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Terry L. Mader University of Nebraska-Lincoln, tmader1@unl.edu

Wanda Kreikemeier Northeast Research and Extension Center, Concord

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# Growth Promoting Agents and Season Effects on Blood Metabolite and Body Temperature Measures

Terry L. Mader Wanda M. Kreikemeier<sup>1</sup>

#### Summary

To assess growth promoting agents efficacy among seasons, triiodothyronine, thyroxine, blood metabolites, and tympanic temperature were measured in summer and winter studies. Within each season, pens of heifers were assigned to one of six growth promotant treatments. Season by growth promotant treatment interactions (P < 0.05) indicated that the combination of estrogen and trenbolone acetate increased triiodothyronine in the winter, whereas trenbolone acetate alone decreased both triiodothyronine and thyroxine in the winter. Maximum tympanic temperature was greater (P < 0.01) in the summer than in the winter, while minimum tympanic temperature was lowered (P < 0.01) in the summer. Changes in blood metabolite levels resulting from the use of growth promotants do not appear to substantially influence seasonal changes in body temperature.

#### Introduction

Within a season, changes in temperature, wind speed, precipitation, and/or radiation can significantly influence physiological and metabolic processes. Physiological characteristics, particularly when cattle are under environmental stress, could be further influenced by anabolic agents. The objective of this experiment was to assess feedlot heifer responses to cold and heat exposure when administered growth promoting agents as determined by blood endocrine levels, plasma urea nitrogen (PUN), and tympanic temperature.

#### Procedure

During a summer and winter season, crossbred Angus, nonpregnant,

yearling heifers (108/season; mean initial BW = 842 lb) were used for obtaining blood samples and tympanic temperatures (TT). Within a season, heifers had been stepped up to a 65.0 NEg (mcal/cwt; DM basis) high-energy finishing diet by the start of each study. Heifers were fed Rumensin and Tylan (Elanco Animal Health, Indianapolis, Ind.) throughout the experimental feeding period. Details of the vaccination, parasite control, and diet regimens used for the experiments have been reported previously (2003 Nebraska Beef Report, pp. 42-45). In early December (winter season), and early June (summer season), heifers were assigned randomly to 12 pens (nine heifers/pen) based on stratification of individual weights. Six growth promotant treatments (two pens of heifers/treatment/season) were imposed as follows: 1) control, 2) estrogenic implant (E; Compudose  $[24 \text{ mg of Estradiol-17}\beta]$ ; Vetlife, West Des Moines, Iowa), 3) androgenic implant (TBA; Finaplix-H [200 mg of trenbolone acetate]; Intervet, Inc., Millsboro, Del.), 4) E + TBA (ET), 5) no implant and fed MGA (MGA; Pharmacia and Upjohn, Kalamazoo, Mich.), and 6) ET implant and fed MGA (ETM). Heifers were bled via jugular puncture and weighed on days 0, 28, 56, and 84. Cattle were fed 104 and 105 days for the winter and summer feeding periods, respectively.

#### Blood Collection and Assays

In both seasons, heifers (four/pen) were bled via jugular puncture and weights were taken on days 0, 28, 56, and 84, beginning at 0800 and prior to being fed. Ten milliliters of blood for plasma were collected into tubes containing sodium heparin. Five milliliters of blood also were collected for serum. After blood collection, tubes were centrifuged (3,400 rpm) for 10 minutes. Plasma and serum fractions were isolated and frozen until analyzed. Serum samples were analyzed for insulin-like growth factor (IGF-1) concentration using RIA with acid–ethanol extraction. Concentrations of thyroxine ( $T_4$ ), triiodothyronine ( $T_3$ ) were quantified with solid phase RIA kits. Samples for  $T_3$  and  $T_4$ analysis were processed as separate assays.

#### Temperature measures

Individual heifers (two heifers/pen; four heifers/treatment/season) were used for obtaining TT, as a measure of body temperature, when ambient temperature was predicted to be < 32°F in the winter and > 77°F in the summer. Tympanic temperatures were recorded using data loggers and thermistor cables (Stowaway, XTI<sup>®</sup>, Onset Computer Corporation, Pocassatt, Mass.). Data loggers were secured in an ear of the heifer using self-adhesive bandages (Vet-Wrap®, 3M Corporation, St. Paul, Minn.) and 2.25 cm athletic tape (Andover Coded Products Inc, Salisbury, Mass.). Tympanic temperature was read every two minutes, with the average recorded every 15 minutes over a seven and five-day period for winter and summer, respectively. On day 28 of each study period, at the time of weighing, ear surface temperature was measured on four heifers from each pen using a Raynger 3i infrared gun (Raytek Corporation, Santa Cruz, Calif.).

