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Effect of Supplementation with Urea, Natural Proteins or Protected Proteins of Plant or Animal Origin on the Utilization of Poor Quality Roughages. 1-Nutrients Digestibility and Nitroger Utilization.

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TWENTY-FOUR growing Rahmany male lambs of 6 month-old were alloted into 8 equal groups. The animals received wheat straw rations without nitrogen (N.) supplementation or with N. supplements of urea, untreated casein (UCAS), formaldehyde treated casein (FCAS), urea + FCAS, untreated decortecated cottenseed meal (UCSM), formaldehyde treated decortecated cottonseed meal (FCSM), or urea + FCSM. The experimental period lasted for 12 weeks. The digestibility and N-balance trials were carried out at the end of the *1st*, *5th* and *9th* weeks of the experimental poriod. The results showed that supplementation by urea, UCAS or UCSM increased nutrients digestibility compared with the results recorded for the control group Animals given the FCAS or FCSM supplements showed higher values for nutrients digestibility compared with those given urea, UCAS or UCSM supplements. Supplementation by both urea and FCAS or FCSM increased greatly nutrients digestibility compared with the values obtained for all other diets. Digestibility values recorded for FCAS diets were somewhat higher for nutrients digestibility than those recorded for FCSM diet. Animals given no N. supplementation were in negative N-balance, while the other groups were in positive N-balance supplementation by FCAS or FCSM slightly improved N-balance compared with the groups given urea, UCAS or UCSM. Lambs given the FCAS ar FCSM in combination with urea showed the highest values for N-balance compared with the other groups.

The present study demonstrated clearly the potentiality of using urea, natural or protected protein in improving nutrients digestibility and N-utilization of poor quality roughages by sheep. It also showed the superiority of animal protein on plant protein in this respect

Lack of concentrate feeds for ruminants in Egypt has emphasized the need for improving both the quality and intake of poor quality roughages by these animals. This can be achieved by physical or chemical treatments such as alkali treatment (See Jackson, 1977 and 1978).

Recent work (Kempton and Leng, 1979 ; Knipfel *et al.*, 1981 ; Martin *et al.*, 1981 ; Shoukry, 1982 and Soliman *et al.*, 1984 a) suggested that supplementing or spraying poor quality roughages with urea or urea solution can be an alternative method to alkali treatment which may involve different hazards for both human and soil (Jackson, 1977 and 1978). Also supplementation with protected protein has been found to improve feed intake,nutrients digestibility and nitrogen utilization of poor quality roughages by ruminants (Reis and Schinkel, 1963 ; Ferguson *et al.*, 1967 ; Faichney and Weston, 1971 ; Faichney *et al.*, 1973 ; Kempton, *et al.*, 1979 and Kempton and Leng, 1979).

Formaldehyde has been shown to be an effective mean of protecting dietary protein without rendering it indigestible in the small intestine (Ferguson *et al.*, 1967 and Reis and Tunks 1969). The method and level required for protecting different protein sources from rumen degredation by formaldehyde have been examined by many workers (Reis and Schinkel 1963 ; Ferguson *et al.*, 1967 ; Peter *et al.*, 1971 Faichney and Davis 1973 and Reynolds *et al.*, 1978).

The present work was therefore carried out to examine the effect of supplementing poor quality roughages (wheat straw) by urea, unprotected or protected protein sources (plant or animal origin) on nutrients digestibility and nitrogen utilization by growing lambs.

Materials and Methods

Method of protein protection

Two protein sources mainly casein as an animal protein source and decortecated cotton seed meal as a plant protein source available in Egypt were examined in this study.

Casein was treated with formaldehyde according to the procedure of Ferguson *et al.*, (1967).

Decortecated cottonseed meal (D.C.S.M.) was sprayed with different levels of formaldehyde (40%) calculated to provide 0·5,1 and 1·5 gm formaldehyde/100 gm protein. The treated D.C.S.M. was stored for 7 days in plastic bags in room temperature before being used. Dacron bag technique (Ørskov *et al.*, 1980) was used for determination of *In Situ* dry matter (DM) and Nitrogen (N) disappearance in the rumen of cannulated sheep for untreated or formaldehyde treated D.C.S.M. at different intervals mainly 4, 6, 8, 10, 12, 16, 20 and 24 hr. This technique was used for determination of the proper level of formaldehyde required for treating D.C.S.M.

