate and accordingly the blood magnesium concentrations and

daily retentions of Mg were greater in the sheep given the magnesium ammonium phosphate treatment. It was concluded that magnesium ammonium phosphate was a satisfactory source of both phosphorus and magnesium for growing lambs.

It has been shown that phosphorus (e.g. Ling and Smith 1940; Bond 1951; Ford 1956; Smith and Comrie 1948; Hemingway et al. 1968) and magnesium (Hemingway et al. 1968; Hemingway, 1971) inadequacies in relation to the ARC (1965) proposed minimum recommended intakes are very common where herbage and grass products form an important component of the diet of ruminants. In contrast, the dietary calcium requirement is frequently satisfied without recourse to special supplementation. The most common materials used to rectify these mineral deficiencies are dicalcium phosphate (c. 17 % P, c. 25 % Ca) and calcined magnesite (c. 87 % MgO). Magnesium phosphates would seem to warrant investigation as potential dietary supplements and to be of interest where it may be desirable to increase the phosphorus and magnesium intakes of livestock without at the same time providing additional and perhaps undesirable amounts of calcium.

#### Experiment 3.1

# Magnesium phosphate and calcium magnesium phosphate as dietary phosphorus supplements for sheep.

In the present experiment, growing wether sheep were fed a low phosphorus diet which was variously supplemented with either (a) Mg P (20.0 % Mg, 18.5 % P) or (b) Ca Mg P (16.1 % Ca, 6.0 % Mg, 18.5 % P). Comparisons were made in each case with equivalent amounts of phosphorus and magnesium supplied as dicalcium phosphate (DCP; 16.0 % P, 26.5 % Ca) and magnesium oxide (MgO; 60.0 % Mg).

#### Materials and Methods.

Five wether sheep (Suffolk X Greyface cross) with an initial mean liveweight 30 kg and aged about 12 weeks were maintained in metabolism cages. For an initial acclimatisation period of 14 days each sheep was fed a daily basal diet of 900 g shredded molassed sugar beet pulp (SEP) supplemented with 11.25 g DCP and 11.25 g urea together with 100 g chopped hay.

The experiment was conducted in the form of a 5 x 5 Latin square. Each sheep received each of 5 different diets for 14 consecutive days over a total period of 70 days. The first 7 days of each 14-day period were treated as changeover periods and complete collections of urine and faeces were made during the second 7 days of each period for input-output balance purposes.

The intention was that the sheep should grow well during the experimental period. Accordingly, the amount of basal diet was progressively increased for each of the 14-day periods so that the sheep were always fed to slightly less than appetite. The amounts fed (fresh matter basis) for each period were: Period 1, 900 g SEP + 100 g hay; Period 2, 1100 g SEP + 130 g hay; Periods 3 and 4, 1100 g SEP + 140 g hay; period 5, 1300 g SEP + 160 g hay. A constant addition of 10 g urea (mixed with the SEP) was fed throughout the experiment to all the sheep to increase the intake of crude protein.

The SBP contained 0.06 % P, 0.13 % Mg and 0.68 % Ca (DM basis) and the hay contained 0.16 % P, 0.09 % Mg and 0.40 % Ca (DM basis). The total amounts of phosphorus, magnesium and calcium given each day in the basal diets accordingly increased from 0.63 to 0.93 g P, 1.14 g to 1.66 g Mg and from 5.87 to 8.55 g Ca in the progression from Period 1 to Period 5.

The five diets are given in Table 12. The amounts of each material were such that each of the four phosphorus supplements

added 1.75 g P to the basal diet so that the total intake was increased from 2.38 to 2.68 g P/day from Period 1 to Period 5. The MgO additions in Diets B and D were such that they added amounts of magnesium equivalent to those present in the Ca Mg P (Diet C) and in the Mg P (Diet E), i.e. 0.57 and 1.89 g Mg/day respectively. No attempt was made to equalise the additional calcium intakes which varied with each treatment because of the already high calcium intakes from the basal diet.

Table 12. The amounts of each mineral supplement provided and the effective additions of phosphorus, magnesium and calcium to the basal diet (g/day).

Diet	A	В	С	D	Е
Mg P	-	-	-	-	9•45
Ca Mg P		~	9•45	-	80
DCP		10•94	—	10•94	-
MgO	-	0•95	-	3•15	~
Effective Additions					
Р	-	1.75	1•75	1•75	1.75
Mg		0•57	0•57	1•89	<b>1.</b> 89
Ca	<b>44</b> 3	2•90	1•52	2•90	-

The care and management and the procedures involved in the collection and sub-sampling of faeces and urine from the five sheep have been outlined in Section 2. The sheep were weighed every 14 days on the last day of each feeding period. Blood samples for the determination of phosphorus, magnesium, calcium and urea were obtained prior to the morning feed on the 11th and 14th day of each feeding period. During Period 4 the blood phosphorus and magnesium concentrations of two sheep (nos. 98 and 174) were monitored on alternate days throughout the 14 day experimental period. At the start of Period 4 sheep no. 98

had been changed from a nil to a phosphorus and magnesium supplemented diet (Diet A to Diet C; nil to 1.75 g P + 0.87 gMg/day). Sheep no. 174 received a reverse treatment sequence (Diet E to Diet A; 1.75 g P + 1.89 g Mg/day to nil). Representative samples of food and food residues, faeces and urine from each feeding period were analysed for phosphorus, calcium and magnesium by the analytical methods described in the Appendix. In calculating the digestibility of the dry matter in the molassed sugar beet pulp, the digestibility of the dry matter of the small amount of chopped hay which was given to each sheep throughout the experiment was taken as 60%.

# Results (Table 13.)

#### Food consumption and DM digestibility.

There were no palatability problems and the four supplemented diets were all consumed and there were no food residues apart from one sheep in Period 5. Where no additional phosphorus was given (Diet A) three of the five sheep left some food; two refused some 20 % of both the hay and the SBP and one left some 50 % of the hay. These residues only occurred during the last 4-5 days of each 14day period and recovery of appetite invariably occurred within 1-2 days of subsequent phosphorus supplementation. on change of diet. Where nitrogen alone was added to the basal diet (Diet A) the dry matter in the sugar beet pulp was about 80.4 % digestible and was not affected by further phosphorus.supplementation.

# Live-weight gain.

The mean live-weight gain of the sheep when fed the four supplemented diets was 0.14 - 0.22 kg/day compared with only 0.05 kg/day in the absence of additional phosphorus. These liveweight gains were considered reasonably satisfactory for this type of sheep considering the short-term nature of the feeding periods, the close confinement in cages and provision of food

magnesium and calcium balances (g/day) and mean blood phosphorus, magnesium, calcium and urea concentrations Table 13. Mean daily live-weight gain (kg), total DM intake (g/day), digestibility of the SBP DM (g), mean phosphorus,

(mg/100 ml) at the end of each 14-day period.

Diet		Ą	Ъ	Ю	D	[2]	°, E	
Supplement		TIN	DCP+ MgO	CaMgP	DCP+ MgO	MgP	of mean (±)	Significance
Live-weight gain	gain	0.05	0.18	0.22	0.15	7T*O	0,046	C <b>∀</b> A*
Total DM intake	jake	996	1098	1111	1053	I I I	37.1	N.S.
Dig. of SBP	DM	80.42	81.42	81.87	80.82	82.49	1.225	M.S.
Phosphorus	Input Urine Facces Retention	0.66 0.02 0.81 -0.17	2.52 0.05 1.55 0.92	2.54 0.05 1.61 +0.88	2.41 0.05 1.59 t0.77	2.54 0.06 1.62 +0.86	0.024 0.066 0.073	- N.S. B, C, D, E>A*** B, C, D, E>A***
Magnesium	Input Urine Faeces Retention	1.23 0.49 0.64 +0.10	1.96 0.61 1.01 +0.34	1.97 0.56 1.07 +0.34	3.09 0.96 1.67 +0.46	3. 29 0.88 0.11 0.40	- 0.043 0.078 0.059	- D, E>A, B, C*** D, E>B, C>A** D, E>A**; B, C*
Cal.cium	Input Urine Faeces Retention	6.35 0.21 6.10 †0.04	10.07 0.04 8.33 +1.70	8.74 0.03 7.55 +1.16	9.55 0.06 8.08 +1.41	7.22 0.04 6.02 +1.16	0.022 0.272 0.259	- A>B, C, D, <sub>氏**</sub> B, C, D>A, <sub>E**</sub> B, C, D, 丘>A.***
Blood	P Mg Urea	2.53 2.53 2.23 32.03	4.26 2.54 10.38 33.22	4.41 2.48 10.09 31.25	4.33 2.56 30.98	4.71 2.92 10.02 32.05	0.317 0.101 0.223 2.199	B, C, D, EVA** E>B, C, D*, E>A**; D>A* A>B, C, D, E** N.S.

to rather less than full appetite.

# Blood composition.

All four supplemented diets increased the blood phosphorus concentration significantly (P<0.01) to about 4.3 - 4.7 mg P/100 ml compared with the low mean value of 2.5 mg P/100 ml recorded 11-14 days after feeding the basal low phosphorus diet. There were associated differences in plasma calcium in that all the phosphorus supplemented groups had mean values in the range 10.0 - 10.5 mg Ca/100 ml compared with the significantly (P<0.01) elevated concentration of 11.7 mg/100 ml for the sheep when fed no additional phosphorus.

The mean concentration of magnesium in the blood 11-14 days after feeding the unsupplemented diet (which was not considered inadequate in magnesium) was 2.23 mg Mg/100 ml. This was significantly less (P<0.01) than the mean value of 2.92 mg/100 ml when Mg P providing an additional 1.89 g Mg/day (Diet E) and also significantly less (P<0.05) than the mean value of 2.56 mg Mg/100 ml when the same total amounts of magnesium and phosphorus were provided as MgO + DCP (Diet D). The provision of an additional 0.75 g Mg/day as either MgO + DCP or as Ca Mg P (Diets B and C) failed marginally to increase the blood magnesium concentration to a significant degree.

Observations from the two sheep which were bled on alternate days during Period 4 (Table 14) indicated that changes in both blood phosphorus and magnesium concentrations were initiated within 2-4 days of either the addition or withdrawal of supplementary dietary phosphorus and magnesium.

The mean concentration of urea in the blood plasma of the sheep fed Diet A (with urea only) was not significantly different from the mean concentrations recorded when both urea and phosphorus were provided (Diets B, C, D and E). Table 14. The blood phosphorus and magnesium concentrations (mg/100 ml) of two sheep 0 to 14 days after either the addition or withdrawal of supplementary dietary phosphorus and magnesium.

#### BLood phosphorus

Dietary	Sheep			D	ay				
change	no	0	2	4	7	9	11	14	
nil to + 1•75 g P/day	98	1•65	2•45	2•70	3•10	2•65	3•15	2•65 -	
+ 1•75 g P/day to nil	174	5•50	3•50	3.00	3•10	2•63	2•85	2•85	

#### Blood magnesium

+ 0.87 g Mg/day	98	2•18	2•33	2•45	2•45	2•23	2•45	2•50
+ 1.89 g Mg/day to nil	174	2•95	1•85	2•23	2•28	2•18	2•53	2•23

# Phosphorus retention.

The increase in mean daily phosphorus intake from 0.66 to about 2.5 g P/day resulted in similar and significantly (P<0.001) increased apparent phosphorus retentions between 0.77 and 0.92 g P/day for all four supplemented groups compared with an apparent loss of -0.17 g P/day when the basal diet was fed. There were small, but consistent, increases in urine phosphorus outputs for all four supplemented groups.

# Calcium retention.

In the absence of phosphorus supplementation (Diet A) the mean apparent calcium retention was 0.04 g Ca/day. All forms of phosphorus supplement significantly (P<0.001) increased calcium retention. When DCP was given (Diets B and D) retentions at 1.4 and 1.7 g/day were greater than when either Ca Mg P (Diet C) or Mg P (Diet E) were fed and where the total daily calcium intake

was correspondingly rather less. All forms of phosphorus supplementation significantly (P<0.01) decreased the mean daily output of calcium in the urine from 0.21 g (Diet A) to about 0.03 - 0.06 g Ca.

# Magnesium retention.

Supplementation with 1.89 g Mg/day as either MgO + DCP (Diet D) or as Mg P (Diet E) significantly (P<0.01) increased apparent magnesium retentions from 0.10 (Diet A) to 0.40 and 0.45 g Mg/day respectively. Supplementation with 0.57 g Mg/day as either MgO + DCP (Diet B) or Ca Mg P (Diet C) also increased (P<0.05) the apparent retention to about 0.34 g Mg/day. The higher, but not the lower, rate of magnesium inclusion resulted in significantly (P<0.001) increased outputs of magnesium in the urine. Discussion and Conclusions.

# The primary purpose of the experiment was to assess the efficiency of magnesium phosphate and a calcium magnesium phosphate as phosphorus sources relative to dicalcium phosphate. Both materials significantly and similarly increased blood phosphorus concentrations and apparent phosphorus retentions. They also reduced blood calcium concentrations and urinary calcium outputs relative to the elevated value found when the low phosphorus diet was fed, which would suggest that each material was an equally efficient source of phosphorus.

The mean phosphorus retention over all four supplemented treatments was equivalent to 5.0 g P/kg live-weight gain. This is in agreement with the ARC (1965) conclusion for 35 kg sheep gaining 0.17 kg live-weight/day, and almost exactly the same as the mean storage of 4.9 g P/kg live-weight gain recorded in Experiments 2.1 and 2.2 (Section 2) with comparable sheep fed either urea phosphate, mono-ammonium phosphate or ammonium polyphosphate as phosphorus sources. In the present trial some

1.75 g of the total 2.5 g P/day intake was as a mineral

supplement where as the ARC (1965) recommended minimum intake of 2.9 g P/day (for 35 kg sheep gaining at 0.17 kg/day) presumably anticipates a greater proportion of the total dietary phosphorus coming from vegetable sources.

It is interesting to note that where both nitrogen (as urea) and phosphorus (as either DCP, Mg P or Ca Mg P) were provided, the supplemented diets were invariably well consumed. However, where nitrogen alone was added to the basal low phosphorus diet (Diet A) three of the five sheep left appreciable amounts of SEP and/or chopped hay. Very similar reductions in appetite associated with a low dietary intake of phosphorus were also observed in Experiment 2.1 (Section 2).

The basal diet provided a mean intake of 1.4 g Mg/day which was considerably higher than the ARC (1965) recommended requirement of 0.7 g Mg/day for 35 kg sheep growing at about 0.17 kg/day. When fed the unsupplemented diet the sheep apparently retained 0.10 mg/day which was somewhat surprising as they were in small negative phosphorus balance (-0.17 g P/day) and only small positive calcium balance (+0.04 g Ca/day). Supplementation with 0.57 g Mg/day (as either MgO + DCP or as Ca Mg P) increased the apparent magnesium retention to about 0.33 g Mg/day. Further supplementation with 1.89 g Mg/day (as either MgO + DCP or as Mg P) further increased the apparent retention to about 0.43 g Mg/day. With each supplementation there were also increases in plasma magnesium concentration.

Growing sheep cannot continue to retain as much as 0.3 -0.4 g Mg/day as their total body content (in the order of 360 mg/kg) represents only about 13 g Mg for a sheep of 35 kg liveweight (ARC, 1965). Similarly high rates of apparent magnesium retention by sheep have been previously recorded; eg. Hemingway and Brown (1967) found that 0.3 - 0.4 g Mg/day were

retained when growing sheep were fed 1.65 g Mg/day including 1.25 g Mg present as magnesium ammonium phosphate and Suttle and Field (1966) described daily retentions of 0.26 g Mg of a total of 1.8 g Mg fed as hay and concentrates. Ammerman, Chicco, Loggins and Arrington (1972) have reported that sheep fed a low magnesium diet supplemented with either reagent grade magnesium carbonate, oxide or sulphate to provide a total intake of 0.60 g Mg/day retained 0.10 - 0.15 g Mg/day. Rook, Balch and Line (1958) have also indicated that apparent retentions of about 2 g Mg/day determined for lactating cows could not be continued over long periods.

It is unlikely that there were systematic errors in the measurement of feed inputs and collection of excreta as both the phosphorus and calcium data seem reasonable. It is possible, but very unlikely, to have an analytical error which may contribute towards the large apparent retentions. It is also most unlikely that large amounts of magnesium could be temporarily retained in the gut. The situation could perhaps be resolved by employing a comparative slaughter technique.

It is concluded that under the conditions of this experiment both magnesium phosphate and a calcium magnesium phosphate were as well utilised by growing sheep as were combinations of dicalcium phosphate and magnesium oxide.

#### Experiment 3.2

The supplementary feeding of magnesium phosphate to lactating beef cows at spring grass.

The results of Experiment 3.1 indicated that magnesium phosphate (Mg P; 20.0 % Mg, 18.5 % P) was a satisfactory source of both phosphorus and magnesium for growing sheep. In comparison with magnesium oxide, supplementation with magnesium phosphate maintained a slightly higher mean blood magnesium concentration and daily retention of magnesium, which would suggest that magnesium phosphate was a rather more available source of dietary magnesium.

Calcined magnesite which contains about 87 % MgO is frequently given as a supplement to lactating cattle grazing lush spring and autumn pasture, principally as prophylactic treatment against hypomagnesaemic tetany. It is generally recommended that 2 ozs. of calcined magnesite (= 30 g magnesium) should be fed per day and this is normally given in some form of concentrate feed. However, although calcined magnesite supplementation may reduce the number of clinical cases of hypomagnesaemic tetany it may not eliminate the incidence completely. Furthermore, mineral feeds which contain calcined magnesite are often unpalatable and are not readily consumed by stock and accordingly it is frequently contained in about 1.5 - 2.0 kg concentrate feed.

Magnesium phosphate could be used as an alternative magnesium supplement for cattle at grass. The phosphorus content of magnesium phosphate however, is such that it would be used primarily as a phosphorus source and necessarily the quantity of magnesium supplied would be determined by the degree of required phosphorus supplementation.

Under most grazing conditions only the best young herbage is likely to contain more than 0.35 % P. Concentrations of about

0.20 - 0.30 % P are more likely. Where lactating cattle graze pastures which contain between 0.20 - 0.30 % P it is generally considered that a supplement of about 10 g P/day would provide sufficient additional dietary phosphorus to meet the requirements for maintenance and milk production.

The present experiment was designed to evaluate magnesium phosphate as a combined phosphorus and magnesium supplement for lactating cows at spring grass. The intention was that a supplement of 53 g Mg P/day would be provided. This quantity of magnesium phosphate would supply an additional 10 g P and 11g Mg/ day.

#### Materials and Methods.

The experiment was conducted during spring 1973 at Garscube Estate, Glasgow University Veterinary School. The stock available for the experiment was a herd of 27 beef cows with suckling calves. All the cows had been housed during the preceding winter feeding period. During the early stages of pregnancy the cows had been maintained on a diet of low-protein catstraw and molassed sugar beet pulp variously supplemented with urea and phosphorus. A more detailed account of their nutrition during pregnancy is given in Section 4. After calving and until turnout to grass, poor quality hay (about 7 kg/day) and molassed sugar beet pulp (about 3 kg/day) containing 2.8 % urea and 3.0 % dicalcium phosphate were fed to all the cows.

Calving took place during the period 9th February (9.2) to 26th May (26.5), but because of the range in calving dates not all the cows were put out to pasture on the same date. Sixteen of the earlier calved cows were turned out to grass on 11th May. They were divided according to their date of calving into two groups of eight. The allocation was such that the distribution of individual calving dates in each group was

similar. Each group of cows (control and treated) were grazed in separate but adjacent fields throughout the experiment. On 28th May four later calving cows were introduced to the control and treated groups and subsequently on 4th June a further three cows were put with the control group.

