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Agolin Ruminant®, an Essential Oil Blend, Increases Energy-Corrected Milk and Feed Efficiency in a High Component Dairy Herd

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Abstract

Agolin® Ruminant (Agolin) is a blend of essential oils developed to improve feed efficiency while at the same time reducing rumen enteric methane production. Studies have shown that the product improves lactational performance, but the range of results has been varied. This experiment evaluated the effects of the feed additive Agolinon milk production in a high-component (fat and protein) dairy herd when provided for an extended time. The experiment was conducted at a large commercial dairy in the Pacific Northwest, USA. Eight pens of cows (350 to 500 cows/pen) were blocked by production, and pens within blocks were randomly assigned to treatment. All pens received a common total mixed ration. Treatment consisted of providing 1 g/cow/day of Agolin to the test group. A 4-week adaptation period was followed by an 11-week study period. Milk production and milk composition were determined by cow by averaging daily performance for the one-week period before the start of the trial (covariate period) and the last week of each of the two test periods. Feed intakes were determined by pen at the same time. Milk yield was 1.11 kg/cow/day greater ($P < 0.001$) at the midpoint and 1.48 kg/cow/day greater ($P < 0.001$) at the end of the test period for cows receiving the feed additive. Similarly, protein yields were 0.03 ($P < 0.001$) and 0.07 kg/cow/day greater ($P < 0.001$) at the midpoint and end of the feeding period. Milk fat yield was not different ($P = 0.854$) between treatment groups at the midpoint of the trial but increased with treatment ($P = 0.002$) by the end of the trial. Energy-corrected milk/feed ratio, based on pen parameters, was significantly altered by treatment at the final test period (1.48 vs. 1.64 kg/kg for control and test treatments, respectively).

Keywords

Greenhouse Gas Mitigation, Methane, Feed Efficiency, Lactating Dairy Cows,

Agolin

1. Introduction

The ability to reduce enteric methane production by livestock provides a viable option for the reduction of ongoing greenhouse gas (GHG) emissions. On a global basis, dairy cattle produce an estimated 4% of the total GHG emissions of human origin [1]. While this number appears small, the ability to reduce methane, as opposed to other common GHG moieties, is of vital importance. Because methane is removed from the atmosphere at a relatively rapid rate, the reduction of this gas can act to reverse some of the overall warming effects of GHG [2]. Indeed, the ability to reduce enteric methane production in cattle can significantly contribute to atmospheric carbon reduction.

To that end, several strategies have been developed to reduce enteric methane output. However, for these to be voluntarily adopted by livestock producers, benefits beyond the knowledge that they are ultimately benefiting humankind are needed for the rapid uptake of such technology, and these need to be minimally cost-neutral to the livestock industry.

A central function of the feed additive Agolin Ruminant® (Agolin) is to reduce the production of enteric methane by ruminant animals. A detailed meta-analysis [3] showed that Agolin reduced net methane production per animal per day, production per unit of intake and production per unit of energy-corrected milk yield based on studies where the product was provided for more than 28 days.

As important, however, Belanche *et al.* [3] additionally found that the product, on average, increased energy-corrected milk by 4.1%. Similarly, Williams *et al.* [4] found that mid-lactation cows showed increased energy-corrected milk production and feed efficiency when the product was provided. This suggests that the energy gained through the reduced enteric methane output was available to support lactation. These benefits, if accurately quantified, can offset the economic burden of adopting the technology.

Energy-corrected milk production is an important economic index used by milk producers in many countries. It can be altered by elevating milk yield and maintaining the fat and protein components, or by elevating the components, or both. Interestingly, Brambila and Noricumbo-Saenz [5] saw improved energy-corrected milk when cows were supplemented with Agolin but noted that responses depended upon the start point in the lactation cycle. High-producing cows that first received the product at an average of 118 days in milk (DIM) responded by increasing milk yield, while cows receiving the same diet, but beginning later in their lactation cycle (224 days DIM on average when the product was added to the diet) produced more energy-corrected milk by increasing components rather than milk yield per se. Additionally, in the trial evaluation of Williams *et al.* [4], which began when cows averaged 146 DIM, an improvement in energy-corrected milk yield with the Agolin treatment was again brought about through increased component yield, rather than milk per se.