Five mature male Ossimi sheep of about 40-50 kg live weight and 2 years of age were fitted with rubber rumen cannulae of about 5 cm internal diameter. The animals were maintained on a basal diet of berseem hay given at a rate of 2·5% of body weight /head/day in two equal meals, mainly at 8·00 and 17·00 hrs. They were given access to water twice daily.

About 5 gm of the grinded samples were weighed in each bag which was tied with nylon string. The other end of the string was tied to the top over the cannula through a wire hook fitted to it. The required number of bags were incubated in the rumen of each sheep (not more than 5 bags) for the required period of incubation. Each sample was weighed in 3 bags for each incubation period and incubated in 3 sheep (triplicates). After the required period of incubation, the bags (3) were removed from the sheep (3) and washed throughly in running tap water. They were then dried in an oven at 60° for 48 hr.

Disappearance of DM was calculated as the difference in weight of the sample before and after incubation. The remaining material in each of the three bags (triplicate determination for each incubation period) were pooled together, finely ground and its N content was determined. Disappearance of N was calculated as mentioned above for DM.

Digestibility trials

Digestibility trials were carried out with growing Rahmany lambs to determine nutrients digestibility and N-balance for 8 different experimental rations. A basal ration of wheat straw was used to study the effect of supplementation with urea, unprotected or protected protein sources (plant or animal origin), given alone or in combination, on nutrients digestibility and N-balance of lambs given these ration.

Animals and their management

Twenty-four Rahmany male lambs, of about 22 kg live weight and 6 month-old, were used in this experiment. The animals were divided according to their body weights in 8 groups of three lambs each, and within groups assigned at random to receive one of eight experimental rations. Each group was kept in a separated brick made pens for an experimental period of 12 weeks. The animals were offered their diets *ad-lib.* twice daily in equal parts daily in equal parts at 8·00 and 17·00 . Water and salt block were available freely to the animals. Eight animals (one from each group) were kept in metabolic crates for a preliminary period of 15 days and 7 days for faeces and urine collection. This procedure was repeated three times of 8 animals at the end of the 1*st*, 5*th* and 9*th* weeks of the experimental period.

During the collection period, faeces and urine were collected once daily at 8·00. Sample from rations offered, residues and faeces were taken, finely ground and kept in tight plastic containers for chemical analysis. Urinary samples were taken and kept in tight bottles at +1° until nitrogen determination was done.

Experimental rations

Eight different experimental rations were tested in this experiment. They were based mainly on chopped wheat straw given *ad-lib.* as a basal ration. A supplement of ground co-op-feed mix*. was given at the rate of 100 gm DM/animal/day to each animal. Supplements of urea formaldehyde, treated or untreated casein or D.C.S.M. or combinations of them were added to the basal ration according to treatment as shown below.

Ration

No.

I Wheat straw (basal ration).

II Wheat straw + 2% urea (20 gm urea per kg wheat straw) spray.

III Wheat straw + untreated casein which was calculated to supply the same amount of nitrogen derived from urea as in ration II.

IV Wheat straw + casein treated with 1% formaldehyde calculated to supply the same amount of nitrogen derived from urea or untreated casein (rations II and III).

V Wheat straw + 2% urea + protected casein similar to that in ration III.

VI Wheat straw + untreated D.C.S.M. calculated to supply an amount of nitrogen equal to that of 2% urea.

VII Wheat straw + 1% formaldehyde treated D.C.S.M. calculated to supply an amount of nitrogen equal to that of 2% urea.

VIII Wheat straw + 2% urea + protected D.C.S.M. similar to that of ration VII.

Urea was dissolved in a proper volume of water (50 gm/ Liter) then sprayed on wheat straw, mixed properly and left to dry in the sun for two days. Sodium sulfate was dissolved in the urea solution at the rate of 10% of urea DM.

The amount of co-op-feed mix. and protected or unprotected protein sources were added to diluted cane molasses (calculated to provide 2% of the weight of wheat straw) to ensure good mixing and the mixture was mixed with wheat straw according to the treatment.

Chemical analysis

Dry matter, CP, CF and Ash of the ration used, faeces, residues and DM and N content of sample of protected or non-protected D.C.S.M. before and after incubation ; and urinary N were determined according to A.O.A.C. (1970) procedures.