The unsupplemented concentrate fed to the control group of cows was composed of 89 % ground barley and 10.5 % shredded molassed sugar beet pulp to which was added 0.5 % salt prior to cubing. The supplemented concentrate (fed to the treated group of cows) contained in addition 5.3 % of magnesium phosphate. The respective concentrates were fed as appropriate at 1 kg/head/day and accordingly where the supplemented concentrate was given this provided 10 g P and 11 g Mg/day as magnesium phosphate.

Heparinised blood samples for the determination of magnesium, phosphorus and calcium were obtained from the cows immediately before they were put out to grass and at 3 - 4 day intervals during the course of the experiment. Blood samples were obtained on only one occasion (7 days after turnout) from the 3 cows which were introduced to the control group on 4th June. Representative grass samples were taken from each field on 21st May. Laboratory analysis indicated that the herbage grazed by the control cows contained 0.15 % Mg, 0.34 % P and 0.55 % Ca (DM basis) and that grazed by the treated group 0.15 % Mg, 0.30 % P and 0.62 % Ca (DM basis).

# Results (Tables 15, 16 and 17.)

The individual and mean blood magnesium, phosphorus and calcium concentrations which are presented in Tables 15, 16 and 17 have been arranged on a basis of the number of days from turnout to grass.

#### Blood magnesium.

Both the control and the treated cows were initially

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.

Table 15. Blood magnesium concentrations (mg/100 ml) of the two

groups of suckled beef cows at grass.

		0	-1				0			
Cont	rol Cows				Days	after T	urnout			
No.	Calving									
	date	0	3	7	11	14	18	21	25	28
10	9.2	1.87	2.02	1.91	1.25	0.50	0.42	0.42	0.44	0.42
3	26.2	1.76	1.71	1.76	2.02	1.81	1.81	1.97		1.87
7	2.3	1.60	1.48	1.87	2.14	1.65	2.10	1.97	1.55	2.08
25	12.3	1.97	1.65	1.76			om the			0 51
16 21	15.3 20 <b>.</b> 3	1.91 2.08	$1.00 \\ 1.63$	0.80 1.87	0.67 1.45		0,32* 2,02	0.44 2.08	0.59 2.02	0.54 2.19
13	4.4	1.60		1.02	1.02		1.65			1.20
4	7.4	1.35	1.33	1.35	1.11	0.80		0.42	-	0.67
·								•		•
15 ·	22.4	2.19	1.87	1.55	1.45	-	-		-	
24	2.5	2.32	1.65	0.98	1.45	-	<b>4</b> 20		-	-
22	6.5	2.14	1.40	1.25	1.25	-	-	-		-
17	18.5	2.14	1.57	1.45	1.87	-	-	-	~	-
20	26.5			1 10						
20 23	26.5	-	-	1.40 0.84	-		-	-	-	-
27	26.5	-		0.84 0.27*	· _	-	_	_	-	-
	,									
Mean		1.91	1.51	1.34	1.42	1.17	1.30	1.29	1.16	1.29
S.E. mean		0.083	0.100	0.124	0.133	0.240	0.290	0.307	0.249	0.288
mean	(-)									
Trea	ted Cows									
28	6.2	1.81	2.38	2.14	2.02	1.87	1.57	1.65	1.55	1.60
5	2.3	1.97	2.29	2.19	2.52	2.26	2.26	2.19	2.14	2.19
12	4.3	2.14	2.19	1.87	1.91	1.81	2.14	2.19	2.14	1.97
11	15.3	1.97	2.08	2.26	2.02	2.19	1.89	1.87	1.62	1.87
2 1	20.3	1.40	1.73	1.30	1.65	2.02	1.97	1.97	1.76	1.91
9	23•3 2•4	2.08 1.76	2.45 2.00	2.19 2.08	2.08 1.97	1.76 2.26	1.60 2.32	1.62 2.57	1.97 2.14	1.97 2.19
6	5.4	1.71	2.49	2.08	2.52	2.32	2.14	2.38	2.26	245
	•••			-						
8	18.4	2.26	2.02	2.26	2.52	-	-	-	- '	-
26	1.5	2.32	2.08	2.02	2.14		-	-	-	
32	2.5	2.38	1.81	2.45	1.91	-	-	-	+	
18	25.5	2.19	1.65	1.76	1.91	-	-	-	-	••
Mean		2.00	2.10	2.05	2.10	2.06	1.99	2.05	1.95	2.02
S.E.		0.083	0.079	0.086	0.081	0.079	0.100	0.119	0.095	0.090
mean	$(\underline{T})$							~		•••
Sign	ificance	N.S.	0.001	0.001	0.001	0.005	0.05	0.05	0.01	0.05
Numbe	er <									
	Mg/100ml		,			,	_ 4		,	
		0/12	1/12	4/15	1/12	3/7	3/7	3/7	3/7	3/7
Cont	Mg/100ml	0/12 0/12	1/12 0/12	4/15 0/12	1/12 0/12	3/7 0/8	3/7 0/8	3/7 0/8	3/7 0/8	3/7 0/8

\* Indicates cases of hypomagnesaemic tetany.

reasonably similar with regard to mean blood magnesium

concentrations (1.91 and 2.00 mg Mg/100 ml for the control and treated groups respectively) and to the spread of their individual values. During the first 14 days of the experiment the mean blood magnesium concentration of the control cows declined progressively to a mean level of 1.17 mg/100 ml and remained low at about 1.16 -1.30 mg Mg/100 ml until the end of the experiment.

Supplementation with magnesium phosphate maintained the mean blood magnesium concentration of the treated animals within the range 1.99 - 2.10 mg Mg/100 ml throughout the experiment. On sampling days 3, 7 and 11 after turnout to pasture, the mean blood magnesium concentrations for the magnesium phosphate treated cows were 2.10, 2.05 and 2.10 mg Mg/100 ml respectively and were significantly greater (P<0.001) than the mean concentration of 1.51, 1.34 and 1.42 mg Mg/100 ml recorded for the control cows which received the unsupplemented concentrate. Indeed, on each occasion that blood samples were obtained during the experiment, the mean blood magnesium concentration of the treated cows was always significantly greater than that of the control group.

Among the control cows there were several individuals which had very low blood magnesium concentrations. On the second sampling date, seven days after turnout, there were 4 cows (of the 15) in the control group which had blood magnesium concentrations of less than 1 mg Mg/100 ml. Two days later one of the cows (no.27) developed hypomagnesaemic tetany and subsequently died. The last recorded blood magnesium concentration for cow no.27 (two days earlier) was 0.27 mg Mg/100 ml. On each of the subsequent sampling occasions 3 cows (of the 7 which were still being sampled) consistently had blood magnesium concentrations of 0.8 mg Mg/100 ml or less. Number 16 exhibited clinical symptoms of hypomagnesaemic tetany (eg. muscular tremors and an exaggerated gait) on Day 18.

but recovered after being given a subcutaneous injection of 400 ml 25 % magnesium sulphate solution. Her blood magnesium concentration on that day was 0.32 mg Mg/100 ml.

In comparison, none of the cows fed magnesium phosphate showed any symptoms of hypomagnesaemic tetany and no individual had a blood magnesium concentration of less than 1.30 mg Mg/100 ml. <u>Blood phosphorus</u>.

Both the control and treated groups had an initial mean blood phosphorus concentration of about  $6\cdot28 - 6\cdot29$  mg P/100 ml, and there was a comparable range of individual values within each group. During the first few days of the trial the mean blood phosphorus concentration of the control cows declined and by Day 3 and 7 had reached a mean level of about  $5\cdot03$  mg P/100 ml. On both these sampling dates the mean blood phosphorus concentrations of the treated cows were significantly greater at about  $6\cdot03$  and  $6\cdot17$  mg P/100 ml respectively. Thereafter the mean blood phosphorus concentration of the control group tended to increase to about  $5\cdot80$  mg P/100 ml and on each of the subsequent sampling occasions there were no significant differences between the control and treated cows in respect of mean blood phosphorus concentration. Blood calcium.

The mean blood calcium concentrations of both the control and treated animals were essentially similar throughout the experiment and were within the normal range 9.66 to 10.75 mg Ca/100 ml. Blood calcium concentration was apparently unaffected by magnesium phosphate supplementation. The onset of hypomagnesaemic tetany in cow no.16 was associated with a decline in blood calcium concentration to about 8.20 mg Ca/100 ml, but there was no comparable reduction in the blood calcium concentration of the other 2 cows (nos. 10 and 4) which were consistently hypomagnesaemic during the final 14 days of the experiment.

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Table 16. Blood phosphorus concentrations (mg/100 ml) of the two

groups of suckled beef cows at grass.

Conti	rol Cows				Days	after T	urnout			
No.	Calving date	0	3	7	11	14	18	21	25	28
10 3 7 25 16 21 13 4	9.2 26.2 2.3 12.3 15.3 20.3 4.4 7.4	6.55 6.55 6.25 5.30 6.85 5.30 8.05	5.05 6.40 4.75 5.00 4.35 4.10 2.95 5.75	3.25 5.20 4.60 5.35 3.40 3.25 2.20 5.95	5.90 7.65 5.70 withd 5.10 7.00 5.30 5.95	6.90 6.35 rawn fr 4.00 5.95 4.80 5.60	5.75 6.50 7.45 om the 5.10 6.60 7.05 7.05	4.75 7.30 5.50 experim 5.00 5.85 4.25 5.00	6.75 6.75 6.85 eent 4.35 5.00 3.80 7.35	6.65 6.80 5.20 4.68 5.95 5.40 6.45
15 24 22 17	22.4 2.5 6.5 18.5	6.50 6.55 4.80 6.25	5.20 5.95 5.20 5.85	5.30 6.25 5.75 6.00	5.50 7.15 5.15 5.00	- - -	- - -	- - -	- - -	- - -
20 23 27	26.5 26.5 26.5			5.45 6.20 7.25	-			- - -	- - -	
Mean	~ f*	6.29	5.04	5.03	5.94	5.78	6.50	5.38	5.84	5.87
S.E. mean		0.245	0.271	0.363	0.276	0.409	0,310	0.374	0.535	0.305
mean		0.245	0.271	0.363	0.276	0.409	0,310	0.374	0.535	0.305
mean	(±)	6.15 7.55 5.30 5.40 6.15 7.25 6.35 5.60	5.30 6.10 7.10 5.85 6.80 6.00 6.40 4.35	6.30 4.50 7.20 5.25 4.60 5.00 6.55 6.70	4.50 4.70 8.05 5.70 6.95 6.05 6.20 6.45	0.409 5.65 5.15 6.90 4.80 6.20 5.75 6.50 7.95	4.60 6.60 6.25 6.40 4.95 5.25 7.70 5.70	5.20 4.95 5.30 5.50 5.30 6.35 6.15	4.20 4.70 5.15 5.85 4.40 5.15 6.20 5.20	5.00 5.55 5.55 6.54 4.95 5.30 5.70 5.00
mean Treat 28 5 12 11 2 1 9	(±) ted Cows 6.2 2.3 4.3 15.3 20.3 23.3 2.4	6.15 7.55 5.30 5.40 6.15 7.25 6.35	5.30 6.10 7.10 5.85 6.80 6.00 6.40	6.30 4.50 7.20 5.25 4.60 5.00 6.55	4.50 4.70 8.05 5.70 6.95 6.05 6.20	5.65 5.15 6.90 4.80 6.20 5.75 6.50	4.60 6.60 6.25 6.40 4.95 5.25 7.70	5.20 4.95 5.30 5.50 5.50 5.30 6.35	4.20 4.70 5.15 5.85 4.40 5.15 6.20	5.00 5.55 5.55 6.54 4.95 5.30 5.70
mean Treat 28 5 12 11 2 1 9 6 8 26 32	(±) ted Cows 6.2 2.3 4.3 15.3 20.3 23.3 2.4 5.4 18.4 1.5 2.5 25.5 of	6.15 7.55 5.30 5.40 6.15 7.25 6.35 5.60 5.45 5.45 7.00	5.30 6.10 7.10 5.85 6.80 6.00 6.40 4.35 4.70 6.80 6.80	6.30 4.50 7.20 5.25 4.60 5.00 6.55 6.70 5.40 6.25 9.85	4.50 4.70 8.05 5.70 6.95 6.20 6.45 3.90 4.50 5.20	5.65 5.15 6.90 4.80 6.20 5.75 6.50 7.95	4.60 6.60 6.25 6.40 4.95 5.25 7.70	5.20 4.95 5.30 5.50 5.50 5.30 6.35	4.20 4.70 5.15 5.85 4.40 5.15 6.20	5.00 5.55 5.55 6.54 4.95 5.30 5.70 5.00

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Table 17. Blood calcium concentrations (mg/100 ml) of the two

groups of suckled beef cows at grass.

Cont	rol Cows				Days a	fter Tu	rnout			
No.	Calving date	0	3	7	11	14	18	21	25	28
10 3 7 25	9•2 26•2 2•3 12•3	10•30 9•90 9•60 9•90	9•90 10•10 9•75 10•50	10•70 11•00 9•90 10•30			8•90 10.00 9•60 m the e			9•90 10•30 11•00
16 21 13 4	15•3 20•3 4•4 7•4	9.60 9.60 10.70 9.60	8.45 9.10 10.30 9.90	9.90 9.60 11.40 10.70	10.30 8.60 12.90 10.30	9.90 10.30 11.40 11.00	8.20 10.30 10.70 9.90	8.90 9.90 10.30 9.60	10.30 9.90 10.70 9.90	9.60 9.60 11.40 10.30
15 24 22 17	22.4 2.5 6.5 18.5	10.70 11.00 10.30 9.90	10.30 9.90 10.10 10.70	11.65 10.30 10.70 10.70	10.30 10.30 10.30 10.30		- - -		  	
20 23 27	26.5 26.5 26.5		-	9.90 10.30 9.30	- - -	- - -	- - -	-	-	
Mean S.E. mean	of	10.09 0.144	9.92 0.179	10.42 0.168	10.72 0.346	10.75 0.245	9.66 0.323	9.76 0.183	10.29 0.140	10.30 0.260
Trea	ted Cows						•			
28 5 12 11 2 9 6	6.2 2.3 4.3 15.3 20.3 23.3 2.4 5.4	10.70 9.90 9.60 9.30 10.30 10.30 10.30	9.60 9.75 9.75 9.75 9.75 10.10 9.75 9.10	10.30 10.30 10.70 9.60 9.90 11.00 9.90 10.70	8.60 10.30 14.50 11.00 11.00 10.70 10.30 10.70	10.30 9.90 10.90 10.30 10.70 10.70 10.70 9.90	9.70 9.90 9.80 9.80 9.60 10.30 10.30 10.30	9.70 9.60 10.30 10.10 9.90 10.70 9.30 9.60	11.40 10.30 10.30 10.70 10.55 11.00 10.30 10.30	10.30 10.30 9.90 10.30 10.70 10.70 10.30 10.30
8 26 32 <b>1</b> 8	18.4 1.5 2.5 25.5	9.30 13.30 10.30 10.30	9.90 10.50 9.90 9.90	10.30 10.30 9.30 9.90	9.90 9.90 9.90 9.90		-	- - -		
Mean S.E. mean		10.29 0.305	9.81 0.093	10.18 0.142	10.56 0.405	10.42 0.105	9.96 0.104	9.90 0.159	10.61 0.135	10.35 0.091
Sign:	ificance	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.	N.S.

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# Discussion and Conclusions.

In experimental work with magnesium supplements the efficacy of a particular source of magnesium is determined by the ability of the material to maintain normal blood magnesium concentrations  $(1 \cdot 8 - 2 \cdot 5 \text{ mg Mg}/100 \text{ ml})$  and to eliminate (or reduce) the incidence of hypomagnesaemic tetany. In the present experiment the mean blood magnesium concentration of the control animals fell markedly when they were first put out to grass. The overall mean blood magnesium concentration of the control group was 1.38 mg Mg/100 ml, but there were several individual animals which had extremely low blood magnesium concentrations (< 0.6 mg Mg/100 ml) and eventually hypomagnesaemic tetany occurred in two cows whose blood magnesium concentration fell to less than 0.4 mg Mg/100 ml. This type of situation represents a severe test of a magnesium supplement. However, where magnesium phosphate was given a mean blood magnesium concentration of 2.00 mg Mg/100 ml was maintained throughout the 28 day experimental period, demonstrating the effectiveness of this material as a magnesium supplement. It is of particular significance to note that only 11 g/day of supplementary magnesium were given as magnesium phosphate. This is considerably less than the standard recommendation that 2 ozs. of calcined magnesite (= 30 g Mg) should be given per day and clearly in the present experiment such a large quantity of supplementary magnesium was not required.

It is unlikely that many of the beef cows in the trial were producing in excess of 10 kg of milk/day. The provision of 11 g of supplementary magnesium per day is closely similar to the minimum recommended intake suggested by the ARC (1965) for this level of production and is of course in addition to the small amount of magnesium supplied by the basal concentrate mixture (barley and molassed sugar beet pulp) and that obtained from the herbage.

The herbage grazed by the control and treated groups of animals contained  $0.34 \ \%$  P and  $0.30 \ \%$  P (DM basis) respectively and would not be considered deficient in phosphorus. Nevertheless, the provision of 10 g P/day as magnesium phosphate prevented a reduction in mean blood phosphorus concentration immediately after turnout to pasture and confirms the results of the previous experiment (Experiment 3.1) that magnesium phosphate provides a readily available source of dietary phosphorus.

#### SECTION 4

THE EFFECTS OF UREA AND PHOSPHORUS CONTAINED IN MOLASSED SUGAR BEET PULP ON THE VOLUNTARY INTAKE AND DIGESTIBILITY OF LOW-PROTEIN ROUGHAGE FEEDS BY CATTLE.

## Introduction.

One of the major uses of urea in diets for ruminants is as a low-cost nitrogen supplement for poor-quality roughage feeds. The voluntary consumption of poor roughage materials by ruminants is often limited, partly by the capacity of the rumen, but perhaps of more importance by the slow rate of digestion and consequent reduced rate of disappearance of digesta from the rumen when these materials are fed (e.g. Campling, Freer and Balch, 1962). The rate of digestion of roughage material in the rumen is related primarily to the activity of the various micro-organisms responsible for cellulose digestion. Ammonia nitrogen is essential for the growth and multiplication of these cellulolytic micro-organisms. Frequently the protein content of poor-quality roughage feeds is either too low or unavailable to achieve the ammonia-N concentration required for active microbial growth. Necessarily, the digestion and voluntary intake of roughage DM is impaired.

Many experiments have shown that the addition of non-protein nitrogen (or protein) to poor-quality roughage feeds markedly increases their daily intake by livestock. In many but not all of these experiments the increases in voluntary intake have been accompanied by increases in the digestibility of the dry matter and hence digestible energy intake. Hemsley and Moir (1963) in Australia, found that the addition of 3 % urea to hay (4.6 % CP) increased the intake of DM by sheep from 613 to 839 g DM/day (+54 %). Armstrong and Trinder (1966) (quoting results of Lamb) reported that with sheep, the addition of 1 % urea to straw (4 % CP), to raise the overall CP content of the diet to about 7 %, increased the voluntary consumption of straw DM from 456 to 532 g/day (+ 17 %) and the digestibility of the DM from 47.0 to 51.7 %. The intake of digestible DM was accordingly increased by some 29 %. There was also a corresponding improvement in nitrogen balance from -1.44 g N/day to -0.68 g N/day.

In many areas of the world the intake of poor range-type vegetation is similarly restricted by an inadequate nitrogen (and possibly phosphorus) intake. Elliot and Topps (1963) have reported that for veld grass (Hyparrhenia spp. tall grass) in Rhodesia, there was a significant correlation between the crude protein content  $(x_{\lambda})$ and dry matter intake (y g/kg  $W^{0.73}$ ) by sheep i.e. y = 26.22x + 24.47. Smith (1962) added either urea or groundnut meal to mature Hyparrhenia forage (3% CP) and showed that both materials increased the intake of forage DM by cattle by 40 - 60 %. Altona (1966) measured the live-weight gain of cattle fed veld hay and supplementary urea given as a urea/molasses lick. Where the hay contained 5.6 % CP addition of urea improved the live-weight gain of the steers by 0.62 kg/day compared with an improvement of only 0.12 kg/day recorded when hay containing 9.6 % CP was given. Intermediate growth rates were obtained with hay containing 6.1 and 7.7 % CP. Altona (1966) commented that the results of the trial indicated that "the poorer the hay being fed the better the response to urea supplementation."