Trials with large numbers of animals are needed to uncover factors that alter the response to feed additives to determine the value of the product to the farming community. The primary purpose of this experiment was to determine if cows that remained on Agolin past peak lactation, rather than being introduced to the product for the first time at that point, maintained greater milk yield, or if there would be a shift in response manifested as improved component yield. This trial was conducted in a high component (fat and protein) herd, where the objective of the herd is to produce ECM by challenging the cows to produce fat and protein.

2. Materials and Methods

2.1. Animals and Treatments

This trial was conducted in the Pacific Northwest region of the United States. The herd selected was a high component herd made up of approximately 5500 cows, housed in pens of 350 - 500 cows. Eight pens of lactation cows were blocked by production and days in milk (DIM) and randomly assigned to one of two treatments by farm staff (**Table 1**). The treatment pens of cows received 1 g/cow/day of Agolin ruminant, provided through the mineral component of the ration via a service pack. The control cows received a mineral mix of the identical composition, with the test product omitted.

Fresh feed, in the form of a total mixed ration (TMR), was provided twice daily. Both groups received the identical TMR for the duration of the trial, with the only modification being the mineral service pack, as noted. The alfalfa-corn diets were formulated by an external consulting nutritionist using the feed formulation platform NDS (RUM & N Sas, Reggio Emilia, Italy) to meet nutrient requirements. Cows were given sufficient TMR to allow for 2% - 3% orts. The diets contained 300 mg/cow/day of sodium monensin. Dry matter consumption was recorded by pen daily.

Cows were milked twice daily. Each cow was fitted with an identification collar to permit the recording of milk weights (Metatron® Milk Meters, GEA Farm Technologies, Bonen, Germany). Milk composition was determined weekly.

Routine management practices were not altered. Cows were permitted to move from pen to pen according to the needs of the farm, but only cows that remained in the same pens for the duration of the trial were ultimately enrolled in the study.

2.2. Milk Analyses and Calculations

Weekly milk samples were collected for every cow immediately before the trial began, and for the duration of the trial and were submitted to Dairy Gold Milk Laboratory, Inc, Seattle, WA USA for component (fat and protein) percentage

Table 1. Pen assignments by treatments based on cows available at the start of the trial.

Control					Test				
Pen	DIM[a]	Milk, kg	Fat, %	Protein, %	Pen	DIM	Milk, kg	Fat, %	Protein, %
8	94	32.95	4.85	3.47	10	85	31.73	4.59	3.45
1	108	42.45	4.09	3.20	3	109	43.27	4.55	3.17
7	233	25.45	5.56	3.78	6	222	25.72	5.33	3.78
12	236	26.27	5.09	3.71	5	276	27.64	4.90	3.63

[a]Days in milk.

analyses. Milk yields for every cow were determined at the same time that milk was collected for the component analyses. Component yields were calculated by cow and date of analysis by multiplying milk yield by the component percentage values. Fat corrected milk (FCM) and energy-corrected milk (ECM) was likewise calculated by cow and date using the equations given by Erdman [6]:

Equation 1: FCM= 0.432 * milk yield + 16.23 * fat yield;

Equation 2: ECM = 0.327 * milk yield + 12.95 * fat yield + 7.65 * true protein yield.

3. Statistical Analyses

The trial began on December 17, 2021, at which time pretrial data regarding milk and component yield by cow were recorded. The Trial was 16 weeks long consisting of a 1-week preliminary covariate period, a 4-week dietary adjustment period, followed by the two test periods. Milk weights were averaged by cow based on production from December 11 through December 17 to represent the covariate period (Table 1). Measurements were again taken at the midpoint (February 13 through February 19, 2022) and at the end (March 26 through April 1, 2022) of the trial.

All production data were analyzed using cow as the experimental unit for animals that continually remained in their originally assigned treatment pens. There were 1253 and 1407 individual cows present from the start of the trial until February 19 representing the control and test periods respectively. Similarly, there were 1243 control cows and 1401 test cows that were continually available in their respective pens on April 1. Feed efficiency was analyzed using pens as the experimental unit, and days for replication.