* Consisted of 37% undecortecated cottonseed meal, 25% ground yellow corn, 15% rice germ meal, 3% molasses, 15% wheat bran, 5% lime stone and minerals El-Sabe Tanta feed mill Co.

Statistical analysis

Data obtained for nutrients digestibility and N-balance along with that of *In Situ* DM and N-disappearance were statistically analyzed according to Snedecor and Cochran (1967).

Results

1. *Protection of casein and cottonseed meal proteins from rumen degradation*

1. 1. *Casein*

Casein protein was protected from rumen degradation using the method described by Ferguson *et al.*, (1967). No *In-Situ* studies have been done to determine disappearance of casein in the rumen because of difficulties encountered in incubating untreated or formaldehyde treated casein in the rumen. Incubated samples of casein formed a jelly like material in the dacron bags and this led to large variations in determining DM and N-degradability between animals and between bags incubated in the same animal. A part from possible differences between bags in fermentation rate in the rumen, washing of the bags after incubation represented also a serious problem. It was therefore decided to use the method of Ferguson *et al.*, (1967) for casein. These results showed that, although dacron bag technique is a very useful tool in measuring rate and extent of disappearance of different feeds, it cannot be used with casein.

1.2. *Decortecated cottonseed meal (D.C.S.M.)*

Problems similar to those reported with casein were not encountered with D.C.S.M., and in this case the dacron bag technique proved to be a very useful tool.

Results of *In Situ* disappearance of untreated or treated D.C.S.M. with different levels of formaldehyde increased significantly ($P < 0.01$) as the period of incubation increased from 4 to 24 hr. Rate of disappearance of DM and N. in all cases was highest during the first 4 hr. (see Tables 1 and 2).

In Situ disappearance of DM and N of D.C.S.M. decreased significantly ($P < 0.01$) with increasing the level of formaldehyde up to 1% (see Tables 1 and 2). No significant difference in DM and N disappearance were detected between 1 and 1.5% formaldehyde level, indicating that the 1% level of formaldehyde was suitable for treating D.C.S.M. under the present experimental conditions. This level was therefore used for treating D.C.S.M. used in the digestibility and N-balance trials and was compared with casein treated with formaldehyde according to Ferguson *et al.*, (1967) as mentioned earlier in the materials and methods.

2. *Nutrients digestibility*

Results concerning nutrients digestibility and N-balance values recorded for lambs given different nitrogen supplements are presented in Table 3.

TABLE 1. *In Situ* DM disappearance (%) of treated or untreated decortecated cottonseed meal with different levels of formaldehyde at different incubation periods

Treatment	Incubation period (hrs.)								Mean
	4	6	8	10	12	16	20	24	
Control	37.6	40.5	50.5	54.4	58.9	59.5	62.9	66.3	53.8^{A}
S.E.	±0.90	±0.21	±0.61	±0.47	±0.30	±0.38	±0.15	±0.30	
½% Formaldehyde	27.3	30.7	45.1	44.7	53.8	53.7	55.7	65.8	47.1^{B}
S.E.	±0.56	±0.27	±0.55	±0.06	±0.37	±0.29	±0.56	±0.52	
1% Formaldehyde	25.3	26.2	39.5	42.5	46.7	49.9	53.5	60.3	43.0^{C}
S.E.	±0.20	±0.23	±0.15	±0.47	±0.26	±0.12	±0.38	±0.93	
1½ % Formaldehyde	24.7	24.7	32.7	38.2	48.4	53.5	51.9	60.8	41.9^{C}
S.E	±0.15	±0.75	±0.26	±0.46	±0.38	±0.51	±0.32	±0.86	

A,B,C Means with different superscripts are significantly ($P < 0.01$) different.
S.E. Standard error.

TABLE 2. *In Situ* N-disappearance (%) of treated or untreated decortecated cottonseed meal with different levels of formaldehyde at different incubation Periods.