In more controlled experiments Campling et al. (1962) gave long oatstraw (3 % CP) ad libitum to adult cows either alone or supplemented with up to 150 g urea/day given by continuous infusion per rumen fistula. The mean intake of about 5.5 kg of cat straw DM (unsupplemented) was increased by 43 % when 75 - 150 g urea/day were given and by 26 % when 25 g urea/day was provided, but was not further increased by the addition of 500 g sucrose/day. There was

an accompanying increase in the digestibility of the dry matter with increased straw intake from about 42 to 50 %. This was mainly due to an increased digestibility of the crude fibre and N-free extract fractions of oat straw. In unusual contrast, similar infusion of urea by Minson and Pigden (1961) had no consistent effect on the digestibility of chopped oat straw but decreased voluntary straw intake by 12 %.

In many of the experiments where urea supplementation has increased the intake of low-protein roughage dry matter this most probably resulted from improved cellulolytic activity of the rumen microflora when given an increased supply of nitrogen (Campling et al. 1962; Hemsley and Moir, 1963). Nevertheless, recent evidence would also suggest that the increased voluntary intake of poorquality roughages by ruminants given nitrogenous supplements may also be due in part to the improved protein-status of the animal as a result of the increased absorption of amino acids into the body i.e. to a metabolic effect (Egan 1965ab; Weston 1966).

Many other varied examples of the effect of urea on increasing the intake of low-protein roughage DM could be quoted, but in each case the experimental findings depend on the particular circumstances of the trial. No increase in the voluntary consumption would normally be expected when urea is added to roughage feeds such as hays and straws which contain more than about 6 - 8 % CP in the dry matter. Equally, in assessing the effects of urea on voluntary intake and digestibility, account must be taken not only of the crude protein and crude fibre contents of the roughage material, but also the amount of crude protein supplied in the concentrate or other medium in which the urea is contained. This may explain the observations of Kay, Andrews, MacLeod and Walker (1968) who recorded that the mean daily intake of 7.5 kg barley straw (5.7 to 6.0 % CP in DM) by 450 kg in calf-suckler

cows additionally fed 2 kg barley (10 % CP) was not increased by the inclusion of 50 g urea/day in the barley. It should be noted that the barley alone would have provided 200 g of supplementary crude protein per day, which would be in addition to the 150 g CP/day supplied by the added urea. O'Donovan (1968) employing virtually the same treatments as Kay et al. (1966) with 210 kg beef steers found no improvement in straw intake (4 % CP in DM) when either 75 g urea/day were sprayed on to the straw or when additional barley (1.36 kg/day) was provided with or without added urea. All the animals receiving barley, however, made markedly better live-weight gains than those fed straw or straw plus urea. O'Donovan (1968) reported that where barley was provided the animals maintained "body condition."

A further complicating factor concerned with the voluntary consumption of low-protein roughage feeds, is that in addition to being deficient in nitrogen, many poor-quality roughage materials also contain low concentrations of phosphorus. Smith and Comrie (1948) and more recently Hemingway et al. (1968) have shown that there is a significant correlation between the crude protein and phosphorus content of hay (and silage), over the range of about 5 - 15 % CP. Of the hay samples analysed by Hemingway et al. (1968), 25 % contained less than 5 % CP in the dry matter and almost two thirds less than 0.20 % P. Many oat and barley straws also contain very low concentrations of crude protein (c.3 % CP) and phosphorus (c.0.15 % P). Where urea is added to such feeds to increase voluntary intake, phosphorus supplementation would be necessary if recommended (ARC, 1965) intakes of P were to be maintained. It is uncertain what effects the differential supplementation of low-protein roughage feeds with urea and/or phosphorus would have on appetite, but this would be of considerable interest when molassed sugar beet pulp (c.0.08 % P) is also included

in the diet. Kleiber, Goss and Guilbert (1936) recorded that growing heifers given a low P diet (0.08 %) eventually had only 60 % of the voluntary appetite of those fed a diet adequate in P (0.40 %). They were, however, unable to record any changes in the utilisation of the digestible energy or in the apparent digestibility of the crude protein intake. In contrast, the provision of a phosphate lick has been found to increase the consumption of a poor forage (Brünnich and Winks, 1931). Similar observations were made by Little (1968) and Playne (1969) who increased the intake of low-phosphorus lucerne (0.078 % P) by sheep by the provision of extra phosphorus. Results presented elsewhere in this Thesis (Experiments 2.1, Section 2 and Experiment 3.1, Section 3) have shown that additional phosphorus (1.75 g P/day) in a variety of forms (DCP, Urea P and MAP in Experiment 2.1: DCP, MgP and Ca MgP in Experiment 3.1) increased the voluntary consumption by growing wether sheep of a diet consisting predominantly of urea-supplemented molassed sugar beet pulp (0.08 % P). Where supplementary urea alone was added to the basal low phosphorus diet the mean daily intake of DM was about 7 % (Experiment 2.1) and 14 % (Experiment 3.1) less than where both nitrogen and phosphorus supplements were provided. The mean intake of DM from the basal unsupplemented diet when neither additonal nitrogen nor phosphorus were given was reduced by about 10 % (Experiment 2.1). These changes in appetite generally occurred within a very short period after the introduction of a low P diet (7 - 10 days), but food intake was restored within 2 - 3 days of subsequent change to full P supplementation. There was little evidence from these two experiments to suggest that the dry matter digestibility of the molassed sugar beet pulp was adversely affected by a low P intake. On the other hand no decrease (or increase) would normally be expected with either N or P supplementation of a material which

is already about 30 % digestible. Nevertheless, a reduction in diet digestibility may occur when a low P intake is continued over extended periods and in situations where poor-quality roughage materials of high fibre content are included as a major component of the diet. A low P intake would presumably result in low levels of P in the rumen. Evans and Davis (1966a) found that increasing dietary P from 0.04 to 0.16 and 0.54 % resulted in rumen P levels of 198, 417 and 543 µg/ml respectively. Phosphorus has also been shown to be essential for cellulose digestion (Anderson, Cheng and Burroughs 1956; Chicco, Ammerman, Moore, van Walleghem, Arrington and Shirley, 1965; Evans and Davis, 1966b). Rumen bacterial cells contain 2 - 6 % P on a dry weight basis (Hungate, 1966) so the growth rate of these cells may well be affected by low levels of P in the diet.

## Experiments 4.1 and 4.1a

The comparative effects of two levels of urea included in molassed sugar beet pulp on the voluntary intake and digestibility of oat straw by steers.

These investigations were undertaken to evaluate the effect of urea contained in molassed sugar beet pulp on the voluntary consumption of low-protein oat straw. Experiment 4.1 was concerned with the effects of inclusion of (a) 3.0% and (b) 7.8% urea given in 1 kg/day of molassed sugar beet pulp on the ad libitum intake of oat straw by groups of 300 kg steers. In Experiment 4.1a comparable diets were fed to steers to assess the effects on the digestibility of the oat straw.

## Experiment 4.1 Straw consumption trial

#### Materials and Methods.

Oat straw was given ad libitum to six balanced groups each of four British Friesian steers aged about 15 months and mean liveweight 296 kg. Three molassed sugar beet pulp products (SBP) were fed, as appropriate, at 1 kg/head once per day. The composition of the oat straw and the three cubed (c.1.27 cm diameter, c.2.5 cm length) SBP products are detailed in Table 18. The oat straw contained 1.96 % crude protein (CP) and 45.1 % crude fibre (CF) in the dry matter. Each of the SBP products contained

# Table 18. Experiments 4.1 and 4.1a Composition of the oat straw and the molassed sugar beet pulp products (% dry matter basis).

Diet Supplement	Straw <sup>+</sup>	A Nil	B Urea	C Urea
Crude protein	1.96	10.06	18.97	36.62
Crude fibre	45.12	13.85	13.06	11.68
Ether extract	1.24	0.41	0.44	0.86
N-free extract	45.83	65.61	57.02	39.01
Organic matter	93.98	89.93	89.49	88.17
Ash	5.85	10.07	10.51	11.83
Calcium	0.28	1.77	1.79	1.72
Phosphorus	0.08	0.58	0.54	0.57
<sup>+</sup> Mean of 10 samp	Les			

3 % dicalcium phosphate (to give about 1.76 % Ca and 0.56 % P in DM) plus trace elements and vitamins A and D. The amounts of urea and crude protein (DM basis) contained in each of three cubed materials were: Diet A, no urea, 10.06 % CP; Diet B, 3.0 % urea, 18.97 % CP; Diet C, 7.8 % urea, 36.62 % CP. In total (B) and (C) supplied about twice and three and a half times more additional crude protein than did (A). It was anticipated that the total daily phosphorus intake from the supplemented SBP products and the oat straw (0.08 % P in DM) would be about 10 g P/day, which satisfies the minimum requirement suggested by the ARC (1965) for cattle growing at low rates.

Following a preliminary period of 1 month when oat straw and Diet B were given, the six groups of four steers were arranged in two separate 3 x 3 Latin squares so that each group received 1 kg/ head/day (= 0.88 kg DM) of each SBP product in turn. Oat straw was available ad libitum (from racks where suitable precautions were taken to minimise wastage). The oat straw was replenished twice daily or as required so that at least 25 % more than the anticipated daily intake was always available. Each feeding period lasted 21 days and separate records were kept of the straw consumed by each group of four steers over days 0 to 7 (except during Period 1), days 8 to 14 and days 15 to 21 of each feeding period. Straw residues were collected daily or when necessary and representative samples taken for dry matter analysis. The steers were weighed on the first and last day of the experiment.

## Results (Table 19).

The molassed sugar beet pulp products were well consumed and there were no palatablility problems associated with the inclusion of either 3.0 or 7.8 % urea in products B and C. During the first 7 days after a change of diet the amounts of oat straw eaten by the steers given Diets A, B and C (mean of Periods 2 and 3 only) were 4.88, 4.53 and 5.16 kg DM/day respectively and there were no real. differences in the voluntary consumption of straw DM which could be attributed to the provision of supplementary urea. The amounts of straw DM consumed by the steers fed Diet A (no urea) declined to a mean level of about 4.30 kg DM/day during the periods 8 to 14 days and 15 to 21 days after each change of diet. The addition of 30 g urea (Diet B) and 78 g urea (Diet C) to the SBP significantly (P $\leq 0.05$ ) increased the intake of straw DM to a mean of 5.04 - 5.31 and 5.18 - 5.25 kg DM/day respectively during the second and third weeks of each feeding period. Overall, the provision of either 30 or 78 g of supplementary urea per day increased the intake of oat straw DM by about 20%. It is interesting to note that where supplementary urea was provided in products B and C, this markedly increased the rate at which the offered straw was consumed.

During the course of the 63-day experimental period the steers virtually maintained their initial liveweight (mean loss 3 kg).

Table 19. Experiment 4.1 The amounts of supplementary urea and total crude protein (g/day) supplied by the molassed sugar beet pulp products and the mean voluntary intakes of straw dry matter (kg/day).

Diet Supplement	A Nil	B Urea	Ç Urea	S.E. of mean( $\pm$ )	Significance
Urea supplied Total CP in SBP	0 89	30 169	78 <b>3</b> 22	-	-
Oat straw DM intake Days O - 7 † Days 8 - 14 Days 15- 21	4.88 4.28 4.32	4.53 5.04 5.31	5.16 5.25 5.18	- 0.183 0.244	- B, C>A* B, C>A*
		_			

Refers to Periods 2 and 3 only

# Materials and Methods.

In a parallel digestibility trial, three British Friesian steers (aged about 24 months and mean liveweight 385 kg) fitted with faecal collection harnesses (Balch, Bartlett and Johnson, 1951) were fed the same oat straw and 1 kg/day of the three SBP products given in Experiment 4.1 (Table 18). In a preliminary period of 2 weeks when 1 kg/day of the SBF product supplemented with 3.0 % urea (Diet B) was given to all the steers it was found that about 5.0 to 5.5 kg of oat straw DM offered ad libitum was consumed per day. It was thus decided in the present experiment to give controlled amounts of oat straw DM (5.02 kg DM/day) in two feeds each supplying 2.51 kg DM. The three SBP products were given to the three steers in sequence in a  $3 \times 3$  Latin square design with feeding periods of 21 days. Separate faecal collections were made during days 8 to 14 and days 15 to 21 of each feeding period to ascertain whether a 7-day change-over period was adequate to allow for complete adaption to a change of diet.

The separate 1 kg portions of the SBP products required by the three steers on each day during the 9 week period of the experiment were weighed  $(\pm 1 \text{ g})$  into appropriately labelled paper bags, using a Sauter 5 kg Self-indicating scale balance, on the day prior to the start of the experiment. At the same time representative samples of each product were obtained for laboratory analysis. For the oat straw, the most convenient procedure was to weigh the total of 14 portions required by each animal per 7 days on one day each week. Separate 3 kg (= 2.51 kg DM) portions were weighed ( $\pm$  10 g) into 400 gauge polythene bags (100 cm length x 60 cm width x 40 cm depth) using an Avery 36 kg balance (Model Pk-5u). Samples of oat straw were drawn for analysis on each weighing date.

Oat straw was given at about 9.00 hours and 16.00 hours each day. The appropriate SEP materials were fed once daily at about 8.00 hours. During the faecal collection periods when accurate measurements of straw intake were required, straw residues were removed prior to the morning feed and stored in polythene tags until the end of the recording period, when the total quantity of residues were weighed and samples drawn for dry matter analysis.

Complete collections of faeces were made from the 3 steers during each 7-day recording period. Faeces were collected directly into a 400 gauge polythene bag placed inside the rubberised faecal bag. The polythene bag was changed twice daily at 8.00 hours and 16.00 hours and the contents emptied into a large plastic bin. The total daily output of fresh faeces from each animal was weighed  $\binom{+}{-}$  10 g) in the plastic bin, spread on metal trays and a weighed 1 % sub-sample transferred to a polythene container intended to hold the 7-day bulked sample. The polythene containers, after sealing, were stored under refrigeration until subsequent analysis.

Blood samples were obtained for the determination of blood urea concentration 2 hours after feeding on the 21st day after each change of diet.

Food and faeces were analysed for dry matter, crude protein, crude fibre, ether extract and ash by standard analytical methods (see Appendix). Urea was also determined in the two SBP products B and C.

The digestibility data were analysed in respect of the digestibility of the complete diets and of the oat straw alone. The digestibility of the various components of the complete diets were calculated using Equation 1.

Equation 1 % Digestibility of 
$$x = 100 - \frac{100abc}{100de + fgh}$$

The appropriate input-output data were tabulated on a record sheet as detailed in Figure 2. These data were then analysed using a desk top computer (Olivetti 101, Milan, Italy) programmed with Equation 1. The read out from the computer gave a direct estimate of the digestibility of the various components of the complete diet. The digestibility of the oat straw alone was calculated from the total faecal output data after subtracting the amounts of faeces attributable to the SEP. In determining the quantities of faeces attributable to the SEP the following percentage digestibilities were assumed: dry matter 83.9, crude protein 62.3, crude fibre 91.4, ether extract 23.0, ash 48.2, N-free extract 89.6 and organic matter 83.2. These values have been shown (Parkins, 1972 and other unpublished data) not to be further increased by urea supplementation.

## Results (Tables 20 and 21)

It was noticeable in the present experiment that the inclusion of either 3.0 or 7.8 % urea in the SBP led to a more rapid consumption of the offered straw. The steers fed Diets B and C

Figure 2. An example of the tabulation of the input-output digestibility data from Experiment 4.1a

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			Result % Digestibility	58.78	39.63	62.38	23.80	46.39	59.91	59.50
		භ	h % fraction in straw DM	100.00	1.96	45.12	1.24	5.85	45.83	93.98
100 abc 100 de + fgh ? Period 1	2.	e % fraction in fresh SBP	87°70	16.87	11.86	0.21	8.41	50.35	79.29	
	Date 7 - 14 February 1972 Period 1	Q	c % fraction in faeces DM	100.00	6.68	36.77	2.01	8.33	46.21	9 <b>1.</b> 67
% Digestibility = 100 - Date 7 - 14 February 19	٢Ų	g % DM in oat straw	83.73						·	
% Dige	Date	4-	f g fresh oat straw /day	5891						
	·	ς	d g fresh SBP /day	1000						
	Д	N	b % DM in faeces	14.80						
rt	Diet	+-1	a g fresh faeces /day	16203	•					
Experiment 4.1a	Animal no 4	Entry no		Dry matter	Crude protein	Crude fibre	Ether extract	Ash	N-free extract	Organic matter

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almost completely consumed the 2.51 kg portions of straw DM given at each feed within 1.5 to 2.0 hours. The fixed amounts of straw which were offered however, were slightly in excess of full appetite and inevitably there were some small rejections and residues. In contrast, the steers fed the unsupplemented product (Diet A) were observed to eat their fixed ration of straw slowly over the whole day and a much smaller total quantity of straw DM was consumed. These differences in respect of both the rate and amounts of straw eaten were apparent within 4-5 days of a change of diet involving addition or withdrawal of urea.

Unfortunately, no faecal collections were made during days 8 to 14 of the first feeding period. The mean results found for the DM intake and digestibility of the various dietary fractions during days 8 to 14 (two observations) and days 15 to 21 (three observations) were closely similar. Statistical evaluation has therefore been made on the combined results of the five sets of data available for each dietary treatment.

The addition of 3 % urea (Diet B) and 7.8 % urea (Diet C) to the SEP significantly (P<0.01) increased the intake of oat straw DM by 14 and 17 % respectively. Both levels of urea inclusion in the SEP similarly and significantly increased the apparent digestibility of the dry matter and the crude fibre of both the combined diets and the oat straw alone. The higher and the lower levels of urea supplementation also increased the apparent digestibility of the organic matter of the straw alone, but when considered for the complete diet the lower level of urea supplementation failed to increase the digestibility of this fraction to a significant degree. Urea supplementation had no effect on the apparent digestibility of the N-free extract contained in either the oat straw or the entire straw plus SEP diets.

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Table 20. Intake of oat straw dry matter (kg/day) and urea (g/day) and the apparent digestibility (%) of the dry matter (DM), organic matter (OM), crude fibre (CF), N-free extract (NFE) and crude protein (CP) of the oat straw.

Diet	No.replicates	A	В	С	S.E. of	
Supplement		Nil	Urea	Urea	$mean(\pm)$	Significance
Urea supplied	-	-	30	78	<b>e</b> tra	-
Straw DM intake		•		I		,
Days 8-14	2 3 5	4.31	4.57	4.85	-	
Days 15-21 Combined mean	3	4.00 4.13	4.78 4.70	4.85 4.85	0.091	B,C>A**
Comprised modifi	2	4.1)	4•10	4.09	0.071	De C Minine
DM digestibility	·					
Days 8-14	2	43.8	50.5		-	-
Days 15-21	2 3 5		50.9			-
Combined mean	5	44.0	50.7	51.8	1.36	B,C>A**
OM digestibility						
Days 8-14	2	46.4	52.2	55.0	-	-
Days 15-21	3 5	47.4	52.1	52.4	-	-
Combined mean	5	47.0	52.1	53.5	1.61	B,C>A*
CF digestibility						
Days 8-14	2	50.5	57.0	60.0	-	_
Days 15-21	2 3 5	49.7		57.9	-	-
Combined mean	5	50.0		58.7	2.04	B,C>A*
NFE digestibility Days 8-14		46.0	50 1	52.0		
Days 15-21	~ ~ ~	47.3		48.6	_	-
Combined mean	2 3 5	46.7	50.9		1.55	N.S.
	-	4001	2007	2010	~ • / /	
CP digestibility						
Days 8-14	2	-57.4		63.6	-	-
Days 15-21 Combined mean	3 5	-35.9		57.6	- • • •	
comprised mean	2	-44.5	20.0	60.1	8.05	C>B**; B,C>A**

Table 21. Total intake of dry matter (kg/day) and urea (g/day) from the complete diet, mean blood urea concentration (mg/100 ml) and the apparent digestibility (%) of the dry matter (DM), organic matter (OM), crude fibre (CF), N-free extract (NFE) and crude protein (CP) of the complete diets.

