A Comparative Study on the Rumen Microbial Population of Cattle and Swamp Buffalo Raised under Traditional Village Conditions in the Northeast of Thailand

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ABSTRACT: A comparative study on rumen bacterial and protozoal population and fungal zoospores in cattle (Brahman × Native) and swamp buffalo (Bubalus bubalis) was conducted. Forty animals, twenty of each, with same sex and similar age which were raised under similar condition in the Northeast of Thailand, were used. Rumen digesta were sampled immediately post slaughtering for total microscopic counts of bacteria, protozoa and fungal zoospores. It was found that total bacterial population were higher in swamp buffalo that those in cattle (1.6 vs 1.36×10⁸ cells/ml) having more population of cocci, rods and ovals. Lower rumen protozoal pupulation in swamp buffalo with lower numbers of Holotrichs and Entodiniomorphs were found as compared to those in cattle. Significant higher fungal zoospore counts were in swamp buffalo than those in cattle being 7.30 and 3.78×10⁶, respectively. Study under electron microscope, revealed Anaeromyces sp. with acuminate apex were more predominant in the rumen of swamp buffalo. With these findings, cattle and swamp buffaloes showing differences in rumen bacterial, protozoal population and fungal zoospore counts, offer new additional information as why swamp buffaloes exhibit conditionally better than cattle especially during long dry season without green grass. (Asian-Aus. J. Anim. Sci. 2000. Vol. 13, No. 7: 918-921)

Key Words: Rumen, Bacteria, Protozoa, Fungi, Microorganisms, Swamp Buffaloes, Cattle

INTRODUCTION

Traditionally, swamp buffaloes (Bubalus bubalis) have been kept by small-holder farmers in Southeast Asia, as multi-purpose animals. According to De Haan et al. (n.a) there were 148 millions of buffaloes in the world and 99.9% are raised in the developing countries. The present numbers of swamp buffaloes in Thailand are 3 million heads and more than 80% are in the Northeast which were serving as a vital component in the mixed farming systems along with cattle and/or small ruminants. It has previously been reported that when cattle and buffalo were kept under similar conditions, buffalo utilize feed more efficiently with the digestibility of feeds being typically 2-3 percentage units higher (Wanapat, 1989; Wanapat et al., 1994; Kennedy and Hogan, 1994). Nitrogen utilization in swamp buffalo was found more efficient than that in Malaysian cattle (Devendra, 1985). The superiority is particularly noticeable in situations where the feed supply is of low quantity and/or quality. The reasons for the superior digestive capacity of buffalo over cattle have not been fully elucidated. However it is likely that much of the superiority may be explained by differences in the nature of rumen microbial population which would affect the type of fermentation occurring and the end-products resulting from fermentation. Thus, any variations between cattle and buffalo in the proportions and numbers of ruminal bacteria, protozoa and fungi might contribute to the explanation of differences in digestive capability due to fermentation end-products available for absorption and utilization by ruminants. It is, therefore, the objectives of this experiment to study on differences in rumen microorganisms between swamp buffaloes and cattle raised on similar condition in the Northeast of Thailand.

MATERIALS AND METHODS

The rumen samples for subsequent study of microbial populations were obtained from animals kept under traditional village conditions in the Northeast of Thailand. The experiment was conducted in September to April when swamp buffalo and crossbred cattle (Brahman×Native) were both grazing seasonally on available native grasses, rice stubble and rice straw. No concentrates were given to the animals at this time. A total of 40 animals which were being kept under similar conditions were identified, brought from a local marker and slaughtered. The animals (20 cattle and 20 buffaloes) were of both sexes and were between 2-4 years of age.

Immediately, after slaughtering, samples of fresh digesta (500 g) were taken from the rumen of each

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Table 1. Numbers of bacteria, protozoa and fungal zoospores in the rumen of cattle and buffaloes raised under traditional system in the Northeast of Thailand

Item	Cattle	Buffaloes
Rumen pH	6.58 ± 0.12	6.60 ± 0.07
Microbial population, total direct count		
Bacteria, $\times 10^8$ cells/ ml	1.36 ± 0.14	1.61 ± 0.12
Coccus, ×10 ⁵ cells/ml	1.07 ± 0.70	1.28 ± 0.23
Oval*	<	>
Rod*	<	>
Protozoa, ×10 ⁵ cells/ml	3.82 ± 0.88	2.15 ± 0.41
Holotrich	2.52 ± 0.70	1.80 ± 0.36
Entodiniomorph	1.30 ± 0.34^{a}	0.35 ± 0.13^{b}
Fungal zoospore, ×10 ⁶ cells/ml	3.78 ± 0.78^{a}	7.30 ± 0.93^{b}

a,b Values in the same row with different superscripts differ (p<0.05).