Treatment	Incubation period (hrs.)								Mean
	4	6	8	10	12	16	20	24	
Control	35.8	48.1	56.6	62.3	73.2	75.5	79.5	84.4	64.4^{A}
S.E	±0.48	±0.58	±0.98	±0.29	±0.58	±0.30	±0.35	±0.50	
½% Formaldehyde	29.8	37.5	56.0	55.6	61.6	68.9	76.3	81.6	58.4^{B}
S.E.	±0.06	±0.63	±0.58	±0.61	±0.38	±0.30	±0.29	±0.32	
1% Formaldehyde .	29.1	30.9	45.5	52.8	59.4	64.1	71.6	75.5	53.6^{C}
S.E.	±0.06	±0.43	±0.52	±0.58	±0.60	±0.52	±0.41	±0.43	
1½% Formaldehyde	29.5	27.8	41.1	53.2	60.3	65.2	71.1	76.8	53.1^{C}
S.E.	±0.30	±0.06	±0.61	±0.66	±0.20	±0.22	±0.38	±0.44	

A,B,C Means with different superscripts are significantly ($P < 0.01$) different.
SE = Standard error.

TABLE 3. Nutrients digestibility and N-balance recorded on lambs given rations containing different nitrogen supplements.

Treatment number	Nutrients digestibility %				N-balance g/ Animal/day
	DM	OM	N	CF	
1. Wheat straw	50.6	54.2	46.4	53.2	$-0.22^{A,C}$
2. Wheat straw + urea	54.5	61.0	57.5	60.1	$1.03^{A,B,a,b}$
3. Wheat straw+untreated casein	54.8	62.9	57.0	62.7	$1.07^{A,B,a,b}$
4. Wheat straw+treated casein	57.8	63.1	59.6	63.4	1.16^{B}
5. Wheat straw+urea +treated casein	59.6	64.6	67.7	67.8	$2.12^{B,a}$
6. Wheat straw+decortecated cottonseed meal (DCSM)	54.7	60.8	57.5	59.2	$0.87^{A,B,b}$
7. Wheat straw+treated D.C.S.M.	55.4	62.4	61.5	60.9	$1.14^{A,B,ab}$
8. Wheat straw+urea +treated D.C.S.M	56.3	63.3	62.4	61.4	1.69^{B}

a,b,c Means with different superscripts are significantly ($P<0.05$) different.
A,B, Means with different supercripts are significantly ($P<0.01$) different.

The results showed that supplementation by urea only increased DM, OM, N and CF digestibility compared with the results recorded for the control group.

Supplementation by untreated casein (Treatment 3) or D.C.S.M. (Treatment 6), caused very little change in DM, OM, N and CF digestibility compared with urea supplementation. The group given the on-treated casein supplement (Treatment 3) showed slightly higher values for OM and CF digestibility compared with the groups given urea (Treatment 2) or untreated D.C.S.M. (Treatment 6).

Animals given the formaldehyde treated casein supplement showed higher values for DM, OM, N and CF digestibility compared with those given the urea supplements.

Digestibility of DM, OM, N and CF recorded for the treated casein group was slightly higher than the values obtained for non-treated casein.

Animals given formaldehyde treated D.C.S.M. supplement showed slightly higher values for DM, OM, N and CF digestibility compared with those given untreated D.C.S.M. supplement.

Digestibility values recorded for the treated casein diet were somewhat higher for DM, OM and CF than those recorded for the treated D.C.S.M. diet, which had slightly higher value for N digestibility.

Supplementation by both urea and protected casein increased greatly DM, OM, N and CF digestibility compared with the values obtained for urea or treated casein diets given separately and exceeded the values obtained for all other diets.

Similar supplementation by urea and protected D.C.S.M. caused nearly no change in nutrients digestibility compared with the protected D.C.S.M. diet. Differences in nutrients digestibility were not, however, statistically significant possibly because of large variations within groups.

3. *Nitrogen balance*

The results presented in Table 3, showed that although the group given the control ration was in negative nitrogen balance all other groups were in positive N-balance. Inclusion of urea or either untreated casein or D.C.S.M. changed the N-balance from the negative to the positive side for all the three groups (Treatment 2, 3 &6) with no significant differences between these three supplements. Supplementation by formaldehyde treated casein or D.C.S. M-slightly improved N-balance compared with the groups given urea or untreated D.C.S.M. or casein.

Lambs given the formaldehyde treated casein or D.C.S.M. in combination with urea showed the highest values for N-balance compared with the other groups. Nitrogen balance values recorded for the formaldehyde treated casein group were higher than those recorded for the formaldehyde treated D.C. S.M. group when either of them was given in combination with urea.

