

Small-Scale Dairy Production Study Design

Objective

The objective of the experiment proposed herein is to evaluate the role of “BiOWiSH®” in animal health, growth performance, and milk production of dairy cattle.

Treatments

1. Control - no BiOWiSH®
2. Control + BiOWiSH®

BiOWiSH® Application Options

1. Molasses
2. Water
3. Pelleted, Total Mixed Ration, Mash

Please refer to the relevant BiOWiSH® User Guide (Ruminant Operation Applications) for details on dosage and formulation. In general, BiOWiSH® is incorporated into the feed ration using one of the options below:

- BiOWiSH® MultiBio 3PS is a direct fed microbial (DFM) for all animal species and growth stages. BiOWiSH® MultiBio 3PS can be added to animal mash feeds, animal liquid feed, total mixed or bunk rations, compound feeds, and animal drinking water. Please see our BiOWiSH® Ruminant Operation Applications User Guide for detailed usage directions
- BiOWiSH® MultiBio 3P is a direct fed microbial (DFM) for all animal species and growth stages. Add BiOWiSH® MultiBio 3P to animal feed through the pelleting or extrusion process as a micro addition. Please see our BiOWiSH® MultiBio 3P Compound Pellet Feed Applications User Guide for detailed usage directions.

Animals

Clearly specify animal origin and health status- source animals from the same or similar producer

Body condition score

Arrival weight

Multiparous cows vs heifers

Disease status

Vaccine status

Option 1- Experimental Design

Minimum of 30 animals per pen and 8 replicate pens per treatment

Randomized Complete Block Design

Block by arrival weight (preferred method with many replicates), Randomly allocate animals to pens within each weight block; Randomly allocate pens to treatment or control diets.

Option 2- Small Pen Trial

Completely Randomized Design only- Random treatment/pen allocation (for few replicates)

Statistical analysis at discretion of research partner.

Procedure

Depending on actual measured growth rates and milk production, this schedule may be adjusted to closely mimic commercial dairy cow management.

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Treatment Application and Feed Delivery

BiOWiSH[®] MultiBio 3PS will be provided to the research partner in sufficient quantity to deliver to all treated animals for the duration of the experiment.

Cattle shall remain on the same experimental ration throughout the duration of the experiment (e.g. gestation, lactation and dry off for dairy cattle). Basal ration will be the current “commercial” ration minus any direct-fed microbials and antibiotic growth promoters. Bunk Rations must be read at least twice daily by the bunk reader and managed to provide ad libitum access to feed and minimize refusals. Bunk reader and feed delivery truck drivers are to be blinded of treatment assignment to pen. Feed is to be delivered twice daily. Records of daily feed delivery should be used to calculate average daily intake (Dry Matter basis) by pen. Cattle should be moved through ration “step-ups” in the same manner as all other commercially-fed cattle using standard procedures (e.g. 7-day, 50:50 ration splits for acclimation).

Feed Sampling and Analyses

Feed should be sampled from the bunk of each pen weekly throughout the experiment and monthly composites (triplicate bunk grabs immediately after delivery, prior to cattle contamination; stored frozen until month-end; 1 composite for each ration) may be analyzed for nutrient composition (i.e. Moisture, crude protein, crude fat, ADF, Ash, TDN, Energy (Digestible and Metabolizable), Net Energy, Fe, Ca, Mn, P, Cu, Mg, S, Na, K, Zn) (i.e. BiOWiSH Approved Laboratory). Average Daily Gain (ADG), Dry Matter Intake (DMI), Feed Conversion Ratio (FCR), or Efficiency of Gain (G:F), Cost of Gain (COG) are to be calculated as indices of animal growth performance as well as milk yield (lb/kg), somatic cell count (SCC), % butterfat, lactation duration and number of lactations.

List experimental diets for each study phase		
Basal Ration	Phase	Phase
Ingredient	Approximate Inclusion Rate	Approximate Inclusion Rate

BiOWiSH[®] MultiBio 3P/3PS

Morbidity and Mortality

Research personnel, staff, and technicians should be blinded of treatment assignment to pen. Cattle should be evaluated at least twice daily for clinical symptoms of respiratory disease (nasal or ocular discharge, lethargy, anorexia or depression), digestive upsets or any other ailment. Sick animals removed from the pen should be treated following best management practices and returned to their respective home pen. Animals treated more than three times for respiratory disease and should not be removed from the experiment. If a single animal has been treated three times for respiratory disease, recovers, and relapses after 30 days it will be treated as an initial episode and treated as previously described. Morbidity will be calculated as the percentage of cows in a pen that require 1) one treatment for respiratory disease, 2) two or more treatments for respiratory disease, and 3) Chronic (does not recover after three respiratory treatments). Although instances of morbidity may be minimal, morbidity data should be separated into respiratory, digestive, and “other” categories, as applicable and presented as the percentage of the original pen population that ever required treatment. Veterinary costs should be presented and included in the COG analysis. Mortality data should also be recorded and presented as the percentage of the original pen population that succumbed to death for any reason.