### Statistical Analysis

Blood metabolite concentrations were analyzed using Mixed Models procedures of SAS (SAS Inst. Inc., Cary, N.C.) for a split plot in time design. The model included season, growth promotant treatment, and day (used as repeated measures) plus two and three-way interaction. Unstructured covariance analysis was used for  $T_3$ ,  $T_4$ , and PUN, while auto regressive

(Continued on next page)

procedures were used in the IGF-1 analysis. Tympanic temperature and ear surface temperature data were analyzed using Mixed Models procedures of SAS for a completely randomized design. Least squares means were compared using an F-protected LSD (P < 0.05).

#### Results

For the hot and cold periods, during which TT were obtained, ambient temperature averaged 80.1 and 26.8°F, respectively, and ranged from a daily average of 63.5 to 94.8°F for the hot period, and 2.5 to 51.8 for the cold period. Mean THI [temperature humidity index; THI = temperature - (0.55\*(1-rh/100)\*(temperature -58))] was 76.6 for the hot period and 17.4 for the cold period. Based on the livestock safety index, heifers exposed to hot conditions were on the average in the alert (THI > 74) category, but also also exposed to emergency (THI > 83) category conditions, suggesting cattle were under heat stress during most of this period. During the cold TT collection period, THI ranged between 1.6 and 36.8. A THI < 35 has been suggested as being a cold stress threshold; clearly this threshold was reached.

In general, IGF-1 increased (P < 0.05) from day 0 to day 28 in the winter and in the summer (Table 1). However, IGF-1 levels declined (P < 0.05) after day 28 in the winter but tended to be maintained at day 28 levels throughout the summer. Thyroid hormone levels  $(T_3 \text{ and } T_4)$  followed similar trends among seasons across bleed times. As expected,  $T_{a}$ and T<sub>4</sub> levels were numerically elevated in the winter compared with the summer, but were very similar among season on day 84. In general, by day 84, ambient temperatures were declining in the summer, thus stimulating thyroid gland activity, and increasing in the winter which suppresses thyroid gland activity. On day 56, PUN was elevated in the winter and lowered in the summer when compared with day 28 (P < 0.05); thus PUN tended to peak around day 56 in the

Table 1. Mean blood PUN and endocrine concentration for feedlot heifers for season and time of bleed.

Item <sup>a</sup>		Day of bleed <sup>b</sup>					
	0	28	56	84	SE		
IGF – 1, ng/mL Winter Summer	98.42 <sup>cde</sup> 59.50 <sup>c</sup>	129.03 <sup>f</sup> 104.64 <sup>ef</sup>	101.78 <sup>de</sup> 95.43 <sup>de</sup>	90.64 <sup>c</sup> 109.33 <sup>f</sup>	5.83 5.83		
T <sub>3</sub> , ng/mL Winter Summer	1.44 <sup>c</sup> 1.19 <sup>d</sup>	1.48 <sup>c</sup> 0.94 <sup>c</sup>	1.61 <sup>d</sup> 0.96 <sup>c</sup>	1.46 <sup>c</sup> 1.34 <sup>e</sup>	0.05 0.05		
T <sub>4</sub> , ng/mL Winter Summer	66.12 <sup>c</sup> 66.65 <sup>de</sup>	68.03 <sup>c</sup> 53.57 <sup>c</sup>	77.65 <sup>d</sup> 63.29 <sup>d</sup>	68.52 <sup>c</sup> 68.33e	1.95 1.95		
PUN, mg/dL Winter Summer	9.62 <sup>c</sup> 13.69 <sup>d</sup>	13.50 <sup>e</sup> 17.66 <sup>e</sup>	19.13 <sup>f</sup> 13.11 <sup>d</sup>	12.19 <sup>d</sup> 11.60 <sup>c</sup>	0.54 0.54		

 ${}^{a}T_{3}$  = triiodothyronine;  $T_{4}$  = thyroxine; PUN = plasma urea nitrogen.

<sup>b</sup>Number of days into trial. Day by season interaction (P < 0.05) for all metabolites.

<sup>cdef</sup>Means without a common superscript differ (P < 0.05).

Table 2. Effects of growth promoting treatment and season on blood metabolite concentration.

