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DIGESTIBILITY OF CORN COBS AFTER AMMONIATION BY THE RUMINANT MICROFLORA

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The effect of ammoniation on the chemical composition and digestibility of corn cobs was studied. The increases in dry matter digestibility was more than 80 and 140% on treatment with 5% aqueous ammonia and 4% sodium hydroxide respectively. Maximum increase in dry matter digestibility was found to be almost 200% when corn cobs were successively treated with 4% sodium hydroxide and 5% aqueous ammonia. An improvement in the digestibility of cellulose, minerals and organic matter was also observed after these treatments. A two fold increase in nitrogen contents was observed at a level of 1% aqueous ammonia. However, no significant increase in nitrogen contents was noted by increasing the level of ammonia from 1 to 5%. Reduction in lignin contents was also found after ammonia treatment.

Key words: Corn cobs, Ammoniation, Digestibility.

Introduction

Utilization of crop residues as animal feed is limited due to their poor digestibility and low nitrogen contents. During the past few years, a number of methods had been suggested to increase the digestibility of these materials [1-4]. It had been reported by many workers [5,7] that the rate of degradation of cellulosic materials can be increased by treatments with various alkalies. The findings of Lewis *et al.* [8] and Gould [9] revealed that the digestibility of different crop residues was significantly improved after treatment with alkaline/hydrogen peroxide. In several European countries, ammoniation is commercially applied and widely used for increasing the digestibility of the crop residues [10]. Hartley and Jones [11] observed an increase in the organic matter digestibility of barley straw after ammonia treatment. It had also been observed by Chang *et al.* [12] that the ammoniation of bagasse resulted an increase in nitrogen contents and cellulose digestion.

The present work was undertaken to study the effect of ammonia on the chemical composition and digestibility of corn cobs.

Material and Methods

Corn cobs (without corns) were collected from the local market to carry out these studies. All the corn cobs were ground to 20 mesh size in a hammer mill at the same time before drying at 100° for 4 hrs. Dried ground material was subjected to different chemical treatments as described below;

Ammonia treatment. The material was thoroughly mixed with the solution of aqueous ammonia of different concentrations (1.0-5.0% w/w), keeping 20% moisture contents. The treated material was placed in air tight glass jars at 55 ± 5° for 15 days. After which the material was exposed to air for some time and then it was dried at 100 ± 2° for 4 hrs.

Alkali treatment. The material was thoroughly mixed with 4.0% solution (prepared in tap water) of sodium hydroxide or potassium hydroxide or 2.0% calcium hydroxide (w/w) keeping 20% moisture contents in the substrate. The treated material was placed in air tight glass jars at room temperature for 24 hrs. after that the glass jars were opened and the material was dried at 100 ± 2° for 4 hrs.

Combined treatment. Alkali treated material was also subjected to 5% ammonia treatment as described above.

Digestibility trials. Digestibility of the material was estimated in nylon bags as described by Orskave *et al.* [13]. A dry "Sahiwal" Cow was fitted with a soft rumen cannula (10 cm internal diameter). 10-15 gms of representative sample was placed in nylon bags (in six replicates) and these were introduced in the rumen of cow at the same time and taken out after 48 hrs. These were thoroughly washed with tap water, followed by ethanol and finally with distilled water and then dried at 100 ± 2° to constant weight. Digestibility of the samples alongwith standard deviation (in six replicates) was calculated by the method of Snedecore [14].

Chemical analysis. Nitrogen was estimated by a micro-Kjeldahl method [15] using $CuSO_4$-K_2SO_4-SeO_2 (1:9:0.02) mixture. Dry matter and ash contents were determined according to AOAC methods [16], cellulose was determined by Kurschner and Hanak method [17]. Lignin contents were estimated by the procedure as described by Van Soest and Wine [18] using 72% sulphuric acid.