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Fecal Sample Collection and Analysis

Fecal samples should be collected each week or every other week from each pen for the duration of the experiment. The total number of composited samples collected per pen will be determined by the number of DOF required for cattle to reach their market endpoint or their lactation period. To ensure sample integrity, fecal samples must be collected according to the procedures detailed in Appendix A. Analysis of fecal samples should include moisture, crude protein, crude fat, ADF, Ash, TDN, Energy (Digestible and Metabolizable), Net Energy, Fe, Ca, Mn, P, Cu, Mg, S, Na, K, Zn.

Approximate Timeline - Dairy Cattle

Event	Approximate Timing
Begin receiving cattle for trial	After protocol finalization
Begin experiment	Upon diet acclimation or parturition
Milk yield, Milk Composition etc.	Daily Data Collection
Corresponding Feed and Fecal Sampling	Weekly, Bi-Weekly, or monthly
Completion of Experiment	Dry off period, Cull, or Harvest
Delivery of final Report	Approximately 30 d after cattle ship for harvest

Key Metrics to Measure

- Initial body weight
- Ending body weight
- Body condition score
- Average daily gain
- Daily feed offered
- Daily feed refused
- Daily feed intake
- Feed conversion ratio
- Calf weight
- Milk yield (day or lactation period)
- Milk protein
- Milk somatic cell count
- Milk butterfat
- Days in milk
- Days on feed

Optional Parameters to Measure

- Digestibility/nutrient utilization
- Microbiology/pathogen prevalence
- Daily fecal output
- Water intake
- Feed quality (lab analysis)
- Daily high temperature
- Daily low temperature
- Rectal Temp during heat stress
- Respiration rate
- Steps per day

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Appendix A

Protocol for Fecal Sampling - Pen Floor Samples

Items Needed

- Disposable Gloves
- 1 gal freezer bags
- Permanent Marker
- Tongue Depressors
- Cooler with ice packs or ice

Sample Times

Fecal samples should be photographed, scored, and taken every 14 days, starting at Day 1 through Day 56 for a total of 5 samples per pen. Samples should be taken first thing in the morning prior to ration delivery.

Procedure

1. Label bags with day of collection, treatment ID and date.
2. Establish a 5-foot perimeter along the fence line. No samples should be taken from within 5 feet of the fence line to avoid cross-contamination between pens.
3. Photograph cow pats prior to sampling as a visual observation of differences between treatments.
4. Using scoring system in Table 1, score the fecal cow pat prior to taking a sample. Record your findings for every fecal sample taken.
5. A minimum of 5 fecal grabs (~20-30 grams ea.) from different cow pats should be taken per pen. These will make up the composite pen sample (~100-150 grams total).
6. Sample only from cow pats that appear fresh and firmly mounded (no crust over). Do not sample watery, loose stools or stools that have a marble appearance.
7. Gloves on, place the tongue depressor in the center and stir the cows pat 3-5 times without scraping the ground underneath.
8. The tongue depressor can be used to collect the sample if cow pat is firm enough, or with a gloved hand, grab a fist full of the stool, trying not to scrape the ground with your fingers.
9. Place the sample in the bag and move on to the next stool within the pen until samples from 5 different cow pats have been taken.
10. Seal the bag, then gently massage the bag several times to mix the sample within.
11. Put sample in cooler on ice and move to next pen.
12. After collecting from all pens, samples will be kept on ice for transport for analysis.
13. Should there not be enough cow pats of the correct consistency for sampling in a given pen, samples of marbled cow pats should be taken. Do not sample watery, loose stools.

MANUAL FECAL SCORING TABLE FOR CATTLE

1. Cream soup consistency; contains mostly water with little or no dietary material
2. No stacking; pat less than 1 inch thick; lacks consistent form; consistency of cake batter
3. Ideal with normal pat form; consistency of thick pancake batter; slight divot in middle; deeper than score 2 pat, but will not stack
4. Thick and becoming somewhat deeper, yet not stacking; consistency of peanut butter
5. Firm; stacks over 2 inches in height; clearly defined segments; very dry