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Diet	No replicates	А	В	C S	S.E. of	
Supplement		Nil	Urea	Urea	mean (±)	Significance
Urea supplied	-	-	30	78	-	-
Blood urea		4.7	8.4	18.7	1.10	B>A*; C>B>A***
Total DM intake Days 8-14 Days 15-21 Combined mean	2 3 5	5.19 4.87 5.00		5.70 5.74 5.73		 B, C>A****
DM digestibility Days 8-14 Days 15-21 Combined mean	2 3 5	50.6 50.4 51.6	55.8 56.0 55.9		-	 B, C>A*
OM digestibility Days 8-14 Days 15-21 Combined mean	2 3 5	52•5 53•7 53•1	57.0 56.8 56.9	59.2 56.9 57.8	-	 C > A*
CF digestibility Days 8-14 Days 15-21 Combined mean	2 3 5	52.9 52.3 52.6		61.5 59.4 60.2	81.0	- B, C>A*
NFE digestibility Days 8-14 Days 15-21 Combined mean	2 3 5	56.2 57.8 57.2	59.8 57.6 58.5	57.4 55.3 56.2	-	- N.S.
CP digestibility Days 8-14 Days 15-21 Combined mean	2 3 5	4.6 16.3 11.6		63.3 58.6 60.5	-	- B>A**; C>B>A***

The apparent digestibility of the crude protein fraction of the complete diets was increased from a mean of 11.6 % (Diet: no urea) to 33.5 % and 60.5 % when 30 g urea (Diet B) and 78 g urea (Diet C) were given per day. In respect of the oat straw alone, there was an improvement in apparent crude protein digestibility from -44.5% (Diet A) to +20.0% and +60.1% for Diets B and C respectively. The fact that the apparent crude protein digestibility was negative for Diet A, when the oat straw alone was considered, is not an unexpected result. With roughage feeds which contain as little as 1.96 % CP in the dry matter, the output of nitrogen in the faeces as metabolic faecal nitrogen (5.0 g N/kg DM intake, ARC 1965) greatly exceeds the total nitrogen intake and necessarily the determined crude protein digestibility will be negative. The increase in the digestibility of the straw alone noted when Diets B and C were given does not represent any real improvement in the digestibility of this fraction of the oat straw, but merely an apparent improvement, which results from including in the total CP intake the amounts of CP supplied by both the straw and additional urea. It must be pointed out that there was considerable variation in the apparent digestibility of the crude protein fraction of the oat straw and the complete diets between days 8 - 14 and days 15 - 21 of each feeding period, particularly when Diets A and B were given. This is probably attributable to animal variation in the loss of metabolic faecal nitrogen.

The mean concentrations of urea (mg/100 ml) in the blood of the steers 2 hours after feeding the SBP products were 4.7 (Diet A), 8.4 (Diet B) and 18.7 (Diet C) which tends to confirm the fact that when Diet A (and to a lesser extent Diet B) were given, the nitrogen intakes were very inadequate.

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# Discussion and Conclusions (Experiments 4.1 and 4.1a).

Experiments 4.1 and 4.1a were designed principally as preliminary 'calibration' experiments to determine the effects of the inclusion of urea in molassed sugar beet pulp on the voluntary intake and digestibility of low protein oat straw. The results of Experiment 4.1 indicated that supplementation with 30 g urea/day present in 1.0 kg SBP (to provide a total of about 170 g additional CP/day) increased the intake of low protein cat straw DM (1.96 % CP, 45.10 % CF) by 300 kg steers from about 4.32 kg/day to a probable maximum of 5.31 kg/day. Supplementation with up to 78 g urea/day (to provide a total of about 322 g additional CP/day) was not associated with any further increase in straw intake. Both levels of urea inclusion in the SBP increased the digestibility of the straw DM (Experiment 4.1a) from 44 to 51 - 52 %. This was mainly due to an increase in the digestibility of the crude fibre fraction of the straw. These observations would suggest that the voluntary intake of oat straw DM was restricted when 1 kg/day of unsupplemented SBP (Diet A, no urea) was given primarily by a deficiency of nitrogen in the rumen. At this particular level of energy intake, addition of 30 g urea/day to the SBP (Diet B) was sufficient to correct this inadequacy and to increase straw intake by about 20 %. It is interesting to note that maximum straw intake occurred in Experiment 4.1 when the overall crude protein concentrations of the diet was 4.4 % (Diet B). Further addition of urea increased this to 8.1 % crude protein. This lower value of 4.4 % is considerably less than the value determined by Blaxter and Wilson (1963) who reported that appetite for poor roughages was only maximal when the total crude protein content of the diet was 8.5 %. Since Blaxter and Wilson (1963) studied hays, not straw, the nature of the roughage may account for the observed difference (Elliot and Topps, 1963).

Straw intake generally reached a maximum 7 - 14 days after the introduction of urea to the diet. Withdrawal of supplementary urea resulted in corresponding but reversed changes in straw consumption. During the first 7 days after a change of diet, straw consumption tended to be somewhat variable, and there was no consistent pattern in intake which could be attributed to the provision of supplementary urea. This is perhaps a feature of "change-over" design experiments and is due to a combined "recent inclusion" and "carry-over" effect from a previous dietary treatment. The fact that straw intake tended to stabilise and reach a maximum 7 - 14 days after supplementation with urea is in close agreement with the results of Campling et al. (1962) who reported that the infusion of 150 g urea/day directly into the rumen of adult cows increased straw intake to a maximum about 5 days after the infusion of urea began. The data from the digestibility trial (Experiment 4.1a) further indicates that full adaption to a change of diet occurred within 7 - 14 days, as clearly no real differences in apparent digestibility of the various fractions of the diet (except for the CP component) were recorded when separate faecal collections were made over days 7 - 14 and days 15 - 21 of the respective feeding periods. (Tables 20 and 21). For experimental work of this nature it is suggested that a 7-day preliminary introductory period would be quite adequate unless there was a very marked change of diet.

Only limited data is available from Experiment 4.1 in respect of animal performance. Over the whole nine weeks of the trial the mean change in liveweight of the 24 steers was from 296 kg to 293 kg. However, it would be reasonable to conclude that a diet of ad libitum oat straw and 1 kg/day of molassed sugar beet pulp containing 3 % of added urea would provide a very low cost maintenance diet for cattle weighing 300 kg. Without the urea the cattle would in all probability have lost weight.

## Experiments 4.2, 4.2a and 4.2b

The voluntary intake and digestibility of cat straw by pregnant beef heifers as influenced by urea and phosphorus inclusion in molassed sugar beet pulp.

The results of Experiment 4.1 and 4.1a indicated that the inclusion of 3.0 % urea in 1 kg/day of a supplemented molassed sugar beet pulp product containing about 0.55 % P significantly increased the voluntary intake of low protein oat straw by 300 kg steers by about 20 %. The present experiments describe investigations into the separate and combined effects of supplementary urea and dicalcium phosphate contained in molassed sugar beet pulp on the voluntary consumption of oat straw. Experiment 4.2 was concerned with the measurement of the straw intake by pregnant beef-type heifers given 2.7 kg/day of SEP containing either (a) no supplement (b) 3 % dicalcium phosphate, (c) 3 % urea or (d) 3 % urea plus 3 % dicalcium phosphate. The effects of the dietary treatments on the composition of the rumen liquor and the digestibility of the oat straw are described in Experiments 4.2a and 4.2b. Data is presented in respect of the concentration of various blood constituents. The adequacy of each diet in relation to the recommended (ARC, 1965) protein, energy and mineral requirements of young cattle in mid to late pregnancy are discussed.

## Materials and Methods.

Each experiment involved the feeding of oat straw and four differently supplemented molassed sugar beet pulp products. The total amounts of feed offered were such that they were intended to meet the metabolisable energy requirements of heifers in mid to late pregnancy. It was anticipated that the oat straw and respective SBP materials would provide contrasting total daily intakes of about 6 and 17 g P and 325 and 500 g crude protein.

## Composition of materials

The composition of the oat straw and the four cubed (c.1.27 cm diameter, c.2.5 cm length) SBP products are detailed in Table 22. The oat straw contained 1.66 % crude protein (CP) and about 43 % crude fibre (CF). Product A was the normal unsupplemented commercial molassed SBP material and Products B, C and D were manufactured by the inclusion of approximately 3 % dicalcium phosphate (P) and/or urea. These increased the CP content from about 10 - 11 to about 17 - 18 % and the phosphorus from about 0.08 to about 0.5 - 0.6 % P. There was appreciably more calcium in Products B and D containing added Pand also slightly more magnesium, resulting from a small inclusion of magnesium oxide. Additionally, each contained adequate additions of trace elements (ARC, 1965) and 20 million i.u.s. Vitamin A and 5 million i.u.s. Vitamin D/1000 kg.

Thirty two beef-type females (28 heifers, 4 cows), principally Hereford x British Friesian, Hereford x Ayrshire and Aberdeen Angus x Galloway in mid-pregnancy and mean liveweight 402 kg were housed in eight balanced groups each of four animals. The experiment took the form of two  $4 \times 4$  Latin squares. The mean periods from calving for the two groups of 16 individuals at the start of the experiment were 18 weeks and 23 weeks.

Following a preliminary period of 28 days when oat straw ad libitum and Diet D (2.7 kg/head/day) were fed, the heifers were given once daily 2.7 kg fresh matter (= 2.4 kg DM) one of each of the four supplemented SEP products. The amounts of additional CP, P, Ca and Mg thus provided are given in Table 23. Oat straw was available ad libitum from racks and Norwegian-type feeding boxes, designed to reduce the wastage of straw to a minimum. The racks and feeding boxes were replenished with oat straw twice each day or as required so that at least 25 % more than the anticipated daily intake Table 22. Experiments 4.2, 4.2a and 4.2b Composition of the oat straw and the molassed sugar beet pulp products (% on DM basis).

Diet	$straw^+$	A	В	С	D
Supplement		Nil	P	Urea	Urea + P
Crude protein	1.66	11.49	10.36	18.76	16.82
Crude Fibre	42.98	13.26	13.70	12.47	14.02
Ether extract	1.36	0.71	0.67	0.48	0.65
N-free extract	46.30	66.60	65.13	59.79	58.39
Organic matter	92.30	92.06	89.86	91.50	89.86
Ash	7.70	7.94	10.14	8.50	10.14
Calcium	0.31	0.57	1.38	0.69	1.32
Phosphorus	0.10	0.08	0.61	0.09	0.49
Magnesium	0.05	0.13	0.27	0.11	0.23
-	- <b>f</b>				

T Mean of 19 samples

Table 23. The amounts (g/day) of additional crude protein, phosphorus, calcium and magnesium provided by the SBP products.

	Exper	iments	4 <b>.</b> 2 a	nd 4.2a	Experiment 4.2b			
Diet	A	В	С	D	А	В	С	D
Supplement	Nil	Р	Urea	Urea 4P	Nil	Р	Urea	Urea +P
Dry matter kg/day	2.4	2.4	2.4	2.4	1.8	1.8	1.8	1.8
Crude protein	277	251	445	409	206	186	330	303
Phosphorus	1.9	14.6	2.2	11.8	1.4	11.0	1.6	8.8
Calcium	13.7	33.1	16.6	31.7	10.3	24.8	12.4	23.8
Magnesium	3.1	6.5	2.6	5.5	2.3	4.9	2.0	4.1

was always available. The four feeding periods were each of 21 days and straw consumption was recorded during the last seven days of each period, residues being collected daily or when necessary. This procedure has previously been shown (Experiments 4.1 and 4.1a) to lead to a satisfactorily low experimental error.

The heifers were weighed at the start of the experiment and at the end of each feeding period when blood samples were also obtained. Two heifers calved in the last week of the experiment but calving generally took place from 3 to 14 weeks later. Experiment 42a Composition of rumen liquor

Four adult non-pregnant cows (mean liveweight, 526 kg) each fitted with a rumen fistula (Avon Rubber Co. Ltd) were fed in sequence in a  $4 \ge 4$  Latin square design with 21-day periods the same four SEP products at the same rate (2.4 kg DM) as in Experiment 4.2. A fixed amount of oat straw (5.5 kg fresh matter = 4.56 kg DM) was offered twice each day in two feeds each supplying 2.28 kg DM. It was anticipated as a result of a preliminary feeding period of three weeks with Diet D that this amount would be slightly in excess of the ad libitum intake of the cows.

Blood samples and samples of rumen liquor were obtained three hours after feeding the SBP products on Day 19 of each feeding period. Experiment 4.2b Oat straw digestibility trial

Four British Friesian castrates (mean liveweight, 393 kg) fitted with faecal collection harnesses (Balch et al., 1951) were fed the same four SBP products in sequence in a 4 x 4 Latin square design with 21-day feeding periods. The amount of each SBP product given was 2.0 kg fresh matter (1.8 kg DM), being rather less than the amount given in Experiments 4.2 and 4.2a as it was anticipated that the steers would also consume less straw. A fixed daily amount of oat straw (5 kg fresh matter = 4.14 kg DM) was given in two equal. feeds each of 2.07 kg DM. A preliminary feeding period of three

weeks when oat straw and Diet D were fed had indicated that this amount would be substantially consumed.

The routine procedures involved in the feeding of fixed amounts of straw and SBP, and the weighing of food materials followed the same pattern as was described for Experiment 4.1a. Faecal collections were made during days 15-21 of each feeding period. The technique employed for the collection and sub-sampling of faeces, has similarly, been detailed previously. Blood samples were taken three hours after feeding on day 15 of each period.

The digestibility data has been presented in respect of both the digestibility of the complete diets and the oat straw alone. The digestibility coefficients of the various fractions of the complete diets were determined by computerised analysis. The digestibility of the oat straw alone was calculated from the straw input and total faecal output figures, after making an allowance for the amounts of faeces attributable to the SEP (see Experiment 4.1a).

# Methods of analysis

Blood and food samples were analysed for calcium, magnesium and phosphorus. Blood samples were also analysed for urea, total protein, free fatty acids and glucose.

Rumen liquor samples were filtered through muslin and ammonia was determined following deproteinisation with sodium tungstate. Quantitative analyses for volatile fatty acids (VFA) in rumen liquor were determined by gas/liquid chromatography following treatment with 20 % W/v metaphosphoric acid.

Further analyses were undertaken to determine dry matter, crude protein, crude fibre, ether extract and ash in foods and faeces and urea in the two SBP products C and D.

## RESULTS

Live-weight gain and the intake and digestibility of oat straw (Table 24)

There was no significant difference in the live-weight gains of the pregnant heifers in Experiment 4.2. The mean daily gain on Diet D (urea + P) of 0.41 kg/day appeared to be superior to the gains when the other diets were fed. Over the whole period of 84 days of Experiment 4.2 the heifers consumed a daily average of 4.34 kg oat straw DM and 2.4 kg SBP DM and had a mean live-weight gain of 12 kg. They did however lose a considerable amount of their initially good body condition and most of the apparent small increase in liveweight must have been associated with the increasing size of the foetus.

In Experiment 4.2 the provision of supplementary urea (Diets C and D) significantly (P<0.01) increased the voluntary consumption of oat straw DM from about 3.93 to about 4.75 kg/day, i.e. by about 21 %. In Experiments 4.2a and 4.2b there were significant reductions of straw intake when urea was not given amounting to 16 % and 6 % respectively of the fixed quantities offered each day. It is possible that in Experiment 4.2a and 4.2b more than the fixed amounts of straw offered may have been consumed when urea was fed. Throughout the three experiments it was noticeable that the oat straw was more rapidly consumed when supplementary urea was included in the SEP.

Supplementary phosphorus had no effect on straw consumption either in the presence or absence of additional urea.

The apparent digestibility of the DM, OM and NFE of either the complete diets or of the straw alone were not increased by either urea or P inclusion (Experiment 4.2b). Supplementary urea appeared to increase the digestibility of the CF of the complete diet from about 60 - 63 to 65 - 66 % and of the straw alone from about 56 - 58

Table 24. Mean live-weight gain (kg/day) of the pregnant beef heifers (Experiment 4.2) and total intakes of straw dry matter (kg/day) (Experiments 4.2, 4.2a and 4.2b) and the digestibility (%) of the dry matter (DM), organic matter (OM), crude fibre (CF), N-free extract (NFE) and crude protein (CP) of the complete diets and the oat straw alone (Experiment 4.2b).

Diet	A	В	C	D	S.E. of	
Supplement	Nil	Р	Urea	Urea +P	$mean(\frac{+}{-})$	Significance
Expt 4.2						
Live-weight gain	0.05	0.09	0.02	0.41	0.118	N.S.
Straw DM intake Expt 4.2 Expt 4.2a Expt 4.2b	3.91 4.02 3.92	3.95 3.83 3.86	4•79 4•55 4•11	4.72 4.55 4.13	0 <b>.11</b> 5 0.089 0.035	C,D>A,B*** C,D>A,B** C,D>A,B**
Expt 4.2b						
DM digestibility Complete diet Straw alone	61.9 51.9	60.3 49.4	61.9 52.5	62 <b>.</b> 1 52.6	0.09 1.87	N.S. N.S.
OM digestibility Complete diet Straw alone	63.0 53.7	62.1 52.8	63 <b>.</b> 3 54.9	64.0 55.9	0.92 1.39	N.S. N.S.
CF digestibility Complete diet Straw alone	60.8 56.6	63.0 58.9	65.6 62.4	66.1 62.6	1.69 1.49	N.S. N.S.
NFE digestibility Complete diet Straw abne	67.2 51.5	64.6 47.2	63.8 48.7	65.1 50.9	1.12 1.79	N.S. N.S.
CP digestibility Complete diet Straw alone	37.3 -44.4	32.9 -54.9	52.4 43.9	49.5 38.5	0.308 8.219	C>D>A>B*** C,D>A,B***

to about 62 % but not quite significantly (P=0.05). When the data were combined for Diets A and B (no urea) and for Diets C and D (with urea) the mean increase in the digestibility of the CF in the complete diet was increased significantly (P<0.05) from 61.9 to 65.8 % by urea inclusion.

Supplementation with urea also increased the apparent digestibility of the crude protein fraction of the complete diets and the oat straw alone. For the complete diets, the mean increase in apparent CP digestibility was from about 35 % (Diets A and B) to about 51 % when urea was provided. For the oat straw alone, the apparent digestibility of the CP was negative (- 44.4 and - 54.9 for Diets A and B respectively) when no additional urea was given, but was increased to + 43.9 and + 38.5 % by the inclusion of about 3 % urea in the two SEP products B and C.

# Composition of the rumen liquor (Table 25.)

The concentration of ammonia in the rumen liquor significantly (P < 0.01) increased from about 7 to 23 mg/100 ml by supplementary dietary urea. Neither additional urea nor P increased the concentration of total volatile fatty acids in the rumen liquor. The fermentation of each diet in the rumen was characterised by the production of acetic, propionic and butyric acids in the proportions 57 : 26 : 17. The molar % of each acid in the rumen liquor. Induce the rumen was apparently unaffected by dietary urea or P supplementation. Table 25. Mean concentration of ammonia (mg/100 ml) and total VFA

(m-equiv./l) and the molar % of acetic, propinic and nbutyric acids in the rumen liquor.