Results and Discussion

Digestibility of corn cobs after ammonia treatment. The effect of ammoniation on the chemical composition and digestibility of corn cobs is given in Table 1. Nitrogen contents of corn cobs showed an increase at all levels of aqueous ammonia (1-5% w/w). Maximum increase in nitrogen contents was about 100% (0.8-1.6%) at a level of 1% aqueous ammonia. However, no significant increase in nitrogen contents was noted by increasing the level of ammonia from 1 to 5%. In fact, the proportion of ammonia-N which was captured by corn cobs, decreased from about 98% ammonia to less than 25% with 5% ammonia. Therefore, it is apparent that with 1% ammonia the corn cobs were almost saturated. Thus the nitrogen contents were not increased significantly beyond 1% level of NH_3. The increase in nitrogen contents might be due to formation of ammonium salts as a result of ammonia treatment. Han and Anderson [19] also reported a significant increase in nitrogen contents due to reaction of ammonia with straw components to form nitrogenous compounds. Reduction in lignin contents was also noticed at all levels of ammonia. Maximum reduction in lignin contents by 15% (13.03 - 11.06%) was observed when the level of ammonia was 5%. However, the reduction in lignin contents was not too much when the level of ammonia was less than 5%. The ash contents almost remained unchanged while a slight increase in cellulose was observed after ammoniation. A slight increase in cellulose after treatment of straw with alkali had also been noted by other workers [20]. However, this phenomenon is still unexplained. The results mentioned in Table 1 indicate that the digestibility of the corn cobs improved progressively as the amount of aqueous amonia was gradually increased from 1% to 5% (w/w). Treatment with 5% aqueous ammonia was found to be most effective in increasing the digestibility of corn cobs. Maximum increase in dry matter digestibility was found to be more than 80% with 5% aqueous ammonia whereas it was only 53% with 1% aqueous ammonia. A significant improvement in the digestibility of cellulose, minerals and organic matter was also observed as a result of ammoniation. These results are in agreement with the findings of Singh and Gupta [21] who reported that ammonia treatment increased the swelling action of fibre as well as induced structural changes in cellulose and solubilized some of the lignin and hemicellulose which ultimately increased the digestibility of the straw.

Digestibility of corn cobs after alkali treatment. Results mentioned in Table 2 show the effect of different alkalies [(NaOH, KOH, Ca $(OH)_2$] with and without ammonia on the chemical composition and digestibility of corn cobs. It is apparent from these results that sodium hydroxide was most effective in improving the digestibility of corn cobs. Treatment with 4% sodium hydroxide (w/w) resulted a decrease in lignin content upto 15%. Sodium hydroxide treatment increased remarkably the ash contents but showed no effect on nitrogen contents. Similar results were also obtained in case of potassium hydroxide and calcium hydroxide. The increase in ash contents might be due to formation of complex salts of sodium, potassium or calcium with phenolic compounds (particularly with phenolic acids). It had already been reported by Morrison [22] that the phenolic compounds of lower molecular weight were formed due to degradation of lignin under alkaline conditions. A slight increase in cellulose was also noted after treatment with these alkalies. Garrett *et al.* [23] also observed an increase in cellulose contents alongwith a significant decrease in lignin due to the action of sodium hydroxide with rice straw. It is clear from these results that the digestibility of corn cobs after treatment with different alkalies (NaOH, $Ca(OH)_2$) was significantly higher than those of ammoniated corn cobs (Table 2). The sodium hydroxide treatment increased dry matter digestibility by about 140% whereas calcium hydroxide improved dry matter digestibility of corn cobs by 110%. The digestibility of cellulose, minerals and organic matter was also affected significantly by alkali treatment. Maximum increase in cellulose digestibility was observed in case of sodium hydroxide treatment (Table 2). There was a marked increase in the digestibility of minerals

TABLE-1. CHEMICAL COMPOSITION AND DIGESTIBILITY OF CORN COBS AFTER AMMONIA TREATMENT.

	Untreated	1.0% NH_3	2.0% NH_3	3.0% NH_3	4.0% NH_3	5.0% NH_3
*% Chemical composition**						
Ash	1.69	1.85	1.83	1.88	1.85	1.84
Nitrogen	0.80	1.61	1.63	1.65	1.70	1.75
Cellulose	38.67	40.11	41.32	41.47	41.79	41.99
Lignin	13.03	12.94	12.87	12.53	12.09	11.06
Dry matter	93.57	90.65	89.71	89.53	88.88	88.43
*% Digestibility***						
Dry matter	23.0±2.1	35.2±1.3	35.9±1.7	38.3±2.0	39.0±1.6	41.9±2.0
Cellulose	20.3±1.3	37.2±2.6	33.0±2.7	38.8±1.3	38.4±1.7	34.3±2.7
Minerals	38.1±1.9	42.8±3.4	40.5±2.5	41.2±2.5	46.9±2.6	41.4±4.5
Organic matter	22.7±2.4	34.9±1.4	36.0±1.7	37.4±4.5	39.5±2.7	38.6±3.7

*Average of three replicates; **Average of six replicates alongwith standard deviation

TABLE 2. CHEMICAL COMPOSITION AND DIGESTIBILITY OF DIFFERENT ALKALIES TREATED CORN COBS WITH AND WITHOUT AMMONIA TREATMENT.