Item <sup>a</sup>	С	Е	TBA	ET	MGA	ETM	SE
IGF – 1, ng/ml							
Winter	97.72	109.70	100.69	116.10	92.48	118.10	11.38
Summer	80.70	90.62	92.49	97.55	82.50	109.48	11.38
Mean	86.71 <sup>c</sup>	100.16 <sup>cd</sup>	96.59 <sup>cd</sup>	106.82 <sup>d</sup>	87.49 <sup>c</sup>	113.79 <sup>d</sup>	7.79
T <sub>3</sub> , ng/ml <sup>e</sup>							
Winter	1.49 <sup>f</sup>	$1.44^{\mathrm{f}}$	1.33 <sup>f</sup>	1.72 <sup>g</sup>	1.51 <sup>fg</sup>	1.50 <sup>f</sup>	0.07
Summer	1.17	1.20	1.14	1.02	1.06	1.05	0.07
Mean	1.33	1.32	1.23	1.37	1.29	1.28	0.05
T₄, ng/ml							
Winter	69.06	70.02	65.01	70.57	67.67	78.05	3.21
Summer	59.77	62.49	62.93	63.01	67.22	62.34	3.21
Mean	64.42	66.26	64.02	66.80	67.44	70.19	2.80
PUN, mg/dl							
Winter	13.70	14.87	12.91	12.05	15.12	13.00	0.73
Summer	14.91	15.11	15.12	12.84	14.04	12.08	0.73
Mean	14.30 <sup>g</sup>	14.99 <sup>g</sup>	14.01 <sup>g</sup>	$12.44^{f}$	14.58 <sup>g</sup>	$12.54^{f}$	0.49

 ${}^{a}T_{3} = triiodothyronine; T_{4} = thyroxine; PUN = plasma urea nitrogen.$ 

 ${}^{b}C = Control (no growth promotant), E = estrogenic implant, TBA = trenbolone acetate implant, ET = estrogenic + TBA, MGA = melengestrol acetate, ETM = E + TBA + MGA.$ 

<sup>cd</sup>Means without a common superscript differ (P < 0.10).

<sup>e</sup>Growth promoting treatment by season interaction (P < 0.05).

<sup>fg</sup>Means without a common superscript differ (P < 0.05).

winter and day 28 in the summer.

In these studies, season x growth promotant interactions were not found (P > 0.05) for ADG, although ADG was greater (P < 0.01; 3.18 vs 2.80 lb) in the winter than in the summer (2003 Nebraska Beef Report, pp. 42-45). In data reported herein, serum IGF-1 concentrations increased (P < 0.05) by ~ 43% from day 0 to 28 in the summer but by only 24% in the winter. Also in the winter, IGF-1 levels declined by ~ 21% from day 28 to 56, thus returning to near levels found on day 0. In the summer, IGF-1 levels only declined by ~ 9% (P > 0.05) from day 28 to 56 and remained above (P < 0.05) day 0 level through day 84. Since baseline IGF-1 (98.4 vs 59.5 mg/ mL) were greater in the winter, differences in ADG are not likely due to the rise or change in IGF-1 over time or among seasons, but partially due to the baseline IGF-1 level associated with the cattle at the start of the study. Also, in the winter, during the period when ambient temperatures decline and approach winter lows, feed intake

 Table 3. Effects of growth promoting treatment and time of bleed on IGF-1 and plasma urea nitrogen (PUN) concentrations in feedlot heifers.

Item <sup>a</sup>		Growth promoting treatment <sup>a</sup>						
	С	Е	TBA	ET	MGA	ETM	SE	
IGF – 1, ng/ml								
0 day	73.72	76.85	67.74	90.05	85.26	80.13	10.45	
28 days	104.72 <sup>bc</sup>	121.01 <sup>cd</sup>	114.09 <sup>bcd</sup>	135.11 <sup>d</sup>	93.67 <sup>b</sup>	132.39 <sup>d</sup>	10.45	
56 days	79.25 <sup>b</sup>	93.47 <sup>bc</sup>	113.14 <sup>c</sup>	100.71 <sup>bc</sup>	86.41 <sup>b</sup>	118.64 <sup>c</sup>	10.45	
84 days	89.16 <sup>b</sup>	109.31 <sup>bc</sup>	91.39 <sup>b</sup>	101.43 <sup>bc</sup>	84.61 <sup>b</sup>	124.01 <sup>c</sup>	10.45	
PUN, mg/dl								
0 day	11.09	12.34	11.35	11.37	12.56	11.21	0.97	
28 days	16.99 <sup>g</sup>	17.35 <sup>g</sup>	15.75 <sup>fg</sup>	12.58 <sup>e</sup>	17.18 <sup>g</sup>	13.60 <sup>ef</sup>	0.97	
56 days	17.18 <sup>g</sup>	18.06 <sup>g</sup>	16.39 <sup>fg</sup>	14.45 <sup>ef</sup>	16.44 <sup>g</sup>	14.20 <sup>e</sup>	0.97	
84 days	11.95	12.20	12.57	11.37	12.12	11.15	0.97	

 $^{a}$ C = Control (no growth promotant), E = estrogenic implant, TBA = trenbolone acetate implant, ET = estrogenic + TBA, MGA = melengestrol acetate, ETM = E + TBA + MGA. Day by growth promoting treatment interaction (P < 0.05).