Diet	А	В	С	D	S.E. of	
Supplement	Nil.	Р	Urea	Urea +P	Mean (±)	Significance
Ammonia-N Total VFA Molar % Acetic Propionic n-Butyric	7.8 63.5 57 25 18	6.2 60.3 57 28 15	21.8 57.8 56 26 18	25.2 55.3 57 27 16	1.09 6.30 2.2 1.5 1.9	C, D>A, B** N.S. N.S. N.S. N.S. N.S.

# Blood composition (Table 26.)

In most respects the concentrations of the various blood constituents measured in Experiment 4.2, 4.2a and 4.2b were essentially similar with regard to their numerical values and statistical significance.

Inclusion of urea in the SBP significantly  $(P \lt 0.01)$  increased the concentrations of the urea in the blood from about 9.5 - 11.0 mg/100 ml to 14.5 to 17.0 mg/100 ml when 2.4 kg SBP DM/day was given in Experiments 4.2 and 4.2a. In Experiment 4.2b where rather less SBP DM was fed (1.8 kg DM/day) the mean increase in blood urea concentrations associated with the inclusion of urea in the SBP was from about 7.5 to 15.0 - 19.5 mg/100 ml. It is interesting to note that in two of the experiments (Experiments 4.2 and 4.2b), significantly greater blood urea concentrations were recorded when urea alone was added to the SBP (Diet C) than when both urea and additional phosphorus were provided (Diet D). It is uncertain whether this effect was due to the provision of supplementary P per se in Diet D, or to the rather lower level of urea inclusion in this product and accordingly where the total daily intake of supplementary CP was rather less (see Table 23). Supplementary urea only marginally increased the concentrations of total protein (from 6.90 to 7.05 g/100 ml) in the blood of the pregnant beef heifers in Experiment 4.2. Inclusion of P significantly (P< 0.001) increased the concentration of blood phosphorus in Experiments 4.2 and 4.2a and in the absence of supplementary P, blood calcium concentrations were significantly elevated. The concentrations of magnesium were all about 1.9 - 2.1 mg/100 ml irrespective of dietary treatment.

The concentrations of free fatty acids in the blood were unaffected by dietary supplementation in all three experiments. Additional urea significantly increased the blood glucose concentration of the pregnant heifers in Experiment 4.2 from about Table 26.Mean concentration of urea, phosphorus, calcium,<br/>magnesium, glucose (mg/100 ml), free fatty acids<br/>(µ-equiv./l) and total protein (g/100 ml) in the blood<br/>of the heifers (Experiment 4.2), fistulated cows<br/>(Experiment 4.2a) and steers (Experiment 4.2b).

Diet	А	В	С	D	S.E. of	
Supplement	Nil	Р	Urea	Urea +P	$mean(\frac{+}{-})$	Significance
Urea Expt 4.2 Expt 4.2a Expt 4.2b	12.53 9.44 7.56	10.96 9.31 7.50	•	14.58 18.00 14.81	0.709 0.634 0.743	C,D>A,B**;C>D* C,D>A,B** C,D>A,B**;C>D**
Phosphorus Expt 4.2 Expt 4.2a Expt 4.2b	4.12 3.89 5.36	5.66 6.70 6.21	3.69 4.01 5.43	5.65 7.44 6.45	0.128 0.224 0.436	B,D≯A,C***; A>C* B,D>A,C*** N.S.
Calcium Expt 4.2 Expt 4.2a Expt 4.2b	10.08 9.80 9.75	9.72 9.30 9.15	10.11 9.43 9.68	9.61 8.95 9.50	0.079 0.074 0.068	A,C>B,D** A>B,C,D**; B,C>D** A,C>B**,D*
Magnesium Expt 4.2 Expt 4.2a Expt 4.2b	1.95 1.85 2.05	1.92 1.97 2.05	1.94 1.82 2.00	1.90 1.87 2.15	0.031 0.102 0,046	N.S. N.S. N.S.
Glucose Expt 4.2 Expt 4.2a Expt 4.2b	62.2 55.0 59.0	61.4 60.0 60.8	64.4 60.3 63.0	64 <b>.1</b> 57 <b>.5</b> 62.0	0.70 2.13 2.26	C,D>B*, C>A* N.S. N.S.
Free fatty acid Expt 4.2 Expt 4.2a Expt 4.2b	78.8 50.0 47.5	92.4 52.5 59.0	78.4 58.0 56.0	83.8 84.0 47.0	5.13 10.21 6.74	N.S. N.S. N.S.
Total protein Expt 4.2 Expt 4.2a Expt 4.2b	6.9 8.2 6.6	6.9 8.3 6.6	7.0 8.1 6.4	7.1 8.1 6.7	0.07 0.17 0.13	N.S. N.S. N.S.

61.8 to 64.3 mg/100 ml. There was also a significant difference (P < 0.01) between the two separate Latin squares in respect of blood glucose; those heifers due to calve within about 6 weeks of the end of Experiment 4.2 had a mean value of 58.6 mg/100 ml compared with 67.4 mg/100 ml for those not due to calve for a further 5 weeks.

# Discussion and Conclusions (Experiments 4.2, 4.2a and 4.2b).

The 21 % increase in voluntary consumption of oat straw DM (1.66 % CP, 43.0 % CF) by the pregnant heifers in Experiment 4.2 resulting from the inclusion of about 80 g/day urea in molassed sugar beet pulp, is in almost exact agreement with that previously recorded (Experiment 4.1) when urea was contained in SEP fed to 300 kg Friesian steers given oat straw containing 1.96 % CP and 45.1 % CF. As also reported previously (Experiments 4.1 and 4.1a) the cattle used in the present experiments consumed the offered straw more rapidly when urea was given. This was particularly noticeable in Experiments 4.2a and 4.2b when fixed amounts of oat straw DM were offered in two feeds per day.

In Table 27 the mean amounts of oat straw DM consumed by the four groups of heifers which were receiving no urea and the four groups which were given supplementary urea have been presented for each of the four consecutive recording periods.

Table 27. The mean intake of oat straw DM as influenced by urea supplementation during the course of Experiment 4.2.

	Period								
	1	2	3	4	Mean				
No urea	4.17	3.88	4.62	3.06	3.93				
With urea	4.86	4.51	4.74	4.90	4.75				
Increase	+ 17%	+ 16%	+3%	+ 60%	+ 21%				

Whilst there was a mean increase of 21 % in straw DM consumption overall there were very marked differences between the four separate periods. This appeared to be mainly due to variations in straw intake when the heifers were given no urea. In period 3 additional urea resulted in only a 3 % improvement in straw consumption but this was as much as 60 % in Period 4, largely because of the very low consumption of only 3.06 kg straw DM/day when no urea was given. It may be possible to conclude that this might be an effect associated with advancing pregnancy, but inspection of the individual data for each group of heifers shows that the variation in intake when no urea was given was equally apparent for both the early and late calving groups. This is further confirmed by the fact that there was no significant difference in straw consumption between the two separate Latin squares formed of the early and late calving heifers.

The results of Experiment 4.2b indicated that there was no marked improvement in the digestibility of the dry matter, organic matter and N-free extract of the complete diets or of the oat straw alone although the digestibility of the crude fibre was somewhat increased. This is in contrast to the results of the preceding experiments (Experiments 4.1 and 4.1a) where significant improvements in dry matter and crude fibre digestibility accompanied the increase in straw intake resulting from urea supplementation. The apparent lack of response, in terms of the digestibility of dietary dry matter and crude fibre resulting from the increase in the crude protein content of the SBP (Diets C and D) is difficult to explain. It is unlikely that this was due to a reduction in the straw: concentrate ratio in the total ration as there was only a marginal decrease (6 %) in straw DM intake where supplementary urea was given. The results of Experiment 4.2a would tend to suggest that inclusion of supplementary urea in the two SBP products C and D would have resulted in an increase in the concentration of ammonia-N in the rumen liquor and this would normally have been expected to promote microbial activity. It is unlikely that the concentrations of ammonia-N produced in the rumen would have been such as to result in inefficiency of utilisation of dietary urea as the actual concentrations recorded in Experiment 4.2a for the two urea supplemented diets (21.8 and 25.2 mg NH3-N/100 ml Diets C and D

respectively) would not be considered excessive. Several authors (notably Egan, 1966a, b) have noted that it is not always necessary to evoke the concept of an increase in digestibility to explain an increase in the voluntary intake of low-protein roughage with nitrogen supplementation. In the present experiment (4.2b) the amount of straw given was fixed and the cattle may have been able of consume more where urea was given and this might have affected the digestibility values obtained.

The results of the straw consumption trial (Experiment 4.2) are in direct contrast with those recorded by Kay et al. (1968) who were unable to show any difference in the voluntary intake of barley straw by pregnant suckler cows receiving 2 kg barley/day supplemented with either nil or 50 g urea/day. As pointed out elsewhere the barley straw used by Kay et al. (1968) contained between 5.7 and 6.0 % CP in the DM and the provision of 2 kg-of unsupplemented barley alone per day would provide sufficient additional nitrogen to increase the crude protein content of total ration to a level where maximum intake would occur. From the data of Kay et al. (1968) it can be calculated that the overall crude protein content of straw plus unsupplemented barley diet would be about 8 %. At this level no further response in straw intake would normally be expected with urea supplementation. In the present trial, inclusion of 80 g urea/day in 2.4 kg SBP DM increased the total dietary crude protein content of the whole diet from about 5 % (Diets A and B) to 7 % (Diets C and D) and accordingly there was a significant improvement in straw intake.

Supplementary phosphorus did not increase straw consumption or digestibility although the basal diet was markedly deficient in phosphorus. Previous experiments have shown (Experiments 2.1, Section 2; Experiment 3.1; Section 3) that where urea alone was added to a diet (sugar beet pulp plus a little hay) which was deficient

in phosphorus, DM intake was rather less than when supplementary phosphorus was also provided. Reduction in appetite generally occurred very rapidly and within 7-10 days of the withdrawal of additional dietary phosphorus. It was anticipated in the present experiment that where Diet C (urea only) was fed, there might have been some differential response in appetite to nitrogen supplementation in the presence of a low phosphorus intake. However, this was not the case. Similarly, a low phosphorus intake apparently did not adversely affect the digestion of roughage material in the rumen as there were no differences in the apparent digestibility of the various fractions of each diet. Nevertheless, phosphorus supplementation may be of greater significance when such low phosphorus diets are fed over more extended periods of pregnancy.

The overall pattern of a very small mean live-weight gain of the heifers in Experiment 4.2 and the obvious loss of their initially good body condition is such as to suggest that with the possible exception of the combined urea + P supplementation (Diet D), the general standard of nutrition was somewhat inadequate. Although normal concentrations of plasma free fatty acids and glucose were maintained on each treatment, the fact that lower blood glucose levels were found in those heifers which were most advanced in pregnancy further suggests a possible marginal level of energy intake.

The estimated nutrient intakes of the heifers in Experiment 4.2 in respect of metabolisable energy (ME), digestible crude protein (DCP), calcium, phosphorus and magnesium are compared in Table 28 with recommended levels of intake suggested by the ARC (1965) for 400 kg heifers in mid and late pregnancy. The ME and DCP intakes were computed in part from the digestibility data obtained in Experiment 4.2b (Table 24), after suitable allowances were made for the rather greater amounts (0.6 kg DM/day) of SBP fed in Experiment 4.2 and the

1.26

generally higher intakes of straw DM. In determining the estimated intakes of ME it was assumed that 1 Kg of digestible organic matter (DOM) would supply 3.638 Mcal ME (Alderman, 1968). The organic matter (OM) digestibility of the additional 0.6 kg SEP consumed was taken as 83.2 %. For the oat straw, the OM digestibility was taken as 52.8, 54.9 and 55.9 % for diets B, C and D. In respect of DCP intakes, it was assumed that the additional quantity of crude protein supplied as SEP (products A, B, C and D) was 62.3 % digestible and that the added urea was 100 % digestible. The intakes of digestible crude protein obtained from the additional amounts of straw consumed by the heifers relative to the amounts consumed by the steers in the digestibility trial were calculated from the data given in Table 24.

Supplementation with urea, by increasing straw consumption, increased the mean intake of ME from about 13.6 to 15.4 Mcal/day. The experimental observations of a marginal live-weight gain coupled with some loss of body condition as pregnancy proceeded suggests that the energy intake of 15.4 Mcal/day from oat straw and supplemented sugar beet pulp, whilst meeting the ARC (1965) recommendations, was hardly adequate. This conclusion is supported by the lower blood glucose concentrations recorded for those heifers nearest to parturition.

Dietary urea supplementation increased the digestible crude protein intake from about 130 g/day to 255 g/day compared with the recommended intake of 220-290 g/day. At the higher level of intake, nitrogen supplementation, whilst adequate to increase the voluntary consumption of straw by some 21 %, resulted in quite low concentrations of blood urea (14-17 mg/100 ml). This is below the normally expected level, indicating that a recommended intake of 290 g DCP/day is not excessive.

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Table 28.	Comparisons betwee	n estima	ted and	recomme	nded int	takes
	of energy (ME, Mca	l/day) d	igestib	le crude	protei	n
	(DCP g/day), phosp	horus, c	alcium	and magn	esium (	g/day)
	for 400 kg in calf	heifers	in mid	and lat	e pregna	ancy.
		ME	DCP	P	Ca	Mg
ARC (1965)	5 months pregnant	11.5	220	22	23	6.5
	8 months pregnant	15.0	290	30	38	7.5
Expt. 4.2	Diet A Nil	13.7	143	5.8	25.8	5.1
	ВР	13.5	122	18.6	45.4	8.5
	C Urea	15.4	264	7.0	31.4	5.0

15.4

246

16.5

46.3

7.9

D Urea + P

Normal concentrations of blood magnesium were recorded over the whole period of Experiment 4.2 at daily intakes of 5.0 - 8.5 g Mg, compared with the ARC (1965) recommendations of 6.5 - 7.5 g/day in pregnancy. Normal blood phosphorus concentrations were maintained at intakes of about 17 gP/day compared with the suggested requirement of 22 - 30 g P/day in mid to late pregnancy. However, of some 11 g of the 17 g P was in the form/added dicalcium phosphate. The dietary calcium intakes of 26 - 46 g Ca/day were close to the ARC (1965) recommendations and were adequate to maintain normal blood calcium concentrations.

These conclusions concerning energy, protein and mineral intakes can only be fully evaluated by means of an experiment involving a longer treatment period for a particular diet.

#### Experiment 4.3

The effect of a prolonged period of inadequate protein intake by beef heifers on calf performance.

At the end of Experiment 4.2 the pregnant beef heifers had been on a straw diet for a total of 16 weeks and were still on average 11 weeks from calving. Although the heifers had lost some body condition, it was decided to keep them on the same four SBP diets which they had received during the last 3-week period of experiment 4.2 until calving, but to feed hay instead of oat straw. Accordingly they continued to receive 2.7 kg/head/ day (= 2.4 kg DM) of molassed sugar beet pulp together with hay ad libitum.

The purpose of the present experiment was to determine the effect of a prolonged period of inadequate (ARC, 1965) intake of digestible crude protein during late pregnancy on the concentration of immune lactoglobulin in the colostrum of the heifers, the quantity of immune lactoglobulin absorbed by the newborn calves, their birthweight and subsequent growth rate. Selman (1969) has previously indicated after an extensive review of the literature that there is no published information concerning the effect of diet on the composition of bovine colostrum.

# Materials and Methods.

Voluntary hay consumption was measured during each third week using on each occasion all those heifers which had not calved. Three recordings were made during weeks 2 - 3, 5 - 6, and 8 - 9 of hay feeding. For convenience, the 8 separate groups of cows were retained, each being given the appropriate SBP products as previously described in Table 22. However, because of the small number of heifers in each group, the extended period of calving and the fact that the addition of phosphorus had been shown in Experiment 4.2 to have no effect on oat straw consumption it was decided to combine the data from the variously supplemented SEP groups into two main groups: (a) nil and P combined (Diet A,SEP) and (b) urea alone and urea + P combined (Diet B,SEPU). For comparative purposes the mean amounts of hay DM consumed by the heifers during the three recording periods are described together with the mean amounts of straw DM consumed during the last period of Experiment 4.2. The mean composition of the molassed sugar beet pulp materials, the oat straw and the hay which were given are detailed in Table 29.

Table 29. The mean composition of the oat straw, hay and SBP materials (% DM basis).

Diet	A	В	Oat straw	Hay
Supplement	SBP	SBPUT		
Crude protein	10.93	17.79	1.66	4.38
Crude fibre	13.48	13.24	42.98	32.15
Ether extract	0.69	0.56	1.36	0.84
N-free extract	65.86	59.09	46.30	56,80
Ash	9.04	9.32	7.70	5.33
Organic matter	90.96	90.68	92.30	94.67

TMean of the SBP materials containing either nil or 3 % of added dicalcium phosphate as used in Experiment 4.2.

As each heifer approached calving it was removed from the experimental area, but continued to receive the same diet for the 1 - 2 days before parturition. Calving took place in individual pens. The heifer was allowed and encouraged to lick the calf dry after birth, the calf was weighed and a blood sample obtained whenever possible before suckling and again 48 hours later for the determination of serum immune lactoglobulin concentration (McEwan, Fisher, Selman and Penhale, 1970). The immune lactoglobulin concentration (ZST units) of the pre-colostral blood sample was subtracted from the post-colostral value and the difference was assumed to be due to absorbed immune lactoglobulin. The calf was encouraged to suckle to satiation as coon as possible after birth. Normally this occurred within a period of two hours. Colostrum samples were obtained at the same time, some 10 ml being withdrawn from each teat and bulked for analysis. The total protein content of the colostral whey was determined. This has been shown (Selman, 1969) to be closely correlated ( $\mathbf{r} = 0.99$ ) with the immune lactoglobulin concentration of the colostrum.

After calving the heifers continued to receive hay ad libitum and an increased amount (3 kg/day) of SEP supplemented with both urea and phosphorus. Subsequently they were turned out to grass. This occurred at intervals ranging from 9 weeks after calving for the earliest calved heifers to a few days for those which were the last to calve. The calves were weighed regularly throughout the experiment and the live-weight gain/day to 63 days obtained. Results.

Of the 32 heifers used to measure straw consumption in Experiment 4.2 only 27 were available for hay intake measurements in the present experiment. Two of the heifers were on loan and were returned to their owner at the end of Experiment 4.2. One heifer was withdrawn from the experiment because of an apparent reaction to a routine tuberculosis test and two heifers calved during the last period of straw consumption. A further heifer calved much later than the remainder and although used to measure hay intake,

no information was obtained from its calf. In total, data were available from 26 calves, 11 of which were born to heifers in the low protein (no urea) group and 15 to heifers which were in the urea supplemented group.

The mean consumption of oat straw DM during the last period of Experiment 4.2 and the mean consumption of hay DM in the present experiment, together with the estimated intakes of metabolisable energy (ME) and digestible crude protein (DCP) are given in Table 30. The mean intakes of ME and DCP when oat straw was given were calculated as described for Experiment 4.2. When hay was fed these values were calculated after the ME and DCP contents of the hay had been estimated from its proximate composition as detailed in Table 29. (A.D.A.S. 1972; Alderman, Collins, Jones,Morgan and Ibbotson, 1967). The hay was estimated to contain 1.90 Mcal ME/kg DM and 0.70 % DCP.

In Experiment 4.2 the mean overall effect of urea supplementation had been to increase the intake of oat straw DM by 21 %. However, in the last period as given in Table 30 the mean increase was from 3.06 to 4.90 kg DM/day, i.e. + 60 %. When hay was given the increase in DM intake resulting from urea supplementation (Diet B) was 11 % (weeks 2 - 3) increasing to + 22 % (weeks 5 - 6) and to + 28 % (weeks 8 - 9). Over the whole period of hay feeding the increase in hay DM intake resulting from the inclusion of urea was + 21 % (ie. 5.68 increased to 6.85 kg DM/day).

