# Impact of methionine hydroxy analog supplemented to cows during the last trimester of gestation to evaluate cow reproduction and calf growth

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# Abstract

The study was conducted to investigate the impact of methionine hydroxy analog supplemented to cows during the last trimester of gestation to evaluate cow reproduction and calf growth for 3 years. The five treatments are allocated to 10 pastures where pasture is an experimental unit and there is one replication for each treatment. There were 11 groups of cows that stayed in the same groups and treatments but were assigned to different pastures each year. Each year, there were total of 150 cows in the study and the five treatments are No supplement **(NS)**, 1lb of a distillers-based cube **(T1)**, 2lb of distillers-based cube **(T2)**, 2lb of a distillers-based cube plus 30g of methionine hydroxy analog **(MET)** and lastly hay with no protein **(HAY)**. The result from the studies shows that treatment was significant for precalf breed weight (T2 = 161.30, p-value = 0.0144) and calf wean weight (HAY = 511.54, p-value = 0.0297). However, there were not any significant difference between MET and other treatments. In addition, the treatment was significant for change in precalf BCS (HAY 0.06379, p-value < .0001) and pregnancy (p-value = 0.0424). The treatments were not significant for any carcass traits. There was significant effect of year for all carcass traits except marbling and quality grades. Furthermore, there was significant sex effect for calf birth (HAY = 76.91, p-value <.0001) and calf wean weight (HAY = 511.54, p-value <.0001). Overall, methionine hydroxy analog does not seem to have significant effect on the variable of interest i.e. weight, BCS and carcass traits.

# Introduction

“Maternal methionine (Met) supply in non-ruminants during pregnancy can affect offspring development and growth” (Alharthi et al., 2018, p. 1). The research concludes that “increasing the maternal supply of Met during late-pregnancy enhanced growth in utero as well as during the pre-weaning and early post-weaning periods” (Alharthi et al., 2018, p.2). Collectively, these studies influence the objective of our study furthermore which is to evaluate the calf growth and cow reproduction when cows are supplemented during the last trimester of gestation with methionine hydroxy analog (MFP). Additionally, “nutritional status during the third trimester of gestation of beef cows regulates placental environment, fetal development (Funston et al., 2022, as cited in Izquierdo et al., 2022) and offspring postnatal performance” (Palmer et al., 2022, 2022a, b as cited in Izquierdo et al., 2022). Hence, considering this last gestation period with protein supplementation is of interest. Methionine hydroxy analog (MFP) has similar chemical structure to that of methionine. More so, the studies indicate that “methionine plays an important role in gestating beef cows impacting epigenetic of offspring” (Silva et al., 2021, p.2). Thus, our interest lies in the impact of methionine hydroxy analog with other treatments that have varied level of protein to evaluate cow reproduction and calf growth.

In this study, there are five treatments of interest which are no supplement (NS), 1lb of a distillers-based cube (T1), 2lb of distillers-based cube (T2), 2lb of a distillers-based cube plus 30g of MFP (MET). (Distillers based cube are good source of energy and protein which contains higher percent energy value of corn grain) and lastly hay with no protein (HAY).

# Materials and Methods

# Experimental design

The experiment is a three-year study. Cows are supplemented on winter range (December through February) with one of the five treatments. There are ten pastures where the pasture is an experimental unit. Five treatments are randomly applied to pastures and each treatment has one replication. There are eleven cows for each treatment allocated to a pasture. These 11 cows stayed in the same groups and treatments but assigned to different pastures each year. Note: Hay 1 and Hay 2 pastures receive Hay treatment in all years. There are a total of 150 cows in each year. The primary response of interests for the cows and calves are weight, Body conditioning scores, and carcass traits.

# Statistical Model

Let denote one of the 14 responses for year, treatment, (NS, MET, T1, T2, HAY), sex (Bull or Heifer) Additionally, a baseline measurement day season was taken into account for some variables. This is represented by B ( - .

Then, model is

Where:

is the overall mean.

is the effect of the year, i = 1, 2, 3

is the effect of the treatment, j = 1, 2, 3, 4, 5

is the random term where treatment is nested within year.

B is the coefficient indicating dependency of on and B

is the measurement made on the day season covariate corresponding to , where day season is equal to the calf birthdate minus the first calving season plus one.

is the average of the day season values (

is the error term, ~ iid N(0, ) where has a covariance structure.

For non-normal data the pregnancy response, a binomial model was used with the logit link function. The final model had fixed effect of treatment and dayseason as covariate with random year\*treatment effect. The cows who are not pregnant were dropped along with any repeated measurements.