<sup>bcd</sup>Means without a common superscript differ (P < 0.05).

<sup>efg</sup>Means without a common superscript differ (P < 0.10).

Table 4. Effect of season on tympanic (TT) and ear surface (EST) temperature.

	Sea			
Item	Summer	Winter	SE	
EST, °F	92.26 <sup>b</sup>	56.48 <sup>a</sup>	0.22	
TT, mean, °F	102.27	101.97	0.15	
Maximum, °F	104.07 <sup>b</sup>	102.97 <sup>a</sup>	0.05	
Minimum, °F	100.20 <sup>a</sup>	101.05 <sup>b</sup>	0.04	

<sup>ab</sup>Means without a common superscript differ (P < 0.01).

is stimulated which resulted in greater PUN levels that were found on day 56. In the summer, ambient temperature would be peaking around day 56, thus suppressing feed intake resulting in blood PUN being lowered. This decline in summer PUN levels could be due to the decrease in DMI.

There was no (P > 0.05) growth promoting agent by season interaction for serum IGF-1,  $T_4$ , or PUN concentration (Table 2). Across both seasons, IGF-1 tended to be increased (P < 0.10) in ET and ETM treated heifers when compared with control heifers. No differences in  $T_4$  were observed among growth promotant treatments within or among season. There was a growth promoting treatment by season interaction (P < 0.05) for  $T_4$ 

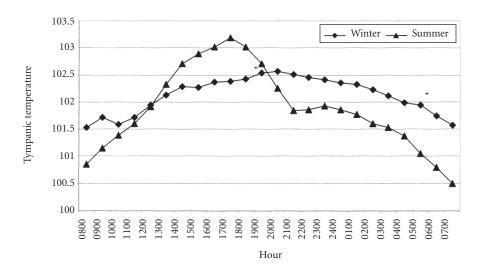


Figure 1. Effects of season on tympanic temperature over a 24-hour period. \*Means differ (P < 0.05; SE = 0.18).

concentration. The ET treated heifers had increased (P < 0.05) T<sub>3</sub> levels in the winter when compared with control and other implanted heifers. Across season, heifers receiving ET (ET and ETM treatments) had lower PUN levels.

A bleed time by growth promotant treatment interaction was not found for thyroid hormones but was found (P < 0.05) for IGF-1 and PUN (Table 3). In general, when compared with control heifer groups, ET and ETM treated heifers had greater (P < 0.05) IGF-1 concentrations on day 28, whereas the ETM and TBA treated cattle had greater IGF-1 concentrations on day 56; only the ETM treated heifers had greater IGF-1 concentrations on day 84. Thus, the ETM treated cattle had consistently greater IGF-1 concentration during the feeding period, which is supported by the tendency (P < 0.10) for those same heifers plus the ET treated group to have lower PUN concentrations (days 28 and 56) than the control heifer group.

Ear surface temperatures were 92.3°F and 56.5°F (*P* < 0.01), respectively for summer and winter (Table 4). The ear surface temperatures were recorded in the event growthpromoting agent by season interactions could be attributable to payout of the implant. Average tympanic temperature was not different (P > 0.05) between seasons. A greater range in TT was found in the summer than in the winter. Maximum TT was greater (P < 0.01) and minimum TT was lower (P < 0.01) in the summer than in the winter. Analysis of hourly data (Figure 1) indicate that peak summer TT occurs around 1700 while peaks in winter TT are not as evident. Also, minimum summer TT were found at 0700. Difference in TT between summer and winter were found at 0500, 0600, 0700, 0800, 1600, 1700, and 2100 with the diurnal TT pattern being flatter in the winter than in the summer.

There was a growth promoting treatment by season interaction (P < 0.05) for ear surface temperature (Continued on next page) (Table 5). In the summer, there was no difference between ear surface temperatures across growth promoting treatments while in the winter, the MGA treated heifers had ear surface temperatures similar to control but lower (P < 0.05; 51.1 vs 58.5 °F) than groups receiving implants. These data suggest that, at least in the winter, implanting can elevate ear surface temperatures as much as 10°F, however, overall ear surface temperatures in the winter are over 36°F lower than those found in the summer.