animal. Rumen digesta were squeezed through 4 layers of cheesecloth to ensure a sample which contained microbial population from both the liquid and solid phases. The subsequent rumen fluid was immediately fixed with 10% formalin solution (Galyean, 1989). The total direct count of bacteria, protozoa and fungal zoospores were made using the methods of Galyean (1989) based on the use of a haemacytometer (Boeco) and total numbers was studied for bacterial and protozoal shapes under microscope. Differentiation of rumen fungal zoospores from small protozoa was based on characteristics having flagellae while protozoa had ciliates around. Rumen fluid was diluted using autoclaved distill water (121°C for 15 minutes) as a medium, by 100, 10, and 10 times for bacteria, protozoa and fungal zoospores countings using 10×40, 10×10 and 10×40 ocular × objective of haemacytometer, respectively. For further identifying different shapes of bacteria rumen fluid was made at 10 times and counted at 10×40 ocular × objective of haemacytometer. Scanning electron microscopy was used for further determinations of bacteria, protozoa and fungal zoospores. Samples were fixed in buffered glutaraldehyde dehydrated in a graduated ethanol series and dried in a criteria-point dryer. They were mounted on aluminum stubs and sputter coated with gold prior to viewing. The data were subjected to statistical analysis using students t-test.

RESULTS AND DISCUSSION

The methods used proved successful at estimating the numbers of micro-organisms in the samples. Data on the numbers of bacteria, protozoa and fungal zoospores are shown in table 1. Based on this experiment, it was found that the rumen pH was similar for both species, but significant differences were found in the numbers of micro-organisms. There

was a trend for a higher population of bacteria, a lower population of protozoa and significantly more fungal zoospores in the ruminal fluid of buffaloes as compared with those of cattle.

The results obtained show microbial counts which are broadly in agreement with other studies reported by other workers for cattle and buffalo (Langar et al., 1968). Wattanachant et al. (1990) found higher total bacterial count and cellulolytic bacteria in swamp buffalo than those in cattle. Interestingly, in the present study where animals were subjected to similar feeding, there were strong trends for an higher bacteria and lower protozoa counts but significant fungal zoospore counts in buffaloes than those in cattle. As clearly known, rumen bacteria are far most important among protozoa and fungi, but moreover, their close associations and balances in forming optimal rumen ecolgy are utmost paramount in providing useful fermentation end-products for the ruminants.

The presence and role of fungi in the rumen have until recently been a contentious issue. In earlier years, Orpin (1975), Akin et al. (1983), Akin and Benner (1988) and Ho and Abdullah (1999) reported the findings of rumen fungi and their roles in degrading ligno-cellulosic materials which stimulated further studies to be conducted in these regards. Studies by Ho et al. (1988) indicated the presence of fungi in buffalo and in cattle and it was found that the rate and method of colonization by rumen fungi in swamp buffalo and native cattle of Malaysia were similar when fed on guinea grass. In addition, Ho and Barr (1995) offered detailed information regarding classification of rumen fungi from Malaysia. The present study clearly illustrates the presence of fungi in the rumen digesta of both cattle and buffaloes. It is interesting that, on similar diets, fungal zoospores in swamp buffalo were found twice as many as those found in cattle. Using the method of Ho and Barr

^{*} More or less as compared between cattle and swamp buffaloes.

(1995), in swamp buffaloes the fungi predominantly found were *Anaeromyces sp.* having sporangia with acuminate apex. Previous workers (Williams and Coleman, 1992) have suggested that there is a negative relationship between the number of protozoa and fungi in ruminal fluid that would be supported by the present work where protozoal numbers were lower in buffalo and fungal zoospores were higher and the opposite were found in cattle. As Williams and Coleman (1992) found that rumen protozoa predated on fungal zoospores therefore the lower numbers of protozoa present, the higher fungal zoospores would be. Rumen fungi would possibly act to control protozoal population as well.

Figures 1, 2, 3, 4, 5, 6 and 7 illustrate pictures of rumen bacteria attachment, protozoa distribution, fungal sporangia, rumen fungus, *Anaeromyces sp.*, having sporangium with acuminate apex and rhizoid forming appressorium in swamp buffaloes.

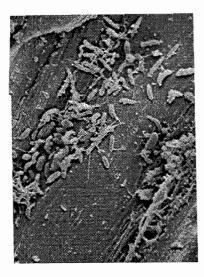


Figure 1. Attachment of rumen bacteria on rice straw of swamp buffalo, Bar=16 μ m

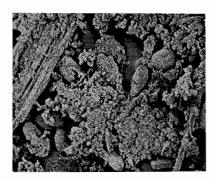


Figure 2. Rumen protozoa of cattle fed rice straw, Bar=16 μ m

Based on this study, interesting findings revealed differences in rumen baterial, protozoal population and fungal zoospore counts in swamp buffaloes and cattle raised under similar conditions. Since rumen microorganisms have impact on fermentation pattern and fermentation end-products particularly volatile fatty acids and NH₃-N, it therefore could be speculated that higher bacteria, lower protozoa and higher fungal zoospores in swamp buffaloes would result in more fermentation efficiency and productivity than in cattle. However, further studies in this regard as well as classification of rumen microbes of swamp buffaloes should be looked at more closely and warrant immediate research undertakings.

