Effects of Cobalt Source on Rate and Extent of Dry Matter and Fiber Degradation in Vitro

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Summary

Positive effects on fiber degradation have been observed when supplemental cobalt was fed to ruminants. This study tested the effects of cobalt carbonate (CoCarb) and cobalt glucoheptonate (CoGH) at different concentrations on in vitro fermentation rate, fermentation end-product concentrations, and degradation of feed dry matter and neutral detergent fiber. Compared to CoCarb, CoGH increased dry matter disappearance and neutral detergent fiber degradation when added at 1 part per million (ppm) cobalt or less. Furthermore, CoGH had limited effects on the biohydrogenation of long chain fatty acids, whereas CoCarb appeared to stimulate this process when added at more than 3 ppm cobalt. Cobalt is not only an important precursor for vitamin B_{12} synthesis, but it also influences ruminal fermentation, with effects that depend on its chemical form.

Key words: minerals, digestibility, fiber, gas production

Introduction

Microbial populations in the rumen use cobalt to produce cobalamin (vitamin B_{12}), which is important to the animal as a cofactor in metabolic processes such as gluconeogenesis and methionine synthesis. Despite the ability of ruminal microbes to synthesize cobalamin from cobalt, the process is inefficient and makes ruminants susceptible to cobalt deficiency. Moreover, vitamin B_{12} is degraded in the rumen and its absorption is reduced as a result.

Increases in fiber degradation have been reported when extra cobalt has been added to rations for dairy cattle. On the other hand, inadequate dietary cobalt has been associated with unstable fermentation patterns and lower apparent nutrient digestibility due to shifts in microbial populations. The objective of this study was to determine the effects of varying CoCarb and CoGH concentrations on in vitro ruminal fermentation rate, dry matter disappearance, fiber degradation, fermentation end-product concentrations, and fatty acid biohydrogenation.

Experimental Procedures

For this study, three cannulated Holstein heifers were used as ruminal fluid donors, and they were fed once a day with a high-forage diet (Table 1) with no supplemental cobalt.

Experiment 1

The experimental treatments consisted of two different sources of cobalt: CoGH (COPRO® 25, Zinpro Corporation, Eden Prairie, MN; CoGH) and feed grade CoCarb. Each source was evaluated at eight different inclusion levels: 0, 0.1, 0.5, 1.0, 2.0, 5.0, 10.0, and 15.0 ppm cobalt. Glucose was also added to the CoCarb treatments such that the total mass added was uniform across sources of cobalt for each concentration. Each treatment combination was added to 4 different flasks (4 repetitions/treatment; 64 treatment flasks in total). Each flask contained 2.5 grams of a basal substrate composed of corn silage (22%), alfalfa hay (21%), corn grain (25%), cottonseed (4%), dried distillers grains (14%), and soybean meal (14%). This mixture was added to each fermentation flask, which was equipped with pressure sensitive membranes and RF transmitters that recorded the volume of fermentative gasses produced at 15-minute intervals.

Finally, 150 milliliters of the ruminal inoculum was added to each flask. The flasks were placed in an incubator at 39°C for 24 hours. The tubes were gently swirled every 3 to 4 hours during the incubation. After 24 hours of fermentation, gas production, pH of the solution, dry matter disappearance and neutral detergent fiber disappearance were measured.

Experiment 2

The extraction of rumen fluid and its preparation were identical to the procedure followed during Experiment 1. The treatments for this experiment were 0, 0.33, 1, 3, and 9 ppm cobalt from each of the two sources. After 24 hours of incubation, the pH of the solutions, concentrations of volatile fatty acids, ammonia concentration, and concentrations of long chain fatty acids were measured.

Results and Discussion

Experiment 1

In vitro dry matter degradation of basal substrate tested with different sources and concentrations of cobalt is presented in Figure 1. The dry matter degradation was similar between control and all other treatments; however, there was a trend for a linear decrease ($P < 0.10$) in dry matter degradation as cobalt levels increased. In addition, CoGH ($P < 0.05$) increased dry matter degradation compared to CoCarb (63.8 vs. 61.3% averaged across all levels); no interaction between concentration and source was observed.

Effects of treatments on neutral detergent fiber degradation are shown in Figure 2. Cobalt carbonate tended to increase neutral detergent fiber degradation ($P < 0.10$) compared to CoGH (50.1 vs. 47.7% across all cobalt inclusion levels). However, at cobalt concentrations of 0.1 to 1.0 ppm, CoGH increased fiber degradation by 21% (60.2 vs. 49.8%) compared to the same levels of CoCarb. On the other hand, CoGH

levels greater than 1 ppm led to a decrease in fiber degradation. In fact, when CoGH was supplemented at 15 ppm cobalt, only 15% of the neutral detergent fiber was degraded. Across all levels of cobalt evaluated, CoCarb had little impact on fiber degradation, with an average of 47.7% degraded compared to 45.2% degraded with no supplemental cobalt. These responses suggested that moderate levels of supplemental cobalt improved ruminal degradation of fiber.

The amount of supplemental cobalt did not affect pH; however, CoGH caused less change ($P < 0.05$) in pH relative to CoCarb.