A growth promoting treatment by season interaction was evident for average maximum TT (P < 0.05) and for average minimum TT (P < 0.10), although the interaction was not evident for mean TT (Table 5 and Figure 1). Mean TT were similar among growth promotant treatment among seasons. Numerically, control heifer groups had greater maximum TT, particularly in the winter, with the MGA heifers having the lowest maximum TT in both seasons. The ET treated cattle had greater (P < 0.05) maximum TT in the summer when compared with MGA fed groups (MGA and ETM). However, in the winter, cattle receiving E and/or MGA (E, ET, MGA, and ETM) had lower maximum TT than control cattle. Differences in minimum TT tended to be found only in the summer, with E treated cattle having greater minimum TT than TBA and ETM treatment groups.

The data indicate that when cattle get hot in the summer, they tend to overcompensate at night by ridding

Table 5. Effect of growth promoting treatment and season on tympanic temperature (TT) and ear surface temperature (EST).

Item	Growth promoting treatment <sup>a</sup>						
	С	Е	TBA	ET	MGA	ETM	SE
EST, °F <sup>b</sup>							
Winter	54.50 <sup>cd</sup>	55.58 <sup>d</sup>	56.48 <sup>d</sup>	59.18 <sup>d</sup>	51.08 <sup>c</sup>	62.24 <sup>d</sup>	0.53
Summer	92.66	92.84	91.04	92.48	93.20	91.40	0.53
Mean	73.58	74.30	73.76	75.92	72.14	76.82	0.33
Mean TT, °F							
Winter	102.63	101.79	100.53	101.64	101.59	101.97	0.37
Summer	102.15	102.25	102.09	103.46	102.85	101.75	0.37
Mean	102.40	102.02	102.18	102.56	102.22	101.86	0.24
Maximum TT, °F <sup>b</sup>							
Winter	104.14 <sup>d</sup>	102.65 <sup>c</sup>	103.14 <sup>cd</sup>	102.45 <sup>c</sup>	102.43 <sup>c</sup>	102.99 <sup>c</sup>	0.17
Summer	104.41 <sup>de</sup>	103.95 <sup>cde</sup>	104.32 <sup>de</sup>	104.79 <sup>e</sup>	103.50 <sup>c</sup>	103.64 <sup>cd</sup>	0.17
Mean	104.29 <sup>d</sup>	103.30 <sup>c</sup>	103.64 <sup>cd</sup>	103.62 <sup>cd</sup>	102.97 <sup>c</sup>	103.32 <sup>c</sup>	0.08
Minimum TT, °F <sup>b</sup>							
Winter	101.12	100.98	101.23	100.98	100.74	101.26	0.15
Summer	$99.84^{\mathrm{f}}$	100.98 <sup>g</sup>	99.82 <sup>f</sup>	100.31 <sup>fg</sup>	$100.44^{fg}$	99.72 <sup>f</sup>	0.15
Mean	100.49	100.98	100.53	100.65	100.60	100.49	0.07

<sup>a</sup>C = Control (no growth promotant), E = estrogenic implant, TBA = trenbolone acetate implant, ET =

E + TBA, MGA = melengestrol acetate, ETM = E + TBA + MGA.

<sup>b</sup>Growth promotant by climatic condition interaction (P < 0.10).

<sup>cde</sup>Means without a common superscript differ (P < 0.05).

<sup>fgh</sup>Means without a common superscript differ (P < 0.10).

the body of heat (resulting in a lower TT) in preparation for subsequent heat episodes. Thus, the range in TT will be greater in the summer than in the winter. The lower nighttime TT appears to enable cattle to prepare for the heat of the day, while greater overall TT in the winter buffers the animal against cold threats. The greater minimum TT found in the E treatment group in the summer would suggest E implanted cattle may be more susceptible to heat stress. If E increases TT, the mechanism by which MGA tends to lower TT is unclear, since the growth promoting response of both products are mediated through estrogen receptors. The estrus suppressing effect of MGA, which is not present in implants, is possibly responsible for any lowering of TT particularly in the ETM group. However, control heifers had greater overall maximum TT. Although limited growth promotant by season interactions existed, changes in blood metabolite levels resulting from the use of growth promotants do not appear to substantially influence seasonal changes in body temperature.

<sup>&</sup>lt;sup>1</sup>Terry Mader, professor; Wanda Kreikemeier, former graduate student, Department of Animal Science, Northeast Research and Extension Center, Concord.