JOURNAL OF PURE AND APPLIED MICROBIOLOGY, October 2011. Vol. **5**(2), p. 769-774

Animal Feed Utilization using Anaerobic Microbial Flora in Cattles

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(Received: 18 December 2010; accepted: 21 January 2011)

Anaerobes grow in the absence of oxygen were predominant in the rumen region of the cattles such as goat, sheep and cow. The role of anaerobic microbial flora in cattles is very broad and the intensive research in this area was carried out here with the special reference to evaluation of plant fibre degradation by anaerobic microflora. The plant fibre degradation in cattle was evaluated by anaerobic microbial flora. The results reveal that the anaerobic fungi play a major role in plant fibre degradation than the bacteria and protozoa. The ruminants feed on cellulosic agricultural by products like cereals straws,stovers, sugarcane bagasse and tree foliages. The ruminants have the efficiency to utilize a wide variety of feeds due to the presence of bacteria, ciliate protozoa and anaerobic fungi. Hence the rumen of cattle has a rich potential of microbial diversity. The present study has been undertaken by collecting the rumen fluid from cattles such as sheep and goat to analyse the protozoa in the rumen fluid microscopically and by evaluating the fibre degradation activity by anaerobic flora.

Key words: Ananerobic Microflora, Plant fibre degradation, Ruminants.

The anaerobes inhabit the rumen in which the anaerobic microflora grows and help in the digestion of certain substances which are not digested by other microorganisms. The rumen is a dynamic continuous fermentation compartment that provides a suitable environment for a variety of species of anaerobic bacteria, protozoa and fungi. These microorganisms have a complex series of interactions with the foods supplied to the host. Due to the metabolic activity of all microbial populations, feeds were converted to microbial matter and the fermentation end products serve as nutrients for the ruminant(Gocmen & Atatur, 2002).

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Mostly the ciliates, protozoans and anaerobic fungi were predominating the rumen of the ruminants for the digestion of fibres. Ruminal ciliate, protozoans were metabolically versatile as they ferment carbohydrate including starch, hemicellulose and cellulose. They are proteolytic and some species hydrogenate unsaturated fatty acids, while others are able to denature fatty acids (Imai, 1986 and 1985).

The rumen harbours various types of bacteria which are active in degradation of those components of the feed.The interactions among themselves with other microbial groups in the rumen were responsible for synergistic effect on the production of volatile fatty acids and microbial proteins in the rumen. The cellulose degrading bacteria get stimulated when the buffaloes were fed with *S. cerevisiae* in their diets not will be due to the release of some undefined micronutrients by the yeast that are essentially required by the

cellulose degrading bacteria1(Grenet ,1989 and Hungate, 1955).

Protozoa are not essential to the ruminant digestive system, it doesn't mean that protozoa will not have a role in the rumen ecosystem but the characterization of the role of protozoa was complicated as the bacteria appear to occupy biological niches vacated by the ciliates. Hence the bacteria are present in greater concentration in defaunted animals than in animals with the ciliate protozoa. There are several studies on the rumen fauna of domestic cattle, sheep and goats (Gocmen and Atatur, 2002)

Rumen protozoa play an important role by contributing nutrients to the host animal also play an important role in the digestion of carbohydrates and proteins containing fed stuffs by secreting the saccharolytic and proteolytic enzymes (Shinchi ,*et al.,* 1986).

The obligate anaerobic fungi found in the rumen and also in the other parts of the gastro intestinal tract of herbivorous animals have an active and positive role to play in fibre degradation by the presence of different enzymes involved in the fibre degradation. The removal of fungi from the rumen results in the reduction of invitro gas production and degradation of fibrous feeds indicate a positive role played by fungi in fibre degradation. The rumen fungi produce high level of cellulose and hemicellulase was particularly proficient in producing xylanases. These organisms were better able to colonize and degrade the lignin containing tissues (Wallace *et al*, 1985 and Lowe *et al*., 1987).

The poor quality of lignocellulosic feeds were digested by the manipulation of rumen fermentation by increasing the number of lignocellulolytic microorganisms in the rumen (Giesecke,1970).Anaerobic fungi produces highly active enzyme for the lignocellulose degradation and have the ability to break and penetrate the fibrous feed particles through fungal mycelium by breaking the feed particles and provide more surface area for the action of other microbes (Grenet, 1989 and Mountfort, 1994).