The improved intake of straw DM in Experiment 4.2 led to an increase in ME intake from 12.1 to 15.7 Mcal/day, i.e. + 3.3 Mcal. When hay was fed the improvement in DM intake increased the ME intake by + 1.3 Mcal (weeks 2 - 3), + 2.2 Mcal (weeks 5 - 6) and + 2.9 Mcal (weeks 8 - 9) or by a mean value of 2.2 Mcal/day overall (i.e. 17.1 to 19.3 Mcal).

Table 30.	The mean intakes of straw and hay DM (kg/day) and
	the estimated daily intakes of metabolisable energy
	(ME Mcal) and digestible crude protein (DCP g).

	Weeks‡	DM in	ntake	ME ir	ntake	DCP	intake
Diet		A	В	Α	В	А	В
Supplement		SBP	SBPU	SBP	SBPU	SBP	SBPU
							•
Oat straw 7	2 - 3	3.06(14) <sup>a</sup>	4.90(16) <sup>b</sup>	12.1	15.7	127	251
Hay	2 - 3	5.96(11)	6.65(13)	17.6	18.9	204	371
Hay	5 - 6	5.35 (8)	6.55(11)	16.5	18.7	200	370
Hay	8 - 9	5.72 (6)	7.34 (7)	17.2	20.1	197	375
Hay (mean d	of 3 peri	ods) 5.68	6.85	17.1	19.3	200	372

+i.e. The last 3-week period of Experiment 4.2.

‡Weeks from commencement of this last period of feeding oat straw. At that time the first heifers to calve were 5-6 weeks from calving and the last heifers were 16-17 weeks from calving.

a,b The number of uncalved cows used to estimate straw and hay intakes.

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Inclusion of urea in the SBP effectively increased the total intake of digestible crude protein during the last period of straw consumption in Experiment 4.2 from 127 to 251 g/day. When hay was given in the present experiment the mean increase in digestible crude protein intake resulting from urea supplementation was from 200 g/day to 372 g/day.

Table 31 details the mean effects of additional dietary urea on the birthweight and live-weight gain of the calves.to 63 days, the mean 48-hour serum concentration of absorbed immune lactoglobulin and the total protein concentration of the colostral whey.

The mean number of days for which the heifers received the two contrasting dietary treatments before calving were essentially the same for the no urea and urea fed groups (75.9 and 80.8 days respectively). Additional dietary urea had no effect on the birthweight of the calves when both male and female calves in each treatment group were considered, but the 13 female calves (25.15 kg) were significantly (P< 0.001) lighter than the 13 males (30.46 kg) at birth. The mean overall live-weight gain of the calves to 63 days was about 0.80 kg/day and there were no treatment differences. The rather low mean value of only 0.69 kg live-weight gain per day for the 4 female calves born to heifers given no urea was due to the fact that one calf had congenital cerebral hypoplasia and the mother of one other female calf died of hypomagnesaemic tetany before 63 days had elapsed.

On the assumption that all the calves received colostrum in adequate amounts shortly after birth in the presence of the dam and that this would have ensured proper colostral uptake (Selman, McEwan and Fisher, 1970, 1971) urea was shown to have no effect on the 48-hour serum concentration of absorbed immune lactoglobulin which averaged 26.1 ZST units for calves born to heifers given Diet A (no urea) and 27.3 ZST units for calves born to heifers given

Table 31.	Mean birthweight (kg) and live-weight gain (kg/day)
	of the calves to 63 days, the 48-hour serum concentration
	of absorbed immune lactoglobulin (ZST units) and the total
	protein concentration of the colostral whey (g/100 ml).

Diet	Α	В	
Supplement	SBP	SBPU	
No.days on treatment	Mean S.E. 75.9 <sup>±</sup> 8.39 (11) <sup>2</sup>	Mean S.E. 80.8 <sup>±</sup> 7.95 (15) <sup>b</sup>	Significance -
Birthweight			
Male calves	30.48 <sup>±</sup> 1.841 (7)	30.45 - 1.645 (6)	N.S.
Female calves	23.81 ± 1.990 (4)	26.49 - 1.086 (9)	N.S.
All calves	28.06 ± 1.659(11)	28.07 ± 1.082(15)	N.S.
Live-weight gain			
Male calves	0.84 ± 0.062 (7)	0.80 ± 0.075 (6)	N.S.
Female calves	0.69 ± 0.051 (4)	0.80 <sup>±</sup> 0.061 (9)	N.S.
All calves	0.79 ± 0.050(11)	0.80 ± 0.040(15)	N.S.
Serum immune lactoglobulin	26.1 ± 3.79 (11)	27.3 ± 3.25 (13)	N.S.
Total protein in colostral whey	13.3 ± 1.25 (9)	11.1 + 0.93 (9)	N.S.

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a, b Number of individual values used to determine each mean value.

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Diet B (with urea). Neither dietary treatment significantly affected the total protein content of the colostral whey (mean 12.2 g/100 ml).

## Discussion and Conclusions.

The heifers improved markedly in body condition within 2 - 3 weeks of being given hay and there was a resulting marked improvement in the intake of ME to be more in accord with the ARC (1965) recommendations for 400 kg heifers in late pregnancy (see Table 28). There were no difficulties at parturition and the calves were generally healthy and vigorous.

The provision of an additional 80 g urea/day increased the mean daily consumption of hay DM by about 20 %. However, it cannot be concluded with certainty that this was entirely due to the provision of additional urea per se. The giving of hay was preceded by a period of straw feeding when the heifers receiving the SBP products without urea consumed some 60 % less straw DM per day than those which were given additional dietary urea (Table 30). There may have been some "carry-over" effect from the final period of straw feeding and this could have adversely affected the hay consumption of the heifers fed on the low protein diet (Diet A) in the present experiment.

When hay was given the two contrasting diets provided mean intakes of 200 g DCP/day (Diet A, no urea) and 372 g DCP/day (Diet B, with urea) compared with the recommended (ARC, 1965) requirement of 290 and 440 g DCP/day for 400 kg heifers in the 8th and 9th month of pregnancy. However, this marked difference in protein intake together with the associated difference in ME intake did not seem to influence calf birthweight. Neither did a low level of protein intake affect the protein content of the colostral whey or the subsequent quantity of immune lactoglobulin by the calf. It is uncertain whether this apparent lack of effect of a low protein intake would have been the same if the ME intake had been rather more marginal during the latter stages of pregnancy.

The absence of an effect on the live-weight gains of the calves may, of course, be a reflection on the fact that all the heifers were fed molassed beet pulp containing both urea and added phosphorus from the date of calving and were subsequently grazed together. There were no discernable group differences in conception when the heifers were run with a bull later in the season.

It must be concluded that much further work may be necessary to determine with certainty the effects of a reduced protein intake (and thereby possibly an accompanying reduced energy intake) by beef heifers in late pregnancy on the health and performance of the heifers and their calves.

### SECTION 5

THE USE OF A HIGH UREA-CONTAINING MOLASSED SUGAR BEET PULP PRODUCT AS A PROTEIN CONCENTRATE FOR INTENSIVELY FED CATTLE AND SHEEP. Introduction.

Concentrate feeds containing high concentrations of urea are not readily accepted by ruminants, particularly lactating dairy cows. Molassed sugar beet pulp, however, is invariably well consumed by most classes of livestock. The high molasses content of this material (c.40 %) may be advantageous in overcoming the often unpalatable nature of urea. It was considered that molassed sugar beet pulp could be an ideal medium in which to include up to 8 - 11 % of added urea to give a product with 32 - 40 % crude protein. With suitable additions of minerals, trace elements and vitamins the final cubed product could be used as a protein/vitamin/ mineral concentrate and mixed with appreciably more than its own weight of cereals.

Two experimental high urea molassed sugar beet pulp products were manufactured as described in the General Introduction to this Thesis. The cubed materials were of both the normal (c.2.5 cm)length, c.1.27 cm diameter) and a smaller production size (c.2.5 cm length, c.0.8 cm diameter). The normal sized cube contained about 8 % of added urea and between 32 and 34 % crude protein. The smaller cubes contained rather more urea (10.9 %) which increased the crude protein content to about 40 %. In addition, each material contained 3 % dicalcium phosphate plus trace elements and vitamins A and D such that their final composition was about 1.25 % Ca, 0.55 % P, 0.3 % Mg, 1.0 % Na Cl, 40 ppm Fe, 15 ppm Cu, 2 ppm Co, 60 ppm Zn, 13 ppm I, 120 ppm Mn and with up to 20 million i.u.s. vitamin A and 5 million i.u.s. vitamin D added per 1000 kg. The experiments to be described in this section have been concerned with the evaluation of the normal sized cubes as a protein/vitamin/ mineral concentrate given with barley for intensively fed,

rapidly growing steer calves (Experiments 5.1 and 5.1a). The smaller sized cubes were given to sheep (Experiment 5.2). In a subsequent trial (Experiment 6.1, Section 6) the normal sized high urea product was used to replace one-half of the vegetable protein concentrate in the production diet of dairy cows.

## Experiments 5.1 and 5.1a

The evaluation of a high urea molassed sugar beet pulp product as a protein concentrate for intensively fed Friesian steers.

It is well appreciated that urea can satisfactorily replace vegetable protein sources in diets for intensively fed steers over the liveweight range of about 100 - 250 kg. Preston, Kay, Walker, Bowers, MacLeod, Macdearmid, Philip and Hargrave (1965) replaced half the supplementary protein in a rolled barley diet with urea and noted that the performance of steers between 150 and 300 kg liveweight was not significantly affected. In later studies Walker, Kay, Preston, McDonald and Macdearmid (1968) measured the growth rate of steers from 120 kg to 400 kg liveweight when given either 0.1 % or 0.2 % urea N in the drinking water or a supplement of soya bean meal added to barley offered ad libitum. The steers given no supplementary nitrogen grew more slowly than the others up to 300 kg liveweight, but above 300 kg liveweight there were no significant differences between the three treatments. Neither were there any differences in growth rate at any stage of the experiment between the steers given urea and those given soya bean meal, or between the two urea treatments.

Growth responses from additional nitrogen in the form of urea (or vegetable protein) can only be expected when the basal diet contains insufficient nitrogen to allow full utilisation of the available energy. For steers weighing up to 250 kg liveweight and where the basal cereal diet contains less than 11 % crude

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protein in the dry matter further supplementation would be required to increase the CP content of the diet to about 14 %, if maximum gains were to be achieved (Kay, Bowers and McKiddie, 1968). At this level the diet would supply intakes of digestible crude protein which would be very close to the factorial estimates suggested by the ARC (1965). Beyond a liveweight of about 250 kg the additional response to added crude protein declines and it is doubtful if steers respond economically to levels higher than those normally found in cereals. Kay and Macdearmid (1972) have recently shown that over the liveweight range 120 - 200 kg liveweight, the growth rate of Friesian steers was increased from about 0.88 kg/day to about 1.04 kg/day when either urea or soya bean meal were added to a basal diet composed of 70 % bruised barley and 30 % ground straw offered ad libitum ( and where the crude protein content of the diet was increased by supplementation from 9.4 to 14.9 % in the dry matter). There were also corresponding improvements in food conversion efficiency from about 5.44 to 4.65 kg/kg gain. Beyond 200kg liveweight, rate of live-weight gain and food conversion efficiency were essentially similar at 1.14 kg/day and 6.28 kg/kg gain for each dietary treatment.

Previous views on the use of molassed sugar beet pulp in diets for intensively fed cattle have usually been rather conservative. It has been generally recommended that not more than 25 % should be included. Evidence presented recently would suggest that sugar beet pulp has an appreciable energy value for rapidly growing cattle and that it may be satisfactorily used at up to 50 % (Parkins, 1972) and 70 % (Boucque, Cottyn and Buysse, 1973) in all-concentrate, or concentrate with limited roughage diets. Boucque et al. (1973) summarised the results of a series of experiments in which 322 entire beef bulls were fed ad libitum

various combinations of a rolled barley/vegetable protein mixture and unmolassed sugar beet pulp over the liveweight range 150 -480 kg. Straw was available at all times. The crude protein content of the rolled barley/vegetable protein mixture was such that the experimental diets were isonitrogenous at about 15 -15.5 % CP. Daily live-weight gains of 1.21 kg/day were described for the diet consisting of 50 % barley/vegetable protein and 50 % unmolassed sugar beet pulp. When the sugar beet pulp component of the diet was increased to 60 % and 70 % the live-weight gains were 1.27 and 1.17 kg/day respectively. The corresponding food conversion ratios resulting from 50, 60 and 70 % inclusion of unmolassed sugar beet pulp were 5.91, 6.02 and 6.09 and tended to be increased with increasing levels of sugar beet pulp in the diet.

Many of the experiments reported in the literature concerning urea supplementation of diets containing sugar beet pulp have been conducted with animals initially weighing more than 250 kg. In many instances sugar beet pulp was not included as a major energy source. Nevertheless, improved live-weight gains and food conversions have been reported for steers fed concentrate diets containing sugar beet pulp and additional urea eg. Modjanov and Sul'ga (1965), Sadovnikova (1959), Tisserand and Zelter (1960), Wettereau and Holzschuh (1960).

Parkins (1972) has reported the results of two experiments in which a molassed sugar beet pulp product containing 3 % of added urea and with about 17 % CP was used as a major component (50 %) of an all concentrate diet given ad libitum to Friesian steers growing over the range of about 100 - 270 kg liveweight. This material when mixed with an equal weight of barley (to give an overall dictary crude protein content of 13.5 %) resulted in a mean live-weight gain of 1.13 kg/day which was fully equivalent to that of 1.14 kg/day for a diet composed of 85 % barley and 15 % of a groundnut/cottonseed meal mixture. Inclusion of as much as 50 % of the urea-supplemented molassed sugar beet pulp product in the diet however reduced the efficiency of food conversion from a mean of 3.95 to 4.42 kg/kg gain.

These results are in agreement with the observations of Boucque et al. (1973) and the earlier work of Connolly, Calvill, Caffrey and Ruane (1967) who compared ground and flaked maize, ground and flaked barley, and unmolassed and molassed sugar beet pulp as principal energy sources for housed Friesian steers from two weeks of age (42 kg liveweight) to slaughter at about 380 kg liveweight. Although Connolly et al. (1967) reported very similar live-weight gains for each treatment at about 0.90 to 1.13 kg/day, the mean food conversion ratios of the steers given the maize, barley and sugar beet pulp diets were 4.10, 5.10 and 6.06 kg/kg gain and were accordingly some 16 to 32 % poorer with sugar beet pulp.

Thus it would seem that although substantial quantities of molassed sugar beet pulp may be included as an energy source in all concentrate diets for rapidly growing cattle and that satisfactory live- weight gains may be achieved, a reduction in the efficiency of food conversion occurs but would not seem to be associated with the inclusion of urea. With a molassed sugar beet pulp product which contains up to 8 - 11 % of added urea and which is intended to be mixed with several times its own weight of cereals and used as a protein concentrate rather than a major energy source, it might well be expected that food conversion efficiency would be correspondingly rather better. Such a product would seem to warrant investigation.

The rate of live-weight gain of intensively fed steers could be very much associated with the palatability of the diet. The present two experiments (Experiments 5.1 and 5.1a) involved

essentially similar dietary treatments. In each experiment the high urea molassed sugar beet pulp products (SBPHU; Experiment 5.1; 7.7 % urea, 31.9 % CP; Experiment 5.1a, 8.3 % urea, 33.8 % CP) were included at about 14 - 17.5 % in an otherwise entirely barley diet, and compared as a source of supplementary protein with a mixture of equal parts of decorticated groundnut and decorticated cottonseed meals (VEG protein). Barley with no protein supplement was used as a control treatment. Materials and Methods.

In each experiment, British Friesian castrates (steers) aged about 4 months and initially weighing 110 kg (Experiment 5.1) and 129 kg (Experiment 5.1a) were fed one of the three dists detailed in Table 32. The decorticated groundnut and decorticated cottonseed meals used in Diet B were cubed (c.1.5 cm length, c.0.5 cm diameter) with an equal weight of ground barley. The final product contained 32.1 and 31.7 % CP in Experiments 5.1 and 5.1a respectively. The remaining barley for all the diets was ground, mixed with the mineral supplement and trace element mixture (Table 32) and subsequently cubed (c.2.5 cm length, c.1.27 cm diameter). The appropriate amounts of the various cubed components were mixed before feeding and stored in 25 kg paper bags for each individual.

The two supplemented diets (Diet A, SBPHU; Diet B, VEG protein) contained about 13.1 - 13.2 % crude protein (fresh matter basis) in each experiment. The low protein control diet (Diet C) contained 10.1 % CP in Experiment 5.1 and 8.5 % CP in Experiment 5.1a. The three diets were calculated to have a metabolisable energy value of 3.21 - 3.28 Mcal ME/kg DM (Ministry of Agriculture, Fisheries and Food, 1971). All the diets were supplied with suitable amounts of minerals and vitamins to meet the suggested (ARC, 1965) requirements.

Table 32.	Experiment 5.1 and 5.1a Composition, crude protein
	(% fresh matter basis) and metabolisable energy
	(ME Mcal/kg DM) content of the diets.

	Experiment 5.1.			Expe	5 <b>.</b> 1a	
Diet	А	В	С	A	B	С
Protein supplement	SBPHU	VEG	Nil	SBPHU	VEG	Ni.J.
Barley	85.8	89.6	100	82.4	85.0	100
Sugar beet pulp with urea	14.2	-		17.6		-
Decorticated groundnut meal	-	5.2	£114	-	7.5	<b>4</b> 0
Decorticated cottonseed meal		5.2		-	7.5	-
Crude protein	13.1	13.2	10.1	13.0	13.1	8.5
Calculated ME	3.22	3.28	3.28	3.21	3.28	3.28

<sup>+</sup>All the diets were supplemented with 1 % calcium carbonate, 1% sodium chloride, 0.5 % dicalcium phosphate and with a trace element and vitamin mixture (34 TE; Cooper Nutrition Products Itd). Two kg included in 1000 kg barley supplied 100 mg Fe, 20 mg Mn, 8 mg Cu, 5 mg I, 1 mg Co and 8000 i.u.s. vitamin A and 1000 i.u.s. Vitamin D<sub>3</sub> per kg of barley.

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In each experiment the steers were housed in individual pens measuring 1.22 metres width x 1.68 metres length and were bedded on poor quality wheat straw. Feed and water were offered in separate plastic containers. The diets were offered ad libitum, the feed containers being replenished twice daily so that at least 2 kg of food was always available. Each addition was mixed with any food remaining in the container so that there were no cumulative residues or selection of dietary components.

Experiments 5.1 and 5.1a were conducted according to a randomised block design. The steers (24 in Experiment 5.1 and 18 in Experiment 5.1a) were divided into groups (eight for Experiment 5.1 and six for Experiment 5.1a) each containing three individuals of comparable initial liveweight. Within each group one steer was randomly allocated to each of the three treatments. The diets were fed for a period of 49 days in Experiment 5.1 and 42 days in Experiment 5.1a. The steers were weighed every 14 days and the cumulative total of food consumed recorded. Elood samples for the determination of blood urea concentrations were obtained on day 49 (Experiment 5.1) and on days 14, 28 and 42 (Experiment 5.1a). Results (Table 33)

All the diets were equally palatable and were readily consumed. The ingredients of each diet were eaten without selection and there were no food residues. The steers given barley supplemented with SEPHU (Diet A) had fully satisfactory live-weight gains of 1.06 -1.17 kg/day and food conversion ratios of 3.97 - 4.08 kg/kg gain and in both experiments were much better than those recorded for the steers fed the low-protein diet (Diet C). In Experiment 5.1a the steers fed VEG protein (Diet B) had significantly greater (P<0.01) live-weight gains (1.15 kg/day) than those fed the unsupplemented barley diet (0.78 kg/day). There was an accompanying improvement in food conversion ratio with VEG Table 33. Experiments 5.1 (49 days) and 5.1a (42 days) Mean initial liveweight (kg), final liveweight (kg), liveweight gain (kg/day), food consumption(kg/day), food conversion ratio (kg/kg gain) and mean blood urea concentration (mg/100 ml).