	4.0% NaOH	+5.0% NH_3	4.0% KOH	+5.0%NH_3	2.0% $Ca(OH)_2$	+5.0%NH_3
*%Chemical composition**						
Ash	6.71	7.05	6.39	6.23	4.71	3.05
Nitrogen	0.84	1.75	0.8	1.45	0.81	1.85
Cellulose	42.05	42.37	41.95	41.72	41.88	42.06
Lignin	11.05	10.28	12.77	11.66	12.66	11.86
Dry matter	88.53	88.23	88.12	86.95	89.67	88.79
*%Digestibility***						
Dry matter	55.9±1.7	68.8±4.3	41.5±2.2 5	9.5±2.9	48.5±2.2	57.5±2.2
Cellulose	56.0±1.4	62.9±1.4	33.7±2.2	46.3±1.2	46.0±3.2	45.9±3.2
Minerals	90.3±1.7	95.5±1.5	90.4±2.5	84.6±0.8	75.5±3.4	65.5±3.0
Organic matter	50.0+2.7	62.2+0.6	36.6+3.4	54.3+2.4	45.5+5.9	51.6+1.3

*Average of three replicates; **Average of six replicates alongwith standard deviation

after treatment with various alkalies. However, the digestibility of minerals after treatment with calcium hydroxide was found to be comparatively less as compared with other treatments. Improvement in digestibility may be attributed to greater availability of cellulose for microbial digestion after the removal of some of the lignin from the plant cell wall [24]. Tarkow and Feist [25] also postulated that the main consequences of sodium hydroxide treatment was the saponification of ester bonds linkages between lignin and hemicellulose which changed the crystalline structure of lignocellulose and improved the digestibility of cellulosic materials.

Digestibility of alkali treated corn cobs after ammonia treatment. It is also clear from the results presented in Table 2 that the digestibility of alkali treated corn cobs was further improved after ammoniation. There was a significant increase in nitrogen contents after the ammoniation of corn cobs which were already treated with different alkalies (NaOH, KOH, $Ca(OH)_2$). However, maximum increase in nitrogen contents was by 130% after calcium hydroxide and ammonia treatment. These results also revealed that the proportion of ammonia-N which was captured by cobs was 24% in case of sodium and calcium hydroxide, whereas it was only 16% after ammoniation of potassium hydroxide treated corn cobs. It seems to be that potassium salts of phenolic compounds might have less affinity for ammonia molecules. Therefore, potassium hydroxide treated cobs captured lower amounts of ammonia-N as compared with other treatments. However, this phenomenon is still unexplained. The amount of cellulose contents remained more or less unchanged as a result of these treatments. Lignin contents were further reduced after the ammoniation of alkali treated corn cobs. Maximum reduction in lignin contents (more than 20%) alongwith an increase in dry matter digestibility (about 200%) was observed when corn cobs were successively treated with 4% NaOH and 5% NH_3. Potassium hydroxide and calcium hydroxide treated corn cobs after ammoniation also improved dry matter digestibility by 150%. An improvement in the digestibility of cellulose, minerals and organic matter was observed with these treatment. However, a decrease in the digestibility of minerals was observed after treatment with calcium hydroxide and ammonia. This might be due to formation of insoluble salts of calcium in excess amount which inhibit the growth of rumen micro organisms on the substrate.

It can be concluded from these findings that sodium hydroxide treatment was found to be more effective in decreasing the lignin contents and increasing the digestibility of corn cobs than ammonia treatment. However, sodium hydroxide pretreatment of corn cobs together with ammoniation showed better results than the individual treatments. It is a well known fact that lignin content of ronghage is inversely related to its digestibility, the approach of the present study was to increase the degradability of corn cobs by attacking the lignin moiety. Ammonia was chosen for this purpose, since its small, highly reactive and penetrating molecules, can attack carbon carbon double bonds of the phenolic constituents of lignin. In fact, reduction in lignin content and the concomitant increase in dry matter digestibility shows that ammonia acts efficiency on sodium hydroxide treated corn cobs. These observations are consistent with the previous studies [26,27] showing that the partial delignification may be sufficient to increase the degradability of a cellulosic substrate by micro- organisms. The decrease in lignin contents probably created large spaces within the cellulose matrix, thus enhancing the accessibility of cellulolytic enzymes into cellulosic wastes. This hypothesis is supported by the increased digestibility of corn cobs after different treatments.

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