Discussion

The results obtained in the present study showed clearly that treating D.C.S.M. with different levels of formaldehyde greatly decreased *In Situ* disappearance of DM and N. of D.C.S.M. Similar results have been reported by Peter *et al.*, (1971) ; Faichney and Weston (1971); Sharma *et al.*, (1972) ; Schoeman *et al.*, (1972) Lyon *et al.*, (1976) and Soliman *et al.*, (1984 b). They found that treatment of protein source with formaldehyde decreased ruminal ammonia production, Solubility and degradability of protein sources.

The present results indicated that the 1% level of formaldehyde was suitable for treating D.C.S.M. This level was therefore used for treating D.C. S.M. used in digestibility trials. In these trials, treated D.C.S.M. was used as a supplement for a basal ration of wheat straw and was compared with other supplements such as urea, protected or unprotected casein or uprotected D.C.S.M. which were given alone or in combination to growing lambs in an attempt to improve utilization of poor quality roughages.

It may be argued that the choice of wheat straw is not a successful example of low quality roughages since its price/ton has now exceeded that of subsidised corn. However lack of facilitates for grinding and pelleting of rice

straw which was intended to be used in this experiment led to choice of wheat straw which is know sold in a properly ground form for use as bedding in poultry farms and these facilitates its mixing with the nitrogen supplement.

The present results showed clearly that unsupplemented wheat straw when given as the main feed to growing lambs resulted in low nutrients digestibility. The decreased nutrients digestibility may have been related to the possibility that N-requirements for rumen micro-organisms may have not been met on this treatment. It was also possible that lack of available energy for rumen microbes or at the tissue level may have been responsible for the very poor nutrients digestibility recorded for this group as also evidenced by their negative N-balance.

Urea or unprotected casein or D.C.S.M. supplementations to wheat straw, (Treatment 2, 3 and 6) greatly improved nutrients digestibility and N-balance. Similar results have been reported by Campling *et al.*, (1962); Ammerman *et al.* (1972) ; Kempton and Leng (1979) ; Knipfel *et al.*, (1981) ; Martin *et al.*, (1981) ; Shoukry, 1982 and Soliman *et al.*, (1984a) for animals given low quality roughages supplemented by urea or unprotected proteins. The improved nutrients digestibility and N-balance with nitrogen supplementation may have been due to the possiblity that the requirements of nitrogen for rumen microbes have been at least partially or completely met (Krause and Klopfenstein, 1978 ; Kempton and Leng, 1979 ; and Soliman *et al.*, 1983). This assumption was substantiated by the improved nutrients digestibility and positive N-balance recorded for these treatments compared with the control treatment.

The slight improvement in nutrients digestibility and N-balance with casein compared with D.C.S.M. supplementation may be considered as an indicative to certain essential amino acid requirements by the rumen-micro-organisms. Apart from other essential amino acids, casein contains higher concentrations of sulfur containing amino acids than D.C.S.M. (See Schaible, 1970) and this may explain the improvement in nutrients digestibility and N-utilization for the casein group over D.C.S.M. group. The possibility that differences in the type of branched chain fatty acids produced from deamination of casein or D.C.S.M. in the rumen and required by rumen micro-organisms. may have also been responsible for the slight differences obtained between the two supplements, cannot be rulled out (Ørskov, 1982).

The results of the present study showed clearly that supplementation with protected casein or D.C.S.M. greatly increased nutrients digestibility and N-balance for growing lambs over those given urea or unprotected casein or D.C.S.M. Similar results have been reported by Ferguson *et al.*, (1967); Peter *et al.*, (1971); Sharma *et al.*, (1972); Faichney *et al.*, (1973); Kempton and Leng (1979) and Kempton *et al.*, (1979). This has been on the basis of improved N-status of the animals (Egan and Moir, 1965; Egan, 1966; Weston, 1971; Faichney and Weston, 1971, Weston, 1973; Faichney, 1974; Egan, 1977 and Kempton and Leng, 1979).

The slight improvement recorded for the group given protected casein over those given protected D.C.S.M. may have been partially related to differences in the extent of protection achieved for the two protein sources. It is possible to speculate that degradability of treated casein was lower than that of D.C.S.M. It follows that more casein protein may have reached the abomasum and the small intestine compared with D.C.S.M. (Crooker *et al.*, 1983) and this may have been responsible for improved nutrients digestibility and N-utilization with protected casein over that achieved with protected D.C.S.M. In this case the improved nutrients digestibility and N-balance with protected casein may be resulting from partial degradation in the rumen and digestion in small intestine of both protein sources.