Experiment 2

Supplementing cobalt decreased in vitro ammonia concentrations compared to control (25.5 vs. 24.7 millimoles/liter; $P < 0.05$). Ammonia concentrations were approximately 25 millimoles/liter with the addition of 0.33 to 3 ppm cobalt but decreased to approximately 23 millimoles/liter at 9 ppm (quadratic effect, $P < 0.05$). Individual and total concentrations of volatile fatty acids are shown in Table 2. Relative to the non-supplemented control, concentrations of branched chain VFA (valerate, isobutyrate, and isovalerate) decreased with cobalt supplementation ($P < 0.05$). Concentrations of acetate, isobutyrate, and isovalerate decreased in a quadratic manner as supplemental cobalt increased ($P < 0.05$). The negative response observed for isobutyrate and isovalerate concentrations was likely because the inhibitory effects we observed occurred only at 3 ppm cobalt and greater. These results, accompanied by the decrease in neutral detergent fiber degradation at high concentrations of CoGH, suggest that excessive amounts of cobalt can be detrimental for ruminal microbes, especially with highly soluble sources.

Long-chain fatty acids were grouped into five different categories for assessing impacts on their ruminal metabolism (Table 3). Cobalt carbonate linearly ($P < 0.05$) decreased the amount of polyunsaturated and total unsaturated fatty acids at the end of incubation, and also linearly increased saturated fatty acids. In the case of CoGH, the effect of concentration was minimal, resulting in substantial differences in fatty acid profiles between the cobalt sources at concentrations of 3 and 9 ppm. Considering profiles in the control treatment, levels of CoCarb greater than 1 ppm cobalt seemed to promote fatty acid biohydrogenation.

Published dietary requirements for dairy cattle suggest that 20 ppb (0.02 ppm) is the critical concentration of cobalt in ruminal fluid for adequate vitamin B_{12} production. The responses observed in the current study suggest that some additional benefits may be achieved at higher cobalt concentrations. In general, ruminal cobalt concentrations are expected to be approximately 80% of dietary cobalt concentrations; therefore, the corresponding estimated dietary cobalt concentrations evaluated in the current experiment ranged from 0 to 18.75 ppm.

Conclusions

Overall, our results suggest that cobalt does impact ruminal fermentation. Considering all of the outcomes we measured, the optimal response for ruminal microbes appears to be achieved with as little as 0.1 to 0.5 ppm cobalt. In that range of concentrations, CoGH and CoCarb enhanced dry matter degradation by 6 and 5 per-

centage units and neutral detergent fiber degradation by 18 and 7 percentage units, respectively, relative to no cobalt supplementation. Typical dietary cobalt concentrations should support ruminal cobalt concentrations in this range.

Table 1. Ingredient and nutrient composition of the diet fed to the ruminally-cannulated heifers during Experiment 1 and 2

Item	%, DM basis
Ingredient	
Wheat straw	36.91
Alfalfa hay	23.05
Corn grain, ground	21.73
Corn distillers grain, dried	17.93
Soybean meal	0.87
Trace mineral salt[1]	0.13
Vitamin A premix	0.0091
Vitamin D premix	0.0297
Vitamin E premix	0.0800
Nutrient	
DM, % as fed	60.6
CP	11.9
NDF	48.5
ADF	32.3
EE	2.5
NFC	31.8

[1] Provided 50 ppm Co, 300 ppm Cu, 2000 ppm Fe, 70 ppm I, 2000 ppm Mn, 3500 ppm Zn.

Table 2. In vitro volatile fatty acid concentrations (millimoles/liter) at different sources and concentrations of cobalt

Treatment	Acetate	Propionate	Butyrate	Valerate	Isobutyrate	Isovalerate	Total VFA
No supplemental Co	62.68	38.04	13.72	2.99	1.45	1.73	114.4
CoCarb (Co, ppm)							
0.33	61.27	38.02	13.35	2.87	1.43	1.66	112.6
1.0	62.04	37.85	13.43	2.91	1.46	1.69	113.3
3.0	62.96	38.95	13.61	2.90	1.40	1.65	115.5
9.0	62.07	38.40	13.54	2.86	1.30	1.53	114.0
CoGH (Co, ppm)							
0.33	61.22	37.42	13.43	2.87	1.41	1.66	112.1
1.0	64.10	39.18	14.01	2.93	1.44	1.68	115.8
3.0	61.63	37.31	13.64	2.89	1.40	1.62	112.5
9.0	60.10	37.46	13.39	2.86	1.34	1.51	110.8
SE	1.37	0.53	0.23	0.05	0.03	0.03	1.9
Contrast	D			A	ACD	ACD	

[1] Letters denote significance ($P < 0.05$) for the following statistical contrasts: A = No supplemental Co vs. all other treatments, C = linear dose, D = quadratic dose.