# Results

# Statistics Overview

The PROC GLIMMIX procedure in SAS 9.4 was used to analyze following variables of interest. Residuals and QQ-plots were used to assess normality.

# Statistical Significance

In statistics, p-value is used to determine whether a variable is associated with affecting the difference in the response. The p-value is dependent upon what are called t-values or F-values. If the p-value is less than 0.05, there is a strong significant effect of that variable or there is a significant difference between the responses due to the two variables being compared.

# Weight

The following weight responses were measured in the study. These are birth weight of calf, pre breed calf weight, calf wean weight and cow wean weight. The compound symmetry structure was chosen for these responses by the smallest AICC and BIC.

The type III test of fixed effect (ANOVA) was used to determine significance regarding the birth weight of calf. The sex and year were significant with a p-value < 0.001 and p-value = 0.0276, which we expect to see since birth weight might differ based on the sex. Regarding the pre breed calf weight, all fixed effects year, treatment, and sex were significant with following p-values of 0.0006, 0.0144, <0.0001. Regarding cow wean weight, year is only significant with p-value of 0.0049. Treatments seem to have significant effect on pre breed calf weight, and sex seems to have effect on the birth weight along with significant year effect.

The table below indicates estimated mean of weight variables across different treatments. T2 treatment leads to larger weight for pre breed calf and cow wean weight whereas hay treatment leads to larger weight in calf at birth and when calf is weaned.

**Table 1.** The effect of treatment on the weight responses, point estimates.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Weight  Variables | Treatments | | | | |
| NS | T1 | T2 | MET | HAY |
| Birthweight of calf | 71.20 | 76.43 | 76.47 | 74.25 | 76.95 |
| Prebreed calf weight | 137.97 | 156.70 | 161.30 | 151.64 | 160.62 |
| Cow wean weight | 1087.58 | 1055.84 | 1116.39 | 1039.90 | 1115.54 |
| Calf wean weight | 464.70 | 493.30 | 507.98 | 486.56 | 511.54 |

**Plot 1.** The effect of treatment on the respone calf mean weight



# Body Conditioning score (BCS)

The variables of interest for body conditioning score are Pre calf BCS, Pre breed BCS and BCS. The body conditioning score describe the relative fatness or body composition of cow. The scoring system has a range of 1 to 9, with 1 representing very thin cows, 9 representing very fat cows and 5 representing average cows. Dec BCS is the body conditioning score of cows before the treatments were fed. A new response was made for this analysis (pre calf BCS – Dec BCS). The year seems to be significant for the new response, whereas treatment was not significant.

The table below indicates estimates with positive and negative change across different treatments. The T2 has larger change for pre calf BCS score and T1 has larger change for pre breed BCS whereas change is negative across all treatment for BCS i.e body conditioning score was lower overall compared to cows before treatments were fed.

**Table 3.** The effect of treatment on the BCS, point estimates.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| BCS  Variables | Treatments | | | | |
| NS | T1 | T2 | MET | HAY |
| Precalf -decBCS | -0.7564 | -0.2002 | 0.07459 | 0.03410 | 0.06379 |
| Prebreed -decBCS | -0.6698 | 0.4082 | 0.3005 | -0.2822 | -0.05546 |
| BCS - decbcs | -0.1832 | -0.3085 | -0.2695 | -0.2089 | -0.3111 |

**Plot 2.** The effect of treatment on the respone (precalf BCS – decBCS)



# Carcass traits

Carcass traits refers to the physical characteristics of the animal's body after it has been slaughtered and processed for meat. The following carcass traits were measured hot carcass weight (HCWT), Quality grade (Q grade), Yield grade (YG), Ribeye (Rea), Marbling (Marb) and Backfat (Bkf). Sex and day season were not considered for this model as they do not play any role in the responses mentioned above. Year was the only significant variable. The table below indicates point estimates across different treatments for carcass traits. The hay has higher HCWT followed by T2, where no supplement has lowest HCWT.

In our dataset, quality grade has three categories prime, select and choice. The binomial distribution was used since the variable was categorized into two options choice vs others. This was based off the frequency table where choice was 156 and prime and select combined was 28. The NS has the highest yield grade score whereas lowest is T1. The higher value of ribeye, the better quality. It seems that T2 has highest value followed by MET AND HAY. The hay has a higher marbling score followed by T2 and T1 whereas the lowest is NS. The lower backfat thickness score typically has a higher yield grade. The NS treatment has lower backfat followed by MET.

