EFFECTS OF *ASPERGILLUS ORYZAE* EXTRACT (AMAFERM®) ON RUMINAL FIBROLYTIC BACTERIA AND IN VITRO FIBER DEGRADATION

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Summary

The effect of Amaferm on growth of pure cultures of ruminal cellulose-digesting, hemicellulose-digesting and pectin-digesting bacteria was determined. The addition of Amaferm to the growth medium increased the growth of *Ruminococcus albus* and *Fibrobacter succinogenes*. Amaferm had no effect on the growth of the other bacteria. Additionally, selective antimicrobial compounds were used to assess the influence of Amaferm on microbial contributions to in vitro fiber degradation. Amaferm appeared to stimulate fiber digestibility of only certain feedstuffs, and this increase in digestibility was attributed to its stimulation of bacterial activity. Amaferm did not appear to stimulate fungal activity.

(Key Words: Rumen, Fungal, Microbial Feed Additive, Growth, Fiber.)

Introduction

Reports on the use of fungal supplements in ruminant diets date back to 1924. However, results of those early studies were inconclusive. In recent years, there has been renewed interest in the use of microbial products as feed additives in ruminant diets, partly because of concerns about antibiotics. Microbial feed additives contain either the microorganisms, the dry products of microorganisms, the medium in which they grew, and/or the residues of their metabolism. The microorganisms used are yeast, molds, and/or bacteria. Because microbial products are not identical in composition, mode of action differs between products, and considerable variation in animal performance has been reported.

One of several fungal products commercially available is Amaferm®, a fermentation extract of the mold *Aspergillus oryzae*.The addition of Amaferm or products containing Amaferm have been reported to increase digestion of dry matter, fiber, and crude protein in vivo and in vitro. In our studies with newborn calves, Amaferm supplementation was shown to increase ruminal microbial activity, as evidenced by increased VFA concentration and bacterial numbers, particularly those of fiber-digesting bacteria. The increased microbial activity was associated with increased dry feed consumption in some calves and earlier weaning. Similar increases in intake have been reported in cattle and are probably the consequence of increased rate of fiber digestion in Amaferm-supplemented animals. It has been proposed that fungal supplementation may increase the nutritive value of feedstuffs by increasing the digestion of dietary fiber.

Little work has been done on the effect of Amaferm supplementation on the ruminal protozoa and fungi populations. The fungal population has been shown to have high fiber-digesting ability and may contribute to overall fiber digestibility. The protozoa population has been shown to prey on bacteria; therefore, inhibition of the protozoa population by Amaferm may partially account for increased bacterial numbers. Therefore, our objectives were to determine the effect of Amaferm on growth rate of selected pure cultures of ruminal fibrolytic bacteria and on the extent of degradation of forage components by the different microbial populations.

Procedures

Pure cultures of ruminal fiber-digesting bacteria (*Fibrobacter succinogenes, Butyrivibrio fibrisolvens, Eubacterium cellulosolvens, Ruminococcus flavefaciens, R. albus, Prevotella* (*Bacteroides*) *ruminicola, and Lachnospira multiparus*) were grown in anaerobic, complete carbohydrate rumen fluid medium with filter-sterilized Amaferm at 2 or 5% of the medium to determine its effect on their specific growth. The medium was inoculated with late-log-phase culture, and growth was monitored by measuring absorbance.

Selective antimicrobial compounds (penicillin and streptomycin to inhibit bacterial growth and cycloheximide to inhibit fungal growth) were used to assess the influence of Amaferm on bacterial and fungal contributions to in vitro fiber degradation. A variety of ground, fibrous substrates (alfalfa hay, brome hay, high and low endophyte fescue, pure cellulose, wheat straw, corn silage and prairie hay, 0.5 g) were incubated with ruminal fluid inoculum (1:2 ruminal fluid to buffer). Amaferm was added at .4, .8 or 1.2 g/l. NDF and ADF digestibilities were determined after 96 h incubation and compared to a control.