Rumen fungi play an important catalytic role in the digestion of poor quality fibrous feed. There is a substancial variation among the fungal isolates from domestic as well as wild animals in their fibrolytic enzyme profile of ability to degrade fibrous feed. The microscopic techniques were used to detect the rumen fungi from the colonized lignin containing tissues of forages and was appeared to be active in the fibre degradation (Kopecney, 1995).

MATERIAL AND METHODS

Sample collection

The rumen fluid was collected from the Cattles such as Goat and Sheep from in and around Vellore and Arcot, Tamilnadu. India. The sample was collected in an air tight bottle. The sample was brought to the laboratory under anaerobic conditions for further investigations.

Microscopic examination of rumen fluid

One drop of the rumen fluid was taken and placed in a clean glassslide, over which a cover glass was placed and viewed under the microscope in low and high power objectives. The anaerobic micoflora was observed.

Identification of protozoa

Saline wet mount

One drop of saline was placed on a clean glass slide and a drop of the rumen fluid was added, over which coverslip was placed without air bubbles. Then the slide was placed in the microscope and observed under low and high power objectives for identification of protozoa (Fig – 1).

Saline – 0.85 gms of NaCl (Sodium chloride) was dissolved in 100 ml of distilled water.

Hay Infusion Broth

Composition

Decomposing Hay	-	50 g
Deionized water	-	1 L
Potassium monohydrogen phosphate	-	2 g

The decomposing hay was taken in water and autoclaved for 30 mins and then filtered.The potassium monohydrogen phosphate was added to the filtrate and pH was adjusted to 6.2 .

Evaluation of fibre degradation

The fibre degradation was evaluated using paddy straw and banana pseudostem. The fibres are made into small pieces of about 1 cm each. The rumen fluid was taken in the three airtight 3 container and labelled as Control(C), Test (1) and Test (2) respectively. All three airtight containers were filled with Hay infusion broth. To

the Test (1) air tight container, antibiotic solution streptomycin and penicillin was transferred. The cyclohexamide antibiotic was added to another air tight container Test (Coleman, 1974). The rumen fluid was inoculated in all three air tight containers except in the control. In all three containers paddy straw pieces was inoculated.The containers were incubated in room temperature for 3 days. The fibre degradation was calculated as follows.

Fibre degradation = Initial Weight of the fibre – Final Weight of the fibre

Similarly the procedure was also carried out for banana pseudostem

RESULT

The rumen fluid was collected from cattles such as goat and sheep in and around Vellore

Table 1. Abundances of protozoa in the rumen of goat and sheep

S. No	Name of the organisms	No. of Goats			Abundance (%)	No .of Sheeps			Abundance (%)
		1st	2nd	3rd		1st	2nd	3rd	
1.	Ciliates	4	4	3	3.6	2	1	2	1.3
2.	Flagellates	2	3	3	2.6	4	2	2	2.6

Table 2. Digestion characteristics of paddy straw inoculated with goat rumen fluid

S. No	Samples	Digestion Characteristics					
		Dry Weight Loss (mg)			Wet Weight Loss (mg)		
		Initial Weight (I)	Final Weight (F)	Total Weight (I-F)	Initial Weight (I)	Final Weight (F)	Total Weight (I-F)
1.	Control	0.05	0.05	0	0.08	0.08	0
2.	T1	0.06	0.04	0.02	0.14	0.05	0.09
3.	T2	0.05	0.04	0.01	0.12	0.07	0.05

C- Control
T1 – Rumen fluid with Streptomycin and Penicillin (For Fungi)
T2 – Rumen fluid with Cyclohexamide (For Bacteria)

Table 3. Digestion characteristics of paddy straw inoculated with sheep rumen fluid

S. No	Samples	Digestion Characteristics					
		Dry Weight Loss (mg)			Wet Weight Loss (mg)		
		Initial Weight (I)	Final Weight (F)	Total Weight (I-F)	Initial Weight (I)	Final Weight (F)	Total Weight (I-F)
1.	Control	0.05	0.05	0	0.09	0.09	0
2.	T1	0.07	0.04	0.03	0.12	0.06	0.06
3.	T2	0.08	0.06	0.02	0.10	0.06	0.04

C- Control
T1 – Rumen fluid with Streptomycin and Penicillin (For Fungi)
T2 – Rumen fluid with Cyclohexamide (For Bacteria)

and Arcot, Tamilnadu. India. The rumen fluids were directly examined under the microscope. In this observation, the protozoans are dominant than the other microbial flora. Hence the present investigation was focused on identification of protozoa by saline wet mount.