	Experiment 5.1						
Diet	А	В	C	S.E. of			
Protein supplement	SBPHU	VEG	Nil	Mean(+)	Significance		
Initial liveweight	110.4	109.5	111.4	-	-		
Final liveweight	162.3	153.2	152.3	-			
Live-weight gain	1.06	0.89	0.84	0.069	A> B, C*		
Food consumption	4.24	3.53	3.70	0.018	A> B, C*		
Food conversion ratio	4.08	3.97	4.80	0.381	N.S.		
Blood urea Day 49	21.45	11.25	8.10	1.353	A≻ B, C***		

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Experiment 5.1a

Initial liveweight	130.0	128.6	127.7	-	-
Final liveweight	179.1	176.8	160.5	-	
live-weight gain	1.17	1.15	0.78	0.072	A, B>C**
Food consumption	4.64	4.34	4.29	0.177	N.S.
Food conversion ratio	3.97	3.78	5.50	0.417	C≻A, B**
Blood urea Day 14	19.63	17.00	5.96	-	-
28	15.50	14.54	6.54		-
42	15.35	13.12	4.54	-	. —
Mean	16.82	14.89	5.68	1.008	A, B>C***

protein supplementation from 5.50 to 3.78 kg/kg gain. In Experiment 5.1, however, the steers given VEG protein gained weight only marginally better than those fed the low-protein diet (0.89 and 0.84 kg/day for Diets B and C respectively). Their daily food consumption was comparable to those given the low protein diet and significantly less (P<0.05) than for the SEPHU diet. The food conversion efficiency of the steers fed VEG protein in Experiment 5.1 was nevertheless better than for the low protein diet and comparable to that for the SEPHU diet.

In Experiment 5.1a both SBPHU and VEG protein supplementation significantly increased blood urea concentrations to comparable mean values (15 - 17 mg/100 ml) which could be related to the similarly increased mean daily intakes of crude protein. In Experiment 5.1 the VEG protein diet had only a marginal effect on blood urea concentration relative to the low-protein diet. The differing blood urea concentrations of the three groups of steers were associated with mean daily crude protein intakes of 0.56 kg (SBPHU), 0.47 kg (VEG protein) and 0.37 kg (low protein).

### Discussion and Conclusions.

There is no obvious explanation for the intake of the diet including VEG protein being lower, in Experiment 5.1, than that of the diet containing SEPHU, but undoubtedly this accounts for the poor growth response to VEG protein supplementation in this particular experiment.

In both Experiments 5.1 and 5.1a the inclusion of about 8 % urea in a molassed sugar beet pulp cube to give a product with about 32% CP(and providing about 1.2 % urea in the complete barley-based diet) was as an efficient method of protein supplementation for 100 - 200 kg Friesian steers as that provided by decorticated groundnut and cottonseed meals. These results, giving a fully equal value to crude protein supplied as urea in a molassed sugar beet pulp,

product to that supplied as vegetable protein from an oil seed residue meal is fully in accord with the results of a number of previous workers: for example Kay and Macdearmid (1972) who were unable to show any difference between urea and soya bean meal as protein supplements for intensively fed steers over the range 120 -200 kg/liveweight.

Since the present work was commenced, Randall, Wallenius, Dyer and Hillers (1972) in America have reported that a molassed sugar beet pulp product containing urea and 49 % CP was as efficient as soya bean meal as a supplementary protein source for dairy heifers growing over the range 75 to 200 kg liveweight. The high inclusion of urea did not give rise to any abnormality in the concentration of ammonia in the blood or in the eventual carcass composition.

#### Experiment 5.2.

The evaluation of a high urea molassed sugar beet pulp product as a protein concentrate for intensively fed lambs.

There is no published information concerning the value of urea and molassed sugar beet pulp in diets for intensively fed growing lambs but many observations have shown that these products are well consumed by sheep. In the two proceeding experiments (Experiments 5.1 and 5.1a) it was demonstrated that a high urea molassed sugar beet pulp product was a very effective protein source for rapidly growing steers. A similar product may well be of use as a dietary protein supplement for intensively fed growing lambs.

The present experiment was concerned with the evaluation of a high urea molassed sugar beet pulp product which contained about 11 % of added urea and containing 40 % CP as a protein/vitamin/ mineral concentrate for growing lambs. Comparisons were made between this product and with equivalent amounts of supplementary nitrogen provided as a conventional vegetable protein mixture and a molassed sugar beet pulp material containing 2.5 % urea and 16.9 % crude protein.

## Materials and Methods.

Fourty-eight Greyface cross lambs (32 Suffolk, 8 Dorset Down and 8 Dorset Horn) were available for this experiment. The lambs were weaned at about 6 weeks of age and during the 3 - 4 week interval prior to the start of the experiment were offered ad libitum an admixture of the four experimental diets. Following this preliminary introductory period the lambs (mean initial liveweight 20 kg) were allocated to 12 pens so that there were four lambs in each pen. Within each group there was one castrated male (wether) and three females. Each pen of lambs had an even distribution of individuals with similar liveweights. As far as possible the 12 groups of lambs were balanced with respect to breed.

Three groups of lambs were assigned at random to one of each

of the four experimental diets detailed in Table 34. The three supplemented diets (Diets B, C and D) contained 12.6 % crude protein (fresh matter basis). Diet C consisted of equal parts of barley (8.4 % CP) and molassed sugar beet pulp with 2.5 % of added urea (SBPU; 16.9 % crude protein). In Diet D equivalent amounts of supplementary nitrogen were supplied as a cubed high urea molassed sugar beet pulp product containing 10.9 % urea (SBPHU: 39.6 % crude protein). Diets A and B acted as negative and positive control treatments respectively. Diet A (low protein) and Diet B (VEG protein) each contained 13.6 % of unsupplemented molassed sugar beet pulp (SEP; 10.0 % crude protein). Additional nitrogen was provided in the positive control diet (Diet B) as a mixture of equal parts of decorticated groundnut and decorticated cottonseed meals. The groundnut and cottonseed meals included in Diet B were mixed with the barley component and the mineral/trace element/vitamin addition and cubed (c.1.5 cm length, c.0.5 cm diameter). The barley for the remaining diets was cubed in a similar manner. Prior to feeding, the barley and barley/vegetable protein fractions of the respective diets were mixed with the cubed SBP products, which were all of the smaller production size (c.2.5 cm length, c.0.8 cm diameter). The calculated metabolisable energy value of the diets were: Diet A 3.23; Diet B 3.22; Diet C 3.08; Diet D 3.23 Mcal/kg DM (Ministry of Agriculture, Fisheries and Food, 1971). All the diets were supplied with adequate (ARC, 1965) amounts of minerals and vitamins.

The lambs were housed in an open fronted building in pens with slatted floors. The pens provided 1.45 sq metres of resting area per animal and were each equipped with one "gravity fill" water bowl. Feed was offered ad libitum from 1.2 metres length troughs placed immediately outside the penned area. The troughs were generally replenished twice daily so that at least 20 % more than

Table 34.

24. Composition, crude protein (% fresh matter basis) and metabolisable energy (ME Mcal/kg DM) contents of the diets.

Diet	A	В	С	D
Supplement	Nil	VEG	SBPU	SBPHU
Barley	86.4	73.2	.50.0	86.4
Decorticated groundnut meal	t.m	6.6		-
Decorticated cottonseed meal	-	6.6		<b>i</b>
Unsupplemented SBP	13.6	13.6	-	
SBP 2.5 % urea	•••		50.0	-
SBP 10.9 % urea	-	-	-	13.6
Crude protein	8.6	12.6	12.6	12.6
Calculated ME	3.23	3.22	3.08	3.23

+ All the diets were supplemented with 1 % calcium carbonate, 1 % sodium chloride, 1 % dicalcium phosphate and with a trace element and vitamin mixture (34 TE; Cooper Nutrition Products Ltd.). Two kg included in 1000 kg barley supplied 100 mg Fe, 60 mg Zn, 20 mg Mn, 8 mg Cu, 5 mg I, 1 mg Co and 8000 i.u.s. Vitamin A and 1000 i.u.s. Vitamin D<sub>3</sub> per kg of barley.

the anticipated daily intake of feed was available. Each addition was intermixed with any food remaining in the feed trough in an attempt to reduce selection of dietary ingredients.

The experiment was continued for 10 weeks. The lambs were weighed every 14 days and the cumulative total of food consumed recorded. Blood samples for the determination of blood urea concentration were obtained at 14-day intervals.

## Results (Table 35.)

The VEG protein and SEPHU diets were well consumed throughout the experiment. Initially, the SEPU diet (C) was not readily accepted. This did not appear to be due to a papatability effect per se, but rather to the inclusion of such a high proportion (50 %) of molassed sugar beet pulp in the diet. Gradually however, consumption increased and within 10-14 days full appetite was attained. Indeed over the whole period of the experiment the lambs fed the SEPU supplemented diet (C) had the highest mean daily food intake. The low protein diet was associated with the poorest food

Dietary protein supplementation with either SBPU or VEG protein significantly (P<0.01) increased the daily live-weight gain of the lambs from 0.16 kg/day to about 0.24 kg/day. The response to additional nitrogen supplied as SEPHU (Diet D) was rather less. Mean live-weight gains of 0.20 kg/day were recorded for the lambs fed the SBPHU containing diet and were significantly lower (P<0.05) than the mean gains resulting from SEPU supplementation.

The mean food conversion ratio data presented in Table 35 were calculated from the total live-weight gain and the total food consumption of the three groups of lambs fed on each dict. The determined values represent an overall figure for each treatment. All forms of protein supplementation were associated with an improvement in overall food conversion efficiency. Food conversion Table 35. Mean initial and final liveweight (kg), live-weight gain (kg/day), food consumption (kg/day), overall food conversion ratio (kg/kg gain) and mean blood urea concentration (mg/100 ml) during the 70-day experimental period.

Diet	А	В	С	D	S.E. of	
Supplement	Nil	VEG	SBPU	SBPHU	$Mean(\frac{+}{-})$	Significance
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Initial liveweight	20.7	19.6	19.9	19.7	-	-
Final liveweight	31.8	35.9	37.3	33.7		-
Mean live-weight gain	11.1	16.3	17.4	14.0	-	25.
Live-weight gain/day	0.16	0.23	0.25	0.20	0.12	C>D* D>A*; B,C>A**
Food consumption	1.27	1.41	1.48	1.35	<b>1</b> 74	-
Overall food conversion	†7.9	6.1	5.9	6.7	0.24	A > B,C,D**
Blood urea Day 14	10.71	20.90	214.26	35.16	-	
. 28	19.25	23.79	28.13	46.42	8 m	-
42	11.71	25.19	30.29	44.23		
56	12.70	24.40	28.75	39.40		
70	14.90	32.60	31.33	34.19	-	
Mean	13.85	25.38	28.06	39.89	1.227	B,C,D≯A***
						D>B,C***

<sup>†</sup>Calculated from the total food intake and the total live-weight gain of the three replicates for each dietary treatment.

ratios tended to be somewhat better at 5.9 and 6.1 kg/kg gain when the basal diet was supplemented with either VEG protein or SBPU compared with 6.7 kg/kg gain when the protein addition was as the high urea product SBPHU. The results also indicated that when the diet contained up to 50 % molassed sugar beet pulp this did not materially affect overall food conversion efficiency.

Both VEG protein and SBPU significantly increased the mean blood urea concentration of the lambs from about 14 to 25-28 mg/ 100 ml. Inclusion of the high urea molassed sugar beet pulp product in Diet D further increased the mean blood urea concentration of the lambs to about 40 mg/100 ml.

## Discussion and Conclusions.

The primary purpose of the present experiment was to compare the effectiveness of either 2.5 or 10.9 % urea contained in molassed sugar beet pulp with a mixture of equal parts of groundnut meal and cottonseed meals as dietary protein supplements for growing lambs fed all concentrate diets. The total crude protein content of the respective diets was determined by the level attained in Diet C by mixing barley with an equal weight of the cubed molassed sugar beet pulp product containing 2.5 % of added urea (SEPU). In the event the barley component contained 8.4 % crude protein (fresh matter basis) and accordingly the overall dietary crude protein concentration was only 12.6 % and was lower than the 13.5 - 14.0 % level which was originally intended.

The lambs responded equally well to additional dietary nitrogen supplied as either VEG protein (Diet B) or 2.5 % urea inclusion in molassed sugar beet pulp (Diet C). The overall mean live-weight gain of 0.24 kg/day of the lambs fed Diets B and C and the corresponding food conversion ratios of about 6.0 kg/kg gain were considered reasonably satisfactory under the present ad libitum feeding regime. The poorer growth response which accompanied the inclusion of the high urea molassed sugar beet pulp product (SEPHU, 10.9 % urea) in Diet D was associated with a mean blood urea concentration of about 40 mg/100 ml. This was some 12-15 mg/100 ml higher than the mean blood urea concentration of the lambs given VEG protein and SEPU containing diets and may have been related to an inefficient utilisation of dietary urea. This might explain the slower growth rate of the lambs given SEPHU as a protein supplement.

It is appreciated that a rather heterogeneous group of lambs with respect to breed type were used in this experiment and that much greater numbers of animals would generally be required to fully evaluate the contrasting dietary treatments. Nevertheless, it would be reasonable to conclude that under the conditions of the present trial, 2.5 % urea contained in a molassed sugar beet pulp cube was as well utilised as conventional vegetable protein sources. The high urea molassed sugar beet pulp product (SEPHU; 10.9 % urea) was a less effective method of dietary protein supplementation.

## SECTION 6

# THE USE OF UREA-CONTAINING MOLASSED SUGAR BEET PULP AS A MILK PRODUCTION CONCENTRATE FOR DAIRY COWS.

#### Introduction.

Under present British farming conditions one of the main areas for use of molassed sugar beet pulp is in the maintenance diet of dairy cows. It is generally given at about 1 - 2 kg per day as an energy supplement to compensate for the low nutritional value of poor quality hay and cereal straws. It may also be used with silage with the intention of providing sufficient energy for maintenance and the production of about 5 kg of milk per day. Molassed sugar beet pulp when suitably supplemented with added urea, dicalcium phosphate, trace elements and vitamins, might give a product with adequate nutrients for milk production.

Ronning and Bath (1962) in America, van Es, Nijkamp and Vogt (1971) in Holland and more recently Castle (1972) in Britain, have concluded that for milk production purposes molassed sugar beet pulp and barley have the same energy value. In particular, Castle (1972) recorded similar yields of milk when a diet containing 80 % barley and 20 % groundnut cake was progressively changed to one consisting of 80 % molassed sugar beet pulp and 20 % groundnut cake providing comparisons were made on a dry matter basis.

In the late 1930's much work was conducted, particularly in Germany (eg. Nehring, 1937, 1939; Schmidt, Kleish, and Kamphferr, 1937; Ehrenberg, Nitsche and Muller, 1938) to evaluate the use of concentrate mixtures containing amide slices prepared from sugar beet pulp, molasses and urea. It was generally found that where urea formed about 30 % of the total nitrogen in the concentrate diet milk yields were about 10 % lower than where vegetable protein sources were given. More recently Wettereau and Holzschuh (1958/59) and Wettereau, Schlegel and Holzschuh (1961/62) used similar amide slices to replace cereals and soya bean mixtures and recorded fully comparable milk yields.

Investigations have also been made concerning the use of ammoniated sugar beet pulp for milk production purposes. Provided the ammonia treatment was such that it did not adversely affect the palatability of the final product (Millar 1941; Ferguson and Neave, 1943) reasonably satisfactory results have been obtained. For example, Broster, Balch, Bartlett and Campling (1960) aumoniated sugar beet pulp to increase the crude protein content from 10.4 to 20.4 % and recorded that this material was almost as effective as decorticated groundnut cake in increasing the yields and composition of milk. Chomysyn, Bielinski and Slabon (1962) reported work from Eastern Europe in which up to two-thirds of a milk production concentrate was replaced by anmoniated or urea-containing sugar beet pulp without significant loss in milk yield or reduction in milk fat content. Kaemerrer and Bollmann (1968) fed a proprietary compound based on urea-containing sugar beet pulp which supplied up to 200 g urea per day and reported that this did not lead to a reduction in milk yield or quality.

The feeding of urea to dairy cows is frequently associated with palatability problems. Almost invariably inferior milk yields have been recorded with urea-containing products relative to those attained when vegetable protein sources were used. Armstrong and Trinder (1966) have summarised the results of some 32 experiments in which urea was included at about 2 % in milk production diets for dairy cows. Urea generally provided about 38 % of the total nitrogen in the production diet (i.e. raised the crude protein of cereals from about 10 to 16 %). At this level, the overall mean yield of about 14 kg of milk per day was reduced by about 0.8 kg per day when urea-containing concentrate mixtures were given for the whole of milk production. It is probable that most of the concentrate

mixtures used in the experimental work summarised by Armstrong and Trinder (1966) were based on loose mixtures of cereals and crystalline or prilled urea and the palatability of the diet may have affected milk yield. Nevertheless, a reduction in milk yield was a consistent finding, i.e. in 31 of the 32 experiments described.

More recently Parkins, Hemingway and Ritchie (1974) have reported the results of 3 experiments in which a molassed sugar beet pulp product supplemented with about 3 % urea and 3 % dicalcium phosphate and containing 17 % crude protein, 0.5 % P and 1.25 % Ca was used as a major constituent of a milk production concentrate for dairy cows. The trials described by Parkins et al. (1974) were conducted to change-over designs, with feeding periods 4 - 6 weeks and at mean yields of about 15 kg of milk per day. Substitution was generally made for a milk production concentrate composed of barley mixed with groundnut and cottonseed meals. Where the ureacontaining molassed sugar beet pulp material formed about 35, 50 and 62 % of the concentrate mixture, so that urea provided 13, 18 and 27 % of the crude protein in the concentrate, there was generally a corresponding reduction in milk yield relative to that attained with the control barley plus vegetable protein concentrate of 0.38, 0.42 and 0.90 kg/day. The results of the three experiments formed a series, the mean reductions in milk yield being in approximate proportion to the overall reduction in yield in the many experiments reviewed by Armstrong and Trinder (1966) in which a mean milk yield of about 14 kg/day was reduced by about 0.8 kg/day when urea formed some 38 % of the total crude protein in the milk production concentrate.

It was concluded by Parkins et al. (1974) that the results of their experiments could be explained on a basis of the normal reduction in milk yield to be expected from urea if molassed sugar beet pulp and barley were considered to have the same energy value for milk production purposes. On the other hand, if molassed sugar beet pulp had a lower energy value than barley, the progressive decline in milk yield noted when the proportion of sugar beet pulp in the milk production concentrate was increased could only be explained if urea had a value closely comparable to that of vegetable protein. This could indeed be the case if the high molasses content and the palatable nature of urea-containing molassed sugar beet pulp cubes made this product a superior protein source to the physical forms of urea often used in the experiments reviewed by Armstrong and Trinder (1966).

The experiments conducted by Parkins et al. (1974) involved relatively short term feeding periods (4 - 6 weeks) and it is possible that the rather inferior results obtained with the ureacontaining molassed sugar beet pulp material could have been due to a lack of adaption to urea given in quantity although they argued that this was unlikely to be the case.

The experiment to be described in this Section of the present work was concerned with a trial with dairy cows in which a molassed sugar beet pulp product containing 2.7 % of added urea and 16.8 % crude protein (essentially similar to that given by Parkins et al. 1974) was included at 50 % in a milk production concentrate mixture, but where the diet was fed for a more extended period and with cows having a rather higher initial milk yield. Comparisons were also made with a concentrate mixture which included a molassed sugar beet pulp material containing 7.8 % of added urea and 32.6 % crude protein and which was used as a protein/vitamin/ mineral concentrate. Both urea-containing products provided about the same quantity of dietary urea. The concentrate mixture which included the high urea product however necessarily contained

a much smaller proportion of molassed sugar beet pulp and was otherwise composed of barley and oats as the main energy sources.