**Table 5.** The effect of treatment on carcass traits, point estimates.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Weight  Variables | Treatments | | | | |
| NS | T1 | T2 | MET | HAY |
| HCWT | 782.07 | 802.60 | 820.56 | 804.36 | 820.99 |
| Probability Q grade | 0.9687 | 0.9459 | 0.9421 | 0.9444 | 0.8154 |
| Yield grade | 2.4709 | 2.7461 | 2.7208 | 2.5614 | 2.6910 |
| Rea | 14.0378 | 13.9569 | 14.5065 | 14.3341 | 14.3267 |
| Marb | 509.27 | 536.81 | 539.50 | 522.14 | 585.52 |
| Bkf | 0.4519 | 0.5175 | 0.5526 | 0.4903 | 0.5131 |

# Pregnancy

The pregnancy variable was recorded as binary where 1 means cow is pregnant, and 0 means cow is open or not pregnant. There is significant effect of treatment on pregnancy rate. The probability of success for pregnancy is higher for all three treatments except for NS. The probability of pregnancy is 17 times as large as the probability of not being pregnant for T1 followed closely by Met and Hay.

**Table 6.** The effect of treatment on pregnancy, est. probability, odds ratio

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Treatments | | | | |
| NS | T1 | T2 | MET | HAY |
| Pregnancy probability | 0.8048 | 0.9449 | 0.9392 | 0.9440 | 0.9415 |
| Pregnancy odds | 4.1216 | 17.1572 | 15.4582 | 16.8699 | 16.0818 |

# Conclusion

The findings of the present study indicates that treatment was significant for pre breed calf weight (p-value 0.0144). The treatment differences were significant for T1, T2 and NS in comparison with Hay, where Hay treatment has the highest estimates (160.62) and lowest estimates for NS (137.97). Likewise, there was significant effect of treatment for calf wean weight (p-value 0.0297). The only significant differences occur between Hay and NS and NS and T2. The year seems to be significant across all the weight variables. However, we see some significant effect of sex for calf birth weight (p-value <0.0001) as well as significant effect of sex (p-value <0.0001) for pre breed calf weight. This significance indicates that calf sex has an impact on their birthing weight and pre breed calf weight, since bulls weigh more than heifers. The treatment has significant effect for change in body conditioning score from pre calf BCS to Dec BCS (p-value < 0.0001). The treatment differences that were significant are all the treatments in comparison with NS. We see the biggest change in estimate for body conditioning score was for T2 treatment (0.07459) followed by Hay (0.06379). The findings for the carcass traits showed that there were not any significant treatment effects. Overall, it turns out that MET treatment does not have any significant impact on our variables of interest. Although, we saw that treatment was significant for pre calf breed and calf wean weight, but MET was not significantly different in comparison to other treatments, though Hay and T2 treatments seems to have higher estimates in general than MET for the weight.

# Discussion

The findings of the study were not very interesting, since MET was not significant for any variables. However, the experiment can be improved, and variables can be explored further. Future studies could look at MET with different amount of distillers-based cubes and the amount of MFP. Additionally, different supplementation could be added with MET. Also, the treatment could be fed during different lactation stages to explore calf performance. Instead of assigning treatment to the pasture, the treatments could be fed to cows individually. There are 11 cows receiving same treatment within a pasture so number of cows could be increased and for that conducting power analyses would be beneficial to determine number of cows to detect any differences in treatment.

# Literature Review

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Palmer, E.A. *et al.* (2020) ‘Maternal supplement type and methionine hydroxy analogue fortification effects on performance of Bos indicus-influenced beef cows and their offspring’, *Livestock Science*, 240, p. 104176. doi:10.1016/j.livsci.2020.104176.

Silva, G.M. *et al.* (2021) ‘Effect of rumen-protected methionine supplementation to beef cows during the periconception period on performance of cows, calves, and subsequent offspring’, *Animal*, 15(1), p. 100055. doi:10.1016/j.animal.2020.100055.

Summers, A.F., Blair, A.D. and Funston, R.N. (2015) ‘Impact of supplemental protein source offered to primiparous heifers during gestation on II. progeny performance and carcass characteristics’, *Journal of Animal Science*, 93(4), pp. 1871–1880. doi:10.2527/jas.2014-8297.

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Stroup, W.W. *et al.* (2018) *SAS for mixed models: Introduction and Basic Applications*. Cary, NC: SAS.