Results and Discussion

The addition of Amaferm to the medium increased (P<.1) the growth of *Ruminococcus albus* (Growth rate .71 vs .61/h) (Figure 1) and *Fibrobacter succinogenes* (Growth rate .35 vs .26/h). Amaferm had no effect on growth of other fibrolytic bacteria (Figure 2). Addition of Amaferm increased (P<.1) NDF and ADF digestion of brome and alfalfa hay. Amaferm addition at .4 or .8 g/l, but not 1.2 g/l, increased NDF and ADF digestion of high endophyte fescue (Table 1). The enhanced fiber degradation by Amaferm was attributed to its stimulation of bacterial activity. Amaferm did not appear to stimulate fungal activity, nor did Amaferm alone have any significant ability to digest fiber. Addition of Amaferm had no effect on NDF or ADF digestion of pure cellulose, low endophyte fescue, wheat straw, corn silage and prairie hay. In conclusion, Amaferm appears to stimulate NDF and ADF digestibility of only certain feedstuffs, and this increase in digestibility may be a consequence of growth stimulation of some fibrolytic bacteria.

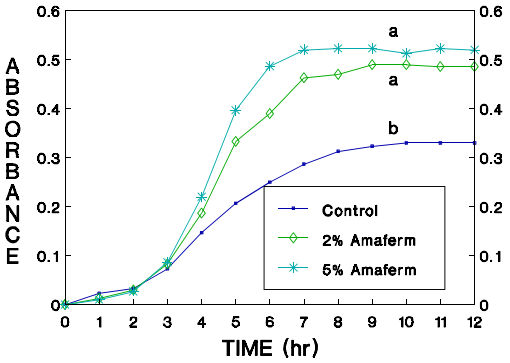

Figure 1. Effect of Amaferm on the Specific Growth Rate of *Ruminococcus albus*. Lines with Uncommon Superscript Letters Differ (P < .10).

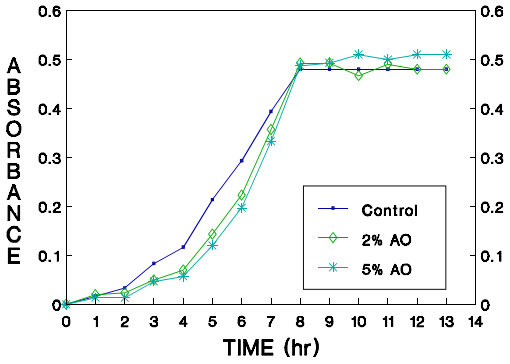

Figure 2. Effect of Amaferm on the Specific Growth Rate of *Eubacterium cellulosolvens*.

Table 1. Effect of Amaferm Supplementation on In Vitro NDF Digestion with Antimicrobial Compounds

	Feedstuff		
Item	Alfalfa hay	Brome hay	High endophyte fescue
% NDF in feedstuff	53.2	69.3	71.0
% NDF digested by:			
Bacteria + Fungi + protozoa (Whole rumen fluid or WRF)			
Amaferm	37.8[a]	55.4[a]	60.0[a]
AO = .4 g/L	42.2[b]	56.8[b]	64.3[b]
AO = .8 g/L	42.3[b]	60.3[b]	65.5[b]
AO = 1.2 g/L	43.0[b]	61.5[b]	59.2[a]
Bacteria (WRF + cycloheximide)			
No Amaferm	32.1[a]	50.8[a]	57.0[a]
AO = .4 g/L	37.8[b]	55.9[b]	62.4[b]
AO = .8 g/L	37.6[b]	56.1[b]	64.9[b]
AO = 1.2 g/L	39.2[b]	56.3[b]	55.2[a]
Fungi and protozoa (WRF + penicillin and streptomycin)			
No Amaferm	25.4	30.0	31.8
AO = .4 g/L	28.1	28.9	31.4
AO = .8 g/L	25.7	25.0	32.2
AO = 1.2 g/L	28.8	27.5	32.4
Negative control (WRF + P, S, C)			
No Amaferm	3.2	0	0
AO = .4 g/L	4.0	0	3.7
AO = .8 g/L	3.5	3.6	0
AO = 1.2 g/L	2.8	3.0	2.3
Amaferm alone (No WRF)			
No Amaferm	0	0	0
AO = .4 g/L	<1	<1	<1
AO = .8 g/L	<1	<1	<1
AO = 1.2 g/L	<1	<1	<1

[a,b]Means within a column with uncommon superscript letters differ (P<.1).