The maximum protozoan populations in goat rumen fluid were found to be ciliates (3.6%) and flagellates (2.6%) where as, the maximum protozoan populations in sheep rumen fluid were found to be flagellates (2.6%) and ciliates (1.3%).(Table 1)

In comparison of anaerobic protozoan in goat and sheep, ciliates are dominant in goat whereas flagellates are dominant in sheep. The diversity of protozoan population is due to feed inhabitat of the cattles.

The anaerobic microbial flora present in the rumen of cattle was evaluated for plant fibre degradation. The present investigations was carried out in two different cattle feed. The digestion characteristics of paddy straw inoculated with goat rumen fluid and sheep rumen fluid shows that maximum degradation activity due to fungi than the bacteria. (Table 2 & Table 3).

The digestion characteristics of banana pseudostem inoculated with goat rumen fluid and sheep rumen fluid shows that maximum degradation activity due to fungi than the bacteria. (Table 4 & Table 5).

The present investigation reveals the maximum degradation activity of plant fibre is due to anaerobic fungi than the bacteria because of the extra cellular enzyme production capability of the fungi and bacteria. The similar findings were also reported (Bauchop, 1981).

Table 4. Digestion characteristics of banana pseudostem inoculated with goat rumen fluid

S. No	Samples	Digestion Characteristics					
		Dry Weight Loss (mg)			Wet Weight Loss (mg)		
		Initial Weight (I)	Final Weight (F)	Total Weight (I-F)	Initial Weight (I)	Final Weight (F)	Total Weight (I-F)
1.	Control	0.08	0.08	0	0.15	0.15	0
2.	T1	0.09	0.05	0.04	0.20	0.09	0.11
3.	T2	0.09	0.06	0.03	0.18	0.09	0.09

C- Control
T1 – Rumen fluid with Streptomycin and Penicillin (For Fungi)
T2 – Rumen fluid with Cyclohexamide (For Bacteria)

Table 5. Digestion characteristics of banana pseudostem inoculated with sheep rumen fluid

S. No	Samples	Digestion Characteristics					
		Dry Weight Loss (mg)			Wet Weight Loss (mg)		
		Initial Weight (I)	Final Weight (F)	Total Weight (I-F)	Initial Weight (I)	Final Weight (F)	Total Weight (I-F)
1.	Control	0.08	0.08	0	0.13	0.13	0
2.	T1	0.07	0.05	0.02	0.18	0.07	0.11
3.	T2	0.07	0.06	0.01	0.16	0.10	0.06

C- Control
T1 – Rumen fluid with Streptomycin and Penicillin (For Fungi)
T2 – Rumen fluid with Cyclohexamide (For Bacteria)

Fig. 1. Microscopic view of Ciliate protozoa

DISCUSSION

Anaerobes are those which grow in the absence of oxygen. Such anaerobes are predominant in the rumen region of the cattles such as goat and sheep . No wide range of research was carried out to find out the characteristic features of rumen anaerobes and their role.

Thus, it is obvious that the role of anaerobic microbial flora in cattles is very broad. Hence, intensive research in the area was carried with special reference to evaluation of plant fibre degradation by anaerobic microflora.

The two major aspects of this study is to carry out this work to get the basic knowledge on anerobic microbial flora and their role in cattles of Vellore District, T.N, India.

The anaerobic protozoans present in rumen fluid were evaluated and the research shows that the maximum protozoan population is ciliates and minimum population is flagellates in goat and the similar were reported (Coleman,1980).While in case of sheep,the maximum protozoan population is flagellates and minimum population is flagellates which is similar to the study by (Giesecke, 1970).

The plant fibre degradation in cattle was evaluated by anaerobic microbial flora. The results reveal that anaerobic fungi plays a major role in plant fibre degradation than the bacteria.

ACKNOWLEDGEMNTS

We thank the Department of Microbiology, Jamal Mohamed College (Autonomous), Tiruchirappalli – 20, Tamilnadu, India for supporting us to carry out this work in a successful manner.

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