## Experiment 6.1

## Materials and Methods.

The experiment was conducted with 15 Ayrshire and 9 British Friesian cows. A continuous-type design was used so that the prolonged effect of each dietary treatment could be examined. The 24 cows were managed uniformly until the experiment started. All the cows received the normal farm concentrate mixture (Diet A, Table 36) and the same hay and silage that was to be fed during the experimental period. In order to minimise the effects of individual animal variation with the small number of cows employed in the experiment, there was a pre-experimental period of three weeks during which the milk yield of each cow was recorded on 12 occasions. Milk samples were obtained twice on Day 11 and Day 16 of this 3-week period. The results obtained during the preliminary period were later used to adjust the results from the experimental period by covariance analysis (Snedecor, 1956).

On the date the experiment commenced (10 January) the 24 cows were divided into eight groups of three. The three cows in each group were randomly allocated to one of the three feeding treatments which were given for the experimental period of 56 days. The cows in each group were as similar as possible in number of previous lactations, stage of lactation, present milk yield and breed. Seven and ten of the cows respectively were in the first or second lactation and the other animals were in their third or fourth lactation. The average number of the previous lactations per cow was 2. The majority of cows had reached their peak lactation yield and were at least 100 days calved. Two of the eight sets of three cows were more recently calved (average 28 days) and were approaching their peak milk yield. The mean milk yield of the 24 cows at the start of the experiment was 20.65 kg/day.

The composition of the three concentrate mixtures given in the experiment are detailed in Table 36. Diet A was the normal farm concentrate in which rolled barley (10.1 % CP) and oats (11.1 % CP) were the major energy sources.

Table 36. Mean composition, crude protein (% fresh matter basis) and calculated metabolisable energy (ME Mcal/kg DM)

contents of the diets.

Diet ,	A	В	С
Supplement	VEG <sup>+</sup>	SBPHU	SBPU
Barley	66.8	62.4	28.5
Oats	12.6	12.6	12.5
Decorticated groundnut meal	10.3	4.5	4.5
Decorticated cottonseed meal	10.3	4.5	4.5
SBPHU 7.8 % urea	-	16.0	. –
SBPU 2.7 % urea	-	-	50.0
Crude Protein	16.5	16.6	16.3

+Additionally this diet contained 0.5 % NaCl, 0.5 % CaCO<sub>3</sub> and 0.5 % dicalcium phosphate plus trace elements and vitamins to ARC (1965) standards.

Additional crude protein was supplied as a mixture of equal parts of decorticated groundnut meal and decorticated cottonseed meal. The groundnut and cottonseed meals for use in Diet A were mixed with about an equal weight of ground barley and the appropriate mineral and vitamin/trace element mixture and subsequently cubed. The final product contained 32.2 % crude protein. In Diet B a cubed (c.2.5 cm length, c.1.27 cm diameter) high urea molassed sugar beet pulp urea crude material (SEPHU) containing 7.8 %/and 32.6 %/protein replaced about one-half of the additional crude protein derived from the groundnut and cottonseed meals in the control concentrate (Diet A). Diet C included 50 % of a similarly cubed molassed sugar beet pulp material containing 2.7 % urea and 16.8 % crude protein. About onehalf of the additional crude protein in Diet C was derived from vegetable protein sources and about one-half from SEPU. The three concentrate mixtures were comparable in overall crude protein content (16.3 - 16.6 % crude protein fresh matter basis). Urea was calculated to supply 22.3 and 21.1 % of the total crude protein in Diets B and C respectively. The concentrates were given at a standard rate of 1 kg/2.5 kg milk produced i.e. 4 lbs/gallon.

The basal maintenance diet consisted of silage and hay. The compositions (% dry matter) were as follows: Silage, 24.9 % DM, 12.1 % CP and 31.3 % CF; Hay 83.0 % DM, 7.6 % CP and 34.9 % CF. The average energy content of the silage and hay, expressed on a dry matter basis, was determined from the crude fibre and crude protein contents (A.D.A.S. 1972). The silage DM contained 45 SE (2.31 Mcal ME/kg) and the hay DM 33 SE (1.89 Mcal ME/kg). The average digestible crude protein contents of the silage DM and hay DM were 7.33 and 3.19 % respectively. A total of 18.2 kg of silage and 3.6 kg of hay/day were fed to all the cows. These were estimated to supply 3.0 kg SE (16.11 Mcal ME) and 0.42 kg digestible crude protein and were adequate to meet maintenance requirements (Evans 1960; ARC 1965).

The cows were housed in double standings in a traditional byre, but were allowed into a bare concrete yard for 2 hours each day. Silage was given mid-morning and hay after the evening milking. The concentrate ration was given in two equal parts at the morning and evening milking which commenced at 05.30 hours and 16.00 hours. The concentrate allocation for each cow was adjusted at the end of each week according to the actual yield recorded over the previous week by the method of Lucas (1943). Milk yields were recorded on 5 days each week during the 8-week experimental period, except during week 6 when milk yield data was obtained on only two occasions. Individual milk yields were measured using Continuous Sampling Milkoscope Recorders (Foss Electric Ltd), calibrated before use and with a determined accuracy of  $\pm 2$  % of the total daily milk yield. Milk samples from consecutive evening and morning milkings were obtained once weekly from each cow and analysed for fat (Gerber, ESI Publ. 696, Parts 1 and 2) and total solids (by both drying and density determination). The daily yields of fat corrected milk (FCM) were calculated by the method of Gaines (1928).

Statistical evaluation of the experimental data was kindly undertaken by Mr. R. Henderson of the ARC Unit of Statistics, Edinburgh University.

## Results.

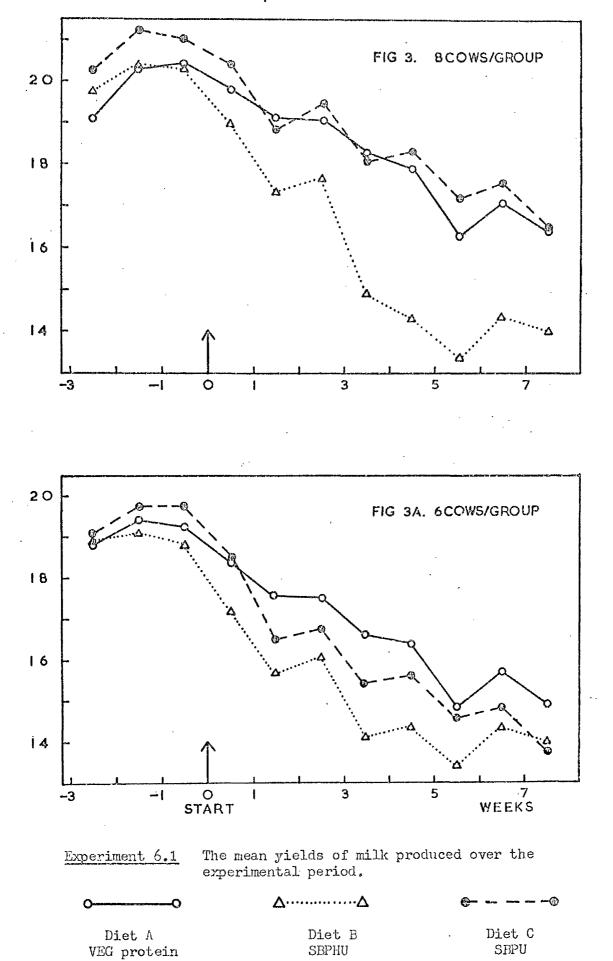
The control VEG protein concentrate (Diet A) and the SBPUcontaining concentrate (Diet C) were well consumed by all the cows and there were no palatability problems. Diet B which included 16 % of the high urea molassed sugar beet pulp material (SEPHU) was not well accepted, particularly by the higher yielding animals. Only 3 of the 8 cows fed Diet B readily ate the concentrate allocation given at each milking. Three cows consumed the concentrate mixture more slowly over a period of 2 hours and a proportion of the high urea cubes were continually rejected each day. The two remaining cows given Diet B (nos 17 and 68) ate very poorly and substantial quantities of both the high urea molassed sugar beet pulp material and the barley/oats/vegetable protein mixture frequently remained uncaten. Both these two cows were more recently calved (no 17, 28 days and no 68, 35 days) at the start of the experimental feeding

period. The actual milk yield of all 8 cows given Diet B declined markedly during the first 28 days of the experiment from an initial mean of 20.41 kg/day to 14.91 kg/day (Fig.3). This was generally the case for all the cows but was accentuated due to the fact both cows no 17 and 68 developed acetonaemia between Day 28 and Day 30, their individual milk yields falling from a mean of about 23 kg to 9 kg/day over the course of about one week. Both cows were subsequently withdrawn from the experiment. Because of the very serious effect which this had on the overall mean milk yield of the 8 cows given Diet B it was decided to omit the data obtained for cow no 17 and 68 and their paired counterparts given Diets A and C when the results of the experiment were statistically analysed. The experiment was accordingly effectively reduced to 6 cows/treatment.

Because of these palatability difficulties the SBPHU material was mixed with the normal milk production concentrate diet given to cows on two other farms. Similar difficulties in consumption were noted and it is concluded that the difficulties encountered in feeding Diet B to the experimental cows was not confined to these particular animals. In an attempt to reduce the serious decline in milk yield and the acetonaemic tendency, 1.8 kg/day of barley were given to the six remaining cows given Diet B from Day 30.

The mean yields of actual milk produced by the cows on the 3 concentrate treatments are presented in Figures 3 and 3a. Figure 3 of represents the mean daily yield/all 8 cows in Groups A and C over the whole 56 days of the experiment and the mean daily yield of the 8 cows in Group B up to Day 28 and the mean yield of the 6 remaining cows in Group B thereafter. Figure 3a represents the mean daily milk yield over the 56 day experimental period when only 6 individuals per treatment were considered. Heference to Figs. 3 and 3a clearly demonstrates the marked effect on mean actual milk yield

MEAN MILK YIELD KG/DAY



of reducing the number of cows in each treatment group from 8 to 6 as those removed had much the highest yields.

When all 8 cows which received either the VEG protein or SEPU-containing diets are considered, the overall pattern of decline in milk yields(Fig 3) were very comparable. Over the 56-day experimental period the mean actual daily milk yields were 18.01 kg/day for the cows given Diet A and 18.33 kg/day for those given Diet C. The inclusion of 50 % SEPU in Diet C apparently had no detrimental effect on the actual milk yield of the 8 cows given this diet.

The results of the experiment when analysed on a basis of 6 cows per treatment are detailed in Table 37. The mean milk yield and milk composition data are given as adjusted values using pre-experimental milk yield and composition as covariates. <u>Table 37.</u> Mean daily yields of milk (kg), fat corrected milk (kg) and milk composition ( $\frac{\pi}{2}$ ).

Diet	A	В	С	S.E. of	
Supplement	VEG	SBPHU	SBPU	$mean(\pm)$	Significance
		•			
Milk yield					
Actual milk	15.66	15.40	15.60	0,500	N.S.
Fat corrected milk	17.27	16.00	16.30	0.363	A > B*
Milk composition					
Fat	4.20	4.38	4.16	0.085	N.S.
Total solids	12.75	13.05	12.49	0.121	B≻ C*

There were no significant differences between the three treatments in daily yield of actual milk, although the milk yield on Diet A (VEG protein) was about 1.1 - 1.3 kg/day greater than that produced on Diet C (SBPU) and Diet B (SBPHU). The yield of FCM with VEG protein supplementation (17.27 kg/day) however, was significantly greater (P $\leq$ 0.05) than the daily yield of FCM (16.00 kg/day) resulting from SBPHU inclusion in Diet B. The fat content of the milk was not significantly affected by either concentrate treatment and was within the range 4.16 - 4.38 %. In respect of the total solids content of the milk, this was significantly greater (P $\leq$ 0.05) when the concentrate mixture contained SBPHU rather than SBPU. <u>Discussion and Conclusions</u>.

The concentrate mixture containing the high urea molassed sugar beet pulp material was associated with considerable palatability problems which were not fully anticipated when the experiment was originally designed. Experimental work with the same product contained in all-concentrate diets given to young rapidly growing steers and young sheep (Section 5 Experiments 5.1, 5.1a and 5.2) presented no such difficulties. Unpalatability of urea is commonly encountered with dairy cows when large amounts of concentrates are fed at milking time (eg. Waite, Castle, Watson and Drysdale, 1968). The high molasses content (c.40 %) of the sugar beet pulp was not sufficient to mask the taste of urea when included at 7.8 % in the high urea product. There were however no palatability problems when urea was included at about 3 % in the SEPU product. Nevertheless, rejection of the high urea molassed sugar beet pulp cube was not the only reason for the reduction in yield of the cows given Diet B as, frequently, considerable amounts of the concentrate mixture were also left, although some of the cows would reject the SBPHU cube selectively. There is also the aspect that disturbance of the normal eating habit of the cows at milking time may have upset milk letdown. No really valid conclusions can be drawn regarding the nutritional value of the SBPHU-containing diet because of these complications.

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The absence of any significant difference between the milk yield of the cows given either the barley/oats/vegetable protein concentrate mixture alone or when replaced with 50 % of SBPU is in agreement with the results of earlier work with this material in short-term change over trials with dairy cows (Parkins et al. 1974). The results of the present experiment would also seem to confirm the findings of other authorities (eg. Castle 1972; van Es et al. 1971) that for milk production purposes there is no material difference between the nutritional value of molassed sugar beet pulp and barley.

# Appendix

## Sampling Procedures.

Feeds

Hay, straw, silage and concentrate feeds were sampled where appropriate at least once per week. For hay and straw which was in bale form the general procedure was to obtain samples from 10-12 separate bales at feeding time when the latter were broken apart. Silage samples were obtained in a similar manner immediately prior to feeding. In the experiments where fixed amounts of roughage and concentrates were given eg. oat straw and SEP in the two digestibility trials (Experiments 4.1a and 4.2b) the most convenient procedure was to take a sample from each portion of food as it was weighed into the appropriate paper or plastic bag. Each separate sample was added to a cumulative sample, which was then used for laboratory analysis. In the experiments where given, representative samples of the separate ingredients were taken prior to mixing and cubing.

# Rümen liquor

A standard procedure was adopted in each experiment. Samples of digesta were removed by hand from several separate sites in the rumen, squeezed, and the extracted liquor collected into 500 ml beakers.

### Blood

Blood samples were taken from the jugular vein using 10 ml vacutainer tubes containing either 100 i.u.s. of heparin (for the determination of calcium and magnesium) or 20 mg potassium oxalate and 25 mg sodium fluoride (for the determination of glucose, phosphorus and urea). Clotted blood samples were obtained for total protein and free fatty acid estimations. Milk

Milk samples were obtained by the use of Milkoscope Recorders (Foss Electric Ltd). This type of recorder collects a continuous 1-2 % sub-sample of the milk flow during the process of milking. The samples of milk obtained at consecutive evening and morning milkings were combined and stored in glass bottles until subsequent analysis on the same day.

### Preparation of samples for analysis.

All roughage feeds were dried before analysis and then ground through a 3 mm screen using a laboratory hammer mill (Christy and Norris Ltd., Chelmsford, England). Concentrate feeds were ground and the appropriate analyses undertaken on the fresh material.

Faecal samples were dried and ground before analysis for all constituent with the exception of nitrogen. For nitrogen determination, 150 g of fresh faeces were macerated in a blender (Ato-Mix, M.S.E. Ltd., London) with 150 ml of water and 5 ml of toluene to form a uniform cream. The cream was subsampled and weighed directly into a Kjeldahl flask (Commonwealth Bureau of Pastures and Field Crops, 1961). Urine samples for laboratory , analysis were obtained by sub-sampling from the Winchester quart containers in which the daily samples had been stored.

Determinations of crude protein (total N x 6.25), crude fibre, ether extract and ash were made by normal, standard procedures (Fertiliser and Feeding Stuffs Regulations, 1973).

Calcium and magnesium in foods, faeces, urine and blood were determined by atomic absorption spectroscopy using lanthanum chloride to suppress phosphorus interference. Phosphorus in foods, faeces and urine was determined by the colorimetric phosphovanado-molybdate method of Cavell (1955). Phosphorus in blood was determined after deproteinisation with trichloroacetic acid using the colorimetric reaction with aminonapthosulphonic acid (Fiske and Subbarow, 1925).

Urea in foodstuffs was determined after decolourisation with activated charcoal of an acid extract followed by the colorimetric reaction with p. dimethylamino benzaldehyde (Fertiliser and Feeding Stuffs Regulations, 1973).

Blood urea was determined following incubation of blood plasma with the enzyme urease. After precipitation of the plasma proteins the colour produced with Nesslers Reagent was compared colorimetrically with that produced under the same conditions with a standard urea solution. The total protein content of blood was determined by the biuret reaction method of Reinhold (1953).

Blood ammonia concentration was determined by the method of Hutchinson and Labby (1962). Care was taken to ensure that all glass ware was ammonia-free. Glass ware was washed in a hot detergent solution, rinsed in hot tap water, rinsed again 3 times in distilled water, soaked in 0.1 N NaOH and rinsed 10 times with ion-free water before drying. Within 1-2 minutes of collection 2 ml of whole blood was added to 4 ml of an ammonium free cation exchange resin (Dowex 50W by 12;200-400 mesh) in a graduated glass stoppered centrifuge tube. The tube was stoppered and shaken vigorously for 3 minutes and then allowed to stand until the resinammonium complex had settled by gravity. The supernatent was decanted carefully and ion-free water added to the 6 ml graduation mark. The tube was shaken for 30 seconds and allowed to settle before decanting. This was repeated for a total of 4 washes. 6 ml of 1.5 diluted Nessler's Reagent were then added and the tube shaken for 3 minutes. The Nesslers Reagent was then decanted and the absorption measured at 415 mp in a spectrophotometer.

The concentration of ammonia in rumen liquor was determined by a modification of the method of Waite and Wilson (1968). Freshly drawn rumen liquor was strained through muslin. 1 ml of liquor strained rumen/was then added to 9 ml of a solution of acidified sodium tungstate (0.75 % sodium tungstate in 0.075 N sulphuric acid). Following centrifugation at 3,000 r.p.m. for 10 minutes, 5 ml of the clear deproteinised supernatent were placed in a Hoskins distillation apparatus to which was added 2 ml of 20 % sodium hydroxide. The liberated ammonia was steam distilled into 10 ml of 2 % boric acid containing methylene red and methylene blue as a mixed indicator. This solution was titrated against standard (approximately 0.015 N) hydrochloric acid.

Blood glucose was determined following deproteinisation with uranyl acetate. The method was based on that described by Werner, Rey and Wielinger (1970) using Boehringer Mannheim standardised reagents.

Plasma free fatty acids were determined by a modification of the method of Itaya and Ui (1965). Following solvent extractions of plasma free fatty acids with chloroform in the presence of a phosphate buffer (pH 6-7), the chloroform phase was treated with Cu-triethanolamine solution. After shaking and allowing to stand for 5 minutes the Cu-triethanolamine solution was aspirated with a fine-tipped pipette. The residual chloroform layer was filtered and then sodium di-ethyldithiocarbamate solution was added. The yellowish brown colour which developed almost immediately was measured at 440 mp in a spectrophotometer.

Quantitative analyses for volatic fatty acids in rumen liquor were determined by gas liquid chromatography following treatment with 20 % w/<sub>v</sub> metaphosphoric acid. The procedure used was based on the method described by Mahadevan and Stenroos (1967) using 150-200 mesh untreated Poropak Q (Waters Associates, Stockport, Cheshire, England). REFERENCES.

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