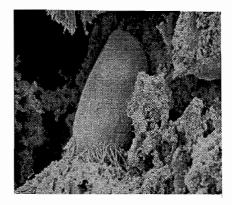


Figure 3. Rumen protozoa, *Entodiniomorph sp.* of swamp Buffalo, Bar=16 μ m



Figure 4. Sporangia of rumen fungi of swamp buffalo burst out from the surface of rice straw, Bar=16 μ m

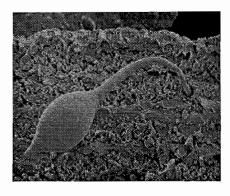


Figure 5. Rumen fungus of swamp buffalo, *Anaeromyces* sp. with acuminate apex, Bar=16 μ m

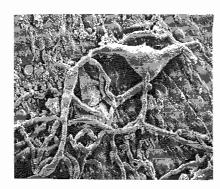


Figure 6. Rumen fungal rhizoid with penetrated appressorium of swamp buffaloes, Bar=16 μ m



Figure 7. Rumen fungal sporangium with flagellae, Bar=16 μ m

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REFERENCES

- Akin, D. E., G. L. Gordon and J. P. Hogan. 1983. Rumen bacterial and fungal degradation of *Digitaria pentzii* grown with or without sulfur. Appl. Environ. Microbiol. 46:738-748.
- Akin, D. E. and R. Benner. 1988. Degradatik on of polysaccharides and lignin by ruminal bacteria and fungi.

- Appl. Environ. Microbiol. 54:1117-1125.
- De Haan, H. Steinfeld and H. Blackburn. n.a. Livestock and the Environment: Finding a balance. European Commission Directorate-General for Development and Natural Resources, EU Publication. p. 115.
- Dehority, B. A. 1984. Evaluation of sub-sampling and fixation procedure used for rumen protozoa. Appl. Environ. Microbiol. 48:182-185.
- Devendra, C. 1985. Comparative nitrogen utilization in Malaysis swamp buffaloes and Kedah-Kelanton cattle. In Proc. The 3rd AAAP Animal science Congress, Seoul, Korea. Vol. 2:873-875.
- Galyean, M. 1989. Laboratory procedure in animal nutrition research. Department of Animal and Range Sciences. New Mexico State University, USA.
- Ho, Y. W. and N. Abdullah. 1999. The role of rumen fungi in fiber digestion. Asian-Aus. J. Anim. Sci. 12:104-112.
- Ho, Y. W. and D. J. S. Barr. 1995. Classification of anaerobic gut fungi from herbivores with emphasis on rumen fungi from Malaysia. Mycologia. 87:655-677.
- Ho, Y. W., N. Abdullah and S. Jalaludin. 1988. Colonization of guinea grass by anaerobic rumen fungi in swamp buffalo and cattle. Anim. Feed Sci. Technol. 22:161-171.
- Kennedy, P. M. and J. P. Hogan. 1994. Digestion and metabolism in buffaloes and cattle: are there consistent differences. In Proc. The 1st Asian Buffalo Association Congress (Ed. M. Wanapat and K. Sommart). Khon Kaen University, Khon Kaen, Thailand.
- Langar, P. N., G. S. Sidhu and I. S. Bhatia. 1968. A study of the micorbial population in the ruminal of buffalo (Bos bulalis) and Zebu (Bos indicus) on a feeding regimen deficient in corbohydrates. Indian J. Vet. Sci. 38:33-336.
- Orpin, C. G. 1975. Studies on the rumen flagellate Neocallimastix frontalis. J. Gen. Microbiol. 91:249-262.
- Wanapat, M. 1989. Comparative aspects of digestive physiology and nutrition in buffaloes and cattle. In Proc. Ruminant Physiology and Nutrition in Asia (Ed. C. Devendra and E. Imaizumi). Jap. Soc. Zootech. Sci. Sendai. pp. 27-43.
- Wanapat, M., K. Sommart, C. Wachirapakorn, S. Uriyapongson and C. Wattanachant. 1994. Recent advances in swamp buffalo nutrition and feeding. In Proc. The 1st Asian Buffalo Association ongress. (Ed. M. Wanapat and K. Sommart). Khon Kaen University, Khon Kaen, Thailand.
- Wattanachant, C., M. Wanapat, S. Sarangbin, S. Chanthai and C. Wachirapakorn. 1990. A Comparative study on rumen cellulolytic bacteria in swamp buffaloes and cattle. In: Proc. 28th Annual Conference, Kasetsart University, Bangkok.
- Williams, A. G. and G. S. Coleman. 1992. The Rumen Protozoa. Springer-Verlag, London. p. 441.