# SAS Appendix

PROC IMPORT OUT= WORK.MEthionine

DATAFILE= "C:\Users\nregmi3\OneDrive - University of Nebraska-Lincoln\3 - 825 and 930\0 - 930-Spring2023\Consultant Files\Neetu\Landon Tadich\GSL Methionine Full Sheet .xlsx"

DBMS=EXCEL REPLACE;

RANGE="Sheet1$";

GETNAMES=YES;

MIXED=NO;

SCANTEXT=YES;

USEDATE=YES;

SCANTIME=YES;

RUN;

proc print data = methionine; run;

data methionine;

set methionine;

if past\_16\_17='Hay2' then past\_16\_17='Hay 2';

run;

proc print ; run;

proc sort data=methionine;

by Cow\_ID year;

run;

proc print; run;

proc freq data=methionine;

table year\*Past\_16\_17;

run;

proc sort data=methionine;

by year;

proc print ;run;

\*determine first day of calving season each year in order to have covariate of dayseason;

data meth2;

set methionine;

if birth\_date=. then delete;

proc print data = meth2; run;

proc sort data=meth2;

by year birth\_date;

proc print data = meth2;run;

data first;

keep year first\_date;

format first\_date date9.;

set meth2;

by year birth\_date;

first\_date=birth\_date;

if first.year then output;

run;

\*this is the cow data;

data methcow;

merge methionine first;

by year;

dayseason=birth\_date-first\_date+1;

proc print data = methcow; run;

\*this is the calf data;

data meth3;

merge methionine first;

by year;

dayseason=birth\_date-first\_date+1;

if sex in ('H','B');

proc print data = meth3; run;

run;

title1 'Initial Analysis of Calf Birth Weight with fixed effects';

title2 'year, treatment, calf gender, and linear day in calving season';

title3 ' and random year\*treatment and repeated measures to account for';

title4 'cows having multiple records';

run;

proc glimmix data=meth3 plot=residualpanel;

class year cow\_id past\_16\_17 trt sex;

model birth\_wt= year trt sex dayseason;

random intercept/subject=year\*trt;

random \_residual\_/subject=cow\_id(trt) type=cs;

lsmeans trt sex year/diff adj=tukey;

run;

title1 'Initial Analysis of Calf Weight prior to breeding season with fixed effects';

title2 'year, treatment, calf gender, and linear day in calving season';

title3 ' and random year\*treatment and repeated measures to account for';

title4 'cows having multiple records';

run;

proc glimmix data=meth3 plot = residualpanel;

class year cow\_id past\_16\_17 trt sex;

model PreBreed\_Calf\_Wt= year trt sex dayseason/s;

random intercept/subject=year\*trt;

random \_residual\_/subject=cow\_id(trt) type=cs;

lsmeans trt sex year/diff adj=tukey;

run;

ods printer close;

\*Weaning weight;

proc glimmix data = meth3 plot = residualpanel ;

class year cow\_id past\_16\_17 trt sex;

model Wt\_\_\_Wean = year trt sex dayseason; \*precalf\_wt as covariate, dayseason and sex were not significant;

random intercept/subject = year\*trt;

random \_residual\_/subject= cow\_id(trt) type = cs;

lsmeans trt year sex /diff adj = tukey;

run;

proc univariate data = meth3;

var Wt\_\_\_Wean;

histogram;

run;

\*Histogram of calf weaning weight;

proc univariate data = meth3;

var Calf\_Wean\_Wt;

histogram;

run;

ods rtf;

\*calf weaning weight;

proc glimmix data = meth3 plot=residualpanel;

class year cow\_id trt sex;

model Calf\_Wean\_Wt = year trt sex dayseason;

random intercept/subject = year\*trt;

random \_residual\_/subject = cow\_id(trt) type = cs;

lsmeans trt sex year/ diff adj = tukey plot = meanplot(join cl);

run;

ods rtf close;

\*Histogram of dec bsc;

proc univariate data = meth3;

var Dec\_BCS;

histogram;

run;

\*BCS

proc glimmix data=meth3;

class year cow\_id past\_16\_17 trt sex;

model chgdeccalv= year trt dayseason/s;

random intercept/subject=year\*trt;

random \_residual\_/subject=cow\_id(trt) type=cs;

lsmeans trt year/diff adj=tukey;

run;

title1 'Analysis of change in Cow BCS from prior to start of treatment (Dec) to';

title2 'Prebreeding with fixed effects year, treatment, sex and day incalving season calved';

title3 ' and random year\*treatment and repeated measures to account for';

title4 'cows having multiple records';

run;

proc glimmix data=meth3;

class year cow\_id past\_16\_17 trt sex;

model chgdecpbrd= year trt sex dayseason/s;

random intercept/subject=year\*trt;

random \_residual\_/subject=cow\_id(trt) type=cs;

lsmeans trt year/diff adj=tukey;

run;

\*Pregnancy;

proc glimmix data = meth3;

class year cow\_id trt sex;

model preg = year trt sex dayseason/dist=binom;

random intercept/subject = year\*trt;;

lsmeans trt year/diff adj = tukey;

run;