Data were analyzed using Minitab 16 statistical software (Minitab Inc., State College. PA, USA). A general linear model was used to account for pretrial differences based on cows enrolled in the study on each test period. A randomized block ANOVA revealed pretrial differences in milk yield, fat percentage and protein percentage, and these factors were therefore applied in the model. Lactation number was used as a covariate.

Lactational persistency was determined for each test period, using the difference in actual milk production by cow between the pretrial period and trial period. Feed efficiency comparisons were evaluated using pens as the experimental unit with days used for replication based on findings for the last week of each period (pretrial, midpoint and end). Pretrial findings were used as covariables in the analyses.

Differences were deemed significant when the probability of a different result was less than 5% ($P < 0.05$). Tendencies were declared when the probability of a different result was between 5% and 10% ($P > 0.05$, $P < 0.10$).

4. Results and Discussion

The goals of this study were firstly to evaluate the feed additive in a high component herd and secondly to determine if there were differences in responses as lactation progressed. Three critical factors were compared: milk production, lactational persistency and feed efficiency.

4.1. Milk Production Parameters

The managerial goal of this herd was to maximize milk fat and milk protein yields and it was not known if the current yields could be maintained when the feed additive was applied. Cows averaged 129 DIM at the start of the trial, and advanced to 192 and 233 DIM at the two testing periods. Results in **Table 2** provide treatment differences after 64 days of exposure to the test product. While ECM was greater by 0.64 kg ($P < 0.05$) for the treatment group, this was primarily associated with increases in milk yield (1.11 kg) and protein yield (0.03 kg), but not fat yield ($P > 0.10$). There was a tendency ($P < 0.10$) for FCM to be elevated for the test group of cows.

In comparison, when cows continued to be exposed to the test product for 105 days (**Table 3**) ECM likewise increased for the treatment group relative to the control group ($P < 0.05$). This increase was associated with greater yields of milk, and protein as well as fat.

The reason for greater fat yield at the longer feeding time is not obvious, but might relate to stage of lactation. In a previous feeding experiment where two groups began receiving Agolin at different stages of lactation [5] milk yield was the main variable that drove ECM for the cows that received the treatment from 118 through 174 DIM. Fat yields were 1.59 kg and 1.61 kg for the control and Agolin treatments, respectively. When the feeding periodbegan at 225 DIM, and continuing through 281 DIM there were significantly greater fay yield with the Agolin (1.55 kg) as compared to the control (1.49) feeding group. This demonstrated that changes to component output were possible in the latter stages of lactation, but unlike the current study, did not provide information on the continuous feeding of the product for an extended period. It seems possible that continuous application might influence persistency, defined as the ability to maintain production post peak.

4.2. Persistency

The ability to maintain milk production past peak (persistency) is an important

Table 2. Least squares means for milk production parameters as determined the week ending February 19 (64 days on study; 1253 control cows and 1407 test cows available).

	Treatment			
Variable	Control	Test	SEM	P-value
Milk yield, kg	36.06	37.17	0.442	<0.001
Fat, %	4.65	4.50	0.006	<0.001
Protein, %	3.36	3.33	0.004	<0.001
Fat yield, Kg	1.64	1.65	0.018	0.854
Protein yield, Kg	1.19	1.22	0.013	0.011
FCM[a], kg	42.23	42.75	0.483	0.085
ECM[b], kg	42.13	42.76	0.478	0.034

[a]Fat corrected milk; [b]Energy-corrected milk.

Table 3. Least squares means for milk production parameters as determined the week ending April 1 (105 days on study; 1243 control cows and 1401 test cows available).

	Treatment			
Variable	Control	Test	SEM	P-value
Milk yield, kg	34.36	35.84	0.451	<0.001
Fat, %	4.36	4.33	0.005	0.002
Protein, %	3.37	3.39	0.003	<0.001
Fat yield, Kg	1.47	1.54	0.018	<0.001
Protein yield, Kg	1.13	1.20	0.014	<0.001
FCM[a], kg	38.70	40.40	0.488	<0.001
ECM[b], kg	38.91	40.81	0.491	<0.001

[a]Fat corrected milk; [b]Energy-corrected milk.

economic parameter in dairying. The costs required to feed and maintain the cow need to be less than the revenue received from the milk she produces, so a slower rate of decline in milk yield is highly desired to achieve this goal. Furthermore, Pedrosa *et al.* [7] noted that enhancing lactational persistency may improve cow health in the subsequent lactation. The length of the lactation cycle for an individual cow is generally not known until a cow is confirmed pregnant [8]. Allowing the usual 60 days for the dry period, and a gestation period of 283 days, the lactation should extend roughly 223 days beyond the date she was confirmed pregnant. If milk production cannot be adequately maintained for that duration, then financial losses result. Drying the cow early is a poor alternative, as the cow may over fatten, which leads toa greater risk for calving difficulties and periparturient diseases [9] [10].