The results obtained in this study showed the superiority of wheat straw supplemented with both urea and protected proteins treatments over all treatments studied. Similar results have been reported by Egan and Moir (1965); Kempton and Leng (1979); Kempton *et al.*, (1979) and Stock *et al.*, (1981). This was probably related to the possibility that addition of urea may have satisfied the microbial need for N on one hand and protected proteins may have satisfied at least partially the need of the host animal for proteins on the other hand. The superiority of casein over D.C.S.M. in supporting nutrient digestibility and N-balance in the presence of urea may have been due to differences in amino acids composition between the two sources as discussed earlier.

Finally, the present results demonstrated clearly the potentiality of using urea, natural or protected proteins in improving nutrients digestibility and N-utilization of poor quality roughages by sheep. It also showed the superiority of animal protein on plant protein in this respect. Further work is needed to examin the effect of these supplements on feed intake and the performance of growing lambs given poor quality roughages in their rations.

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تأثير استخدام الاضافات من اليوريا أو البروتينات الطبيعية غير المحمية أو المحمية ذات الاصل النباتى أو الحيوانى على الاستفادة من المواد الخشنة الفقيرة ـ ١ معاملات الهضم والاستفادة من النيتروجين

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أجريت هذه الدراسة باستخدام ٢٤ من ذكور الحملان الرحمانى النامية عمر ٦ أشهر ، وقد قسمت هذه الحيوانات الى ثمانية مجاميع متساوية فى العدد ، وغذيت حيوانات المجموعة الأولى (المقارنة) على عليقة تحتوى على تبن القمح دون أى اضافة نيتروجينية ، وغذيت المجاميع الأخرى على نفس العليقة مع اضافة أحد الاضافات الآتية : يوريا فقط ، كسب قطن مقشور غير معامل ، كسب قطن مقشور معامل بالفورمالدهيد ، يوريا + كسب قطن مقشور معامل بالفورمالدهيد ، كازين غير معامل ، كازين معامل بالفورمالدهيد ، يوريا + كازين معامل بالفورمالدهيد . وقد استغرقت هذه التجربة ١٢ أسبوعا وأجريت تجارب الهضم والاتزان الأزوتى فى نهاية الأسبوع الأول والخامس والتاسع .

وقد أوضحت النتائج أن اضافة اليوريا والكازين غير المعامل أو الكسب غير المعامل قد أدت الى زيادة معاملات هضم المركبات المختلفة بالمقارنة بعملية المقارنة . كما أدت اضافة كل من الكازين المعامل أو الكسب المعامل الى زيادة معاملات الهضم بالمقارنة بالعلائق المحتوية على اليوريا أو الكازين أو الكسب غير المعامل . أدت اضافة اليوريا مع الكازين المعامل أو الكسب المعامل الى زيادة كبيرة فى معاملات الهضم بالمقارنة بجميع العلائق الأخرى . وبصفة عامة لوحظ أن اضافة الكازين المعامل أو غير المعامل كان له تأثير أكبر على زيادة معاملات الهضم عن الكسب المعامل أو غير المعامل . أعطت الحيوانات المغذاة على عليقة المقارنة ميزان آزوت سالب بينما أعطت بقية مجاميع الحيوانات ميزان آزوت موجب . وقد أعطت الحيوانات المغذاة على الكازين المعامل أو الكسب المعامل قيم ميزان آزوت أعلى من القيم التى أعطتها الحيوانات المغذاة على علائق تحتوى على اليوريا أو الكازين غير المعامل . سجلت الحيوانات المغذاة على علائق تحتوى على كل من اليوريا مع الكازين المعامل أو الكسب المعامل أعلى قيم لميزان الآزوت .

وبصفة عامة فقد أكدت هذه الدراسة امكانية زيادة القيم الهضمية والاستفادة من النيتروجين للمواد الخشنة الفقيرة باستخدام الاضافات من اليوريا أو البروتينات المحمية أو غير المحمية ، ويلاحظ أفضلية استخدام البروتينات الحيوانية عن النباتية فى هذا المجال .