Table 3. Groups of long chain fatty acids produced when different sources and concentrations of cobalt were supplemented in vitro

Treatment[1]	Saturated	Unsaturated	PUFA	CLA
No supplemental Co	58.09	41.91	10.50	1.45
CoCarb (Co, ppm)				
0.33	53.36	46.64	11.89	1.74
1.0	56.50	43.50	11.28	1.57
3.0	63.58	36.42	9.72	1.25
9.0	64.61	35.39	8.83	1.55
CoGH (Co, ppm)				
0.33	54.65	45.35	12.17	1.62
1.0	57.09	42.91	10.46	1.41
3.0	54.07	45.93	12.27	1.66
9.0	57.95	42.05	11.61	1.65
SEM	2.32	2.32	0.87	0.13
Contrast[2]	BCE	BCE	BCE	CE

[1] PUFA = polyunsaturated, CLA = conjugated linoleic acid.
[2] Letters denote significance ($P < 0.05$) for the following statistical contrasts: B = source effect, C = linear dose, E = linear dose × source.

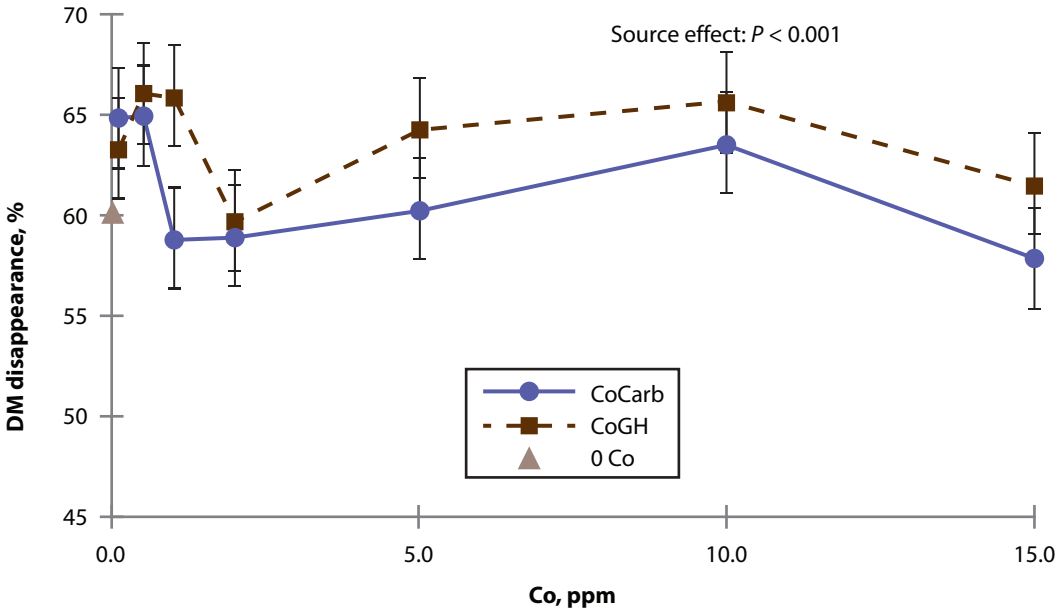

Figure 1. *In vitro* DM disappearance with different sources and concentrations of cobalt.

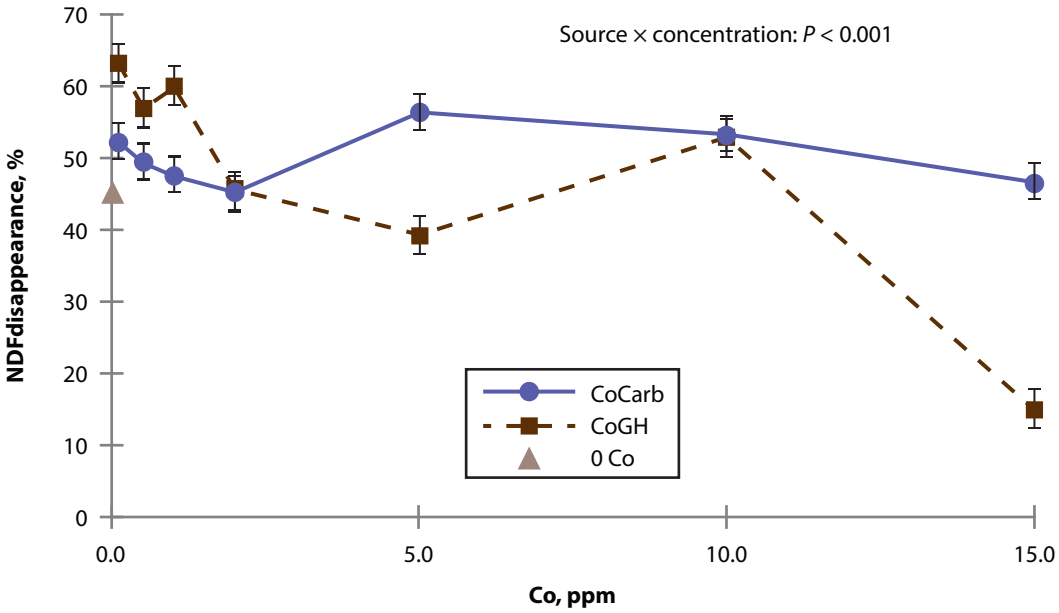

Figure 2. *In vitro* NDF disappearance with different sources and concentrations of cobalt.