Dekkers *et al.* [11] found that the value of greater persistency becomes more important with longer lactation periods. According to Do *et al.* [12], improving lactational persistency is an excellent strategy because the energy deficit and health issues that accompany increases in peak milk are avoided. Generally assumed to be managed through genetic selection [7] [13] [14], Kuehnl *et al.* [15] recently revealed feeding strategies can play a role in increasing persistency, underscoring the findings of the current study in which the feed was modified by the inclusion of Agolin.

Table 4 provides persistency results for the two treatment groups of cows as measured at the midpoint of the feeding trial. These results show that the rate of decline in milk and ECM was greater for the control relative to the test treatment ($P < 0.05$). There were no differences in this parameter for FCM ($P > 0.05$). Similarly, **Table 5** shows persistency results at the end of the feeding period. These results were significantly greater for the test Agolin treatment for milk, FCM and ECM. These results suggest improved greater economic returns for cows in the later stages of lactation.

As **Figure 1** shows, the rate of decline in milk over the course of the feeding trial was impacted less by time for the treatment group as compared to the control group. This might explain why cows receiving the treatment were able to demonstrate greater milk later in lactation in this study, but not in the trial conducted by Brambila and Noricumbo-Saenz [5]. In that study, cows first received treatment when DIM averaged 225, and while component yields increased, milk yield did not.

Agolin was developed in part to reduce methane output, diverting the energy from methane to milk or meat [16]. Therefore the two parameters are correlated. Moate *et al.* [17] determined there were fewer long-term studies investigating the ability of dietary interventions to reduce methane output, and with some intervention technologies. Responses do not persist into later lactation. However, the fact that milk yield declines as the trials progress would be a confounding factor. Knapp *et al.* [1] noted that cattle become less efficient as production declines, and this results in an increase in methane output/unit of product. By evaluating the change in ECM over time (**Figure 1**), it is possible to demonstrate long-term efficacy of dietary modifications. By measuring persistency in the current feeding trial, the results clearly show that the effects of the feed additive Agolin persist into the later stages of lactation. This is an important finding, and strongly suggests that the reduction in methane output likewise persists.

4.3. Feed Efficiency

Table 6 and **Table 7** provide pen related parameters for feed efficiency. Unlike milk parameters, the pen data reflect a mixture of animals present for varying lengths of time. There were no differences in dry matter intakes by pen at either measurement time. The results indicate that there were no differences in efficiency when measured at the trial midpoint ($P > 0.05$) but differences occurred by the end of the trial ($P < 0.05$).

Lovendalh *et al.* [18] reviewed the relationship between greater feed efficiency and reduced methane output in lactating dairy cows. According to the [18], both are correlated because they are influenced by the rumen microbiome. Essential

Table 4. Milk persistency results as determined the week ending February 19 (64 days on study; 1253 control cows and 1407 test cows available).

	Treatment			
Variable	Control (C)	Test (T)	T minus C	P-value
Milk yield change, kg	−0.10	1.02	1.12	<0.001
FCM[a] change, kg	−1.77	−1.33	0.44	0.141
ECM[b] change, kg	−2.03	−1.40	0.63	0.033

[a]Fat corrected milk; [b]Energy-corrected milk.

Table 5. Least squares means for milk production parameters as determined the week ending April 1 (105 days on study; 1243 control cows and 1401 test cows available).

	Treatment			
Variable	Control (C)	Test (T)	T minus C	P-value
Milk yield change, kg	−1.52	−0.04	1.48	<0.001
FCM[a] change, kg	−4.97	−3.33	1.64	<0.001
ECM[b] change, kg	−4.94	−3.03	1.90	<0.001

[a]Fat corrected milk; [b]Energy-corrected milk.

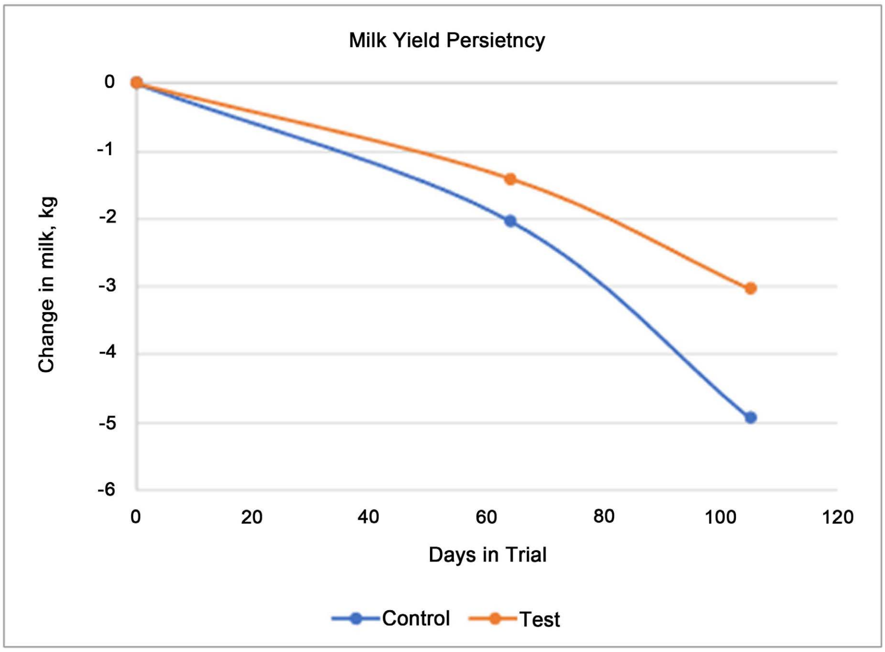

Figure 1. Effects of Agolin treatment on lactational persistency.

oils can reduce the methanogenic archaea in the rumen, thereby altering the overall microbiome and sparing energy [19]. Thus, the gain in feed efficiency found with Agolin may be related to the effects of the essential oils [3] [16] on the rumen the methane producing microbes in the rumen.

Table 6. Least squares means for pen average values for the week ending February 19 (64 days after the start of the study).

	Treatment		
Variable	Control (C)	Test (T)	P-value
Dry matter intake, kg	24.68	24.45	0.799
Milk yield, kg	34.45	34.95	0.495
Milk/intake, kg/kg	1.41	1.44	0.629
ECM[a] yield, kg	40.24	40.28	0.824
ECM[a]/intake, kg/kg	1.64	1.66	0.739

[a]Energy-corrected milk.

Table 7. Least squares means for pen average values for the week ending April 1 (105 days after the start of the study).

	Treatment		
Variable	Control (C)	Test (T)	P-value
Dry matter intake, kg	25.64	25.05	0.295
Milk yield, kg	32.81	34.06	0.151
Milk/intake, kg/kg	1.28	1.36	0.020
ECM[a] yield, kg	37.86	41.04	0.008
ECM[a]/intake, kg/kg	1.48	1.64	0.003

[a]Energy-corrected milk.

5. Conclusions

This trial was conducted on a large dairy with over 1000 cows available for each treatment. With this number of subjects, confidence in the results should be high. The trial compared cows given the same diet for both treatments, except for the inclusion of Agolin at the rate of 1 g/cow/day in the test diet. Unlike many other trials, the feeding period was long: measured over a 105-day period.

The results showed that by the end of the feeding period, milk FCM and ECM yields favored the test treatment by 1.48, 1.64 and 1.90 kg, respectively. Fat and protein yields were both 0.07 kg/cow/day greater with the Agolin treatment by the end of the trial. Thus, Agolin can be of benefit in herds striving to maintain or increase component yields.

An important observation emanating from this trial is the fact the response to Agolin by dairy cows was not diminished as the trial progressed, indicating that the product remained efficacious. The evaluation of persistency, as described here, might be a tool that can be used to determine the long-term benefits of feed additives in dairy cows.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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