

Effects of Ambient Temperature and Relative Humidity on Serum Prolactin and Growth Hormone in Heifers^{1,2} (39273)

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We reported that increasing ambient temperature from 21 to 27° increased basal concentrations of serum prolactin (PRL) and increased PRL release by thyrotropin-releasing hormone (TRH) in heifers (1). Conversely, lowering the ambient temperature from 21 to 10° reduced basal serum concentrations and TRH-induced responses of PRL. Recent work in rats has confirmed that serum PRL increases with warm temperatures and declines with cold temperatures (2). The objective of the present study was to determine the effects of relatively more extreme changes in ambient temperature on serum concentrations of PRL and growth hormone (GH) in heifers. Also, the effects of relative humidity on these hormones was determined.

Material and methods. Twelve Holstein heifers 3 months of age were randomly paired, and the pairs were confined in two environmental chambers. Relative humidity was maintained at $50 \pm 10\%$ and animals were subjected to 12 hr of light daily commencing at 0800 hr. Other management details were as previously described (1).

Heifers were maintained initially for 10 days at $21 \pm 0.5^\circ$. After this period a jugular vein of each heifer was cannulated (Silastic, 95-cm length; 2.16-mm o.d., Dow Corning Corp.). Two pairs of heifers were assigned simultaneously to treatments according to an incomplete block design (3). In the first block, pair one was exposed to $4.5 \pm 0.5^\circ$, whereas the second pair was maintained at $21 \pm 0.5^\circ$. The second block consisted of a

third pair of heifers subjected to 4.5° at the same time the fourth pair was exposed to $32 \pm 0.5^\circ$. The fifth and sixth pairs were housed at 21 and 32° , respectively, and comprised the third block. Duration of exposure was 9 days. On the first day of the experiment, chamber temperatures were decreased from 21 to 4.5° over a period of 12 hr (about 1.4° per hr); the increase from 21 to 32° required 8 hr (about 1.4° per hour).

On the ninth day of the experiment all heifers were injected once via jugular cannula with $10 \mu\text{g}$ synthetic TRH (pyro-glutamyl-histidyl-proline amide)³ between 1100 and 1330 hr. This procedure was used to evaluate the ability of the anterior pituitary to release PRL and GH (4).

Blood samples were collected via cannulae at 2-hr intervals for 16 hr on the first day of the experiment when the ambient temperatures were changed from 21° . Blood was sampled daily on Days 2 and 9 shortly after 0800 hr. On the ninth day, blood was sampled 30, 20, 10, and 0 min before administration of TRH and then at 5, 10, 15, 20, 25, 30, 45, and 60 min after TRH injection. Sera were stored at -20° until assayed for PRL (5) and GH (6). Bovine PRL (NIH-B₁)⁴ and bovine GH (NIH-B₁₂)⁴ were used as reference standards.

In a preliminary experiment four heifers were subjected to $50 \pm 10\%$ relative humidity for 6 days; simultaneously four other heifers were exposed to $90 \pm 10\%$ relative humidity. Temperature was maintained at 21° , and they received 12 hr of light daily. Blood was sampled once daily at 0800 hr, and serum PRL and GH measured as described above.

Results and discussion. As ambient tem-

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⁴ Kindly supplied by National Institutes of Health, Bethesda, Maryland.

perature was reduced from 21 to 4.5° during 12 hr, serum PRL decreased linearly ($P < 0.01$) 0.60 ± 0.14 ng/ml/° (Fig. 1). Conversely, as ambient temperature was increased from 21 to 32°, serum PRL increased linearly ($P < 0.05$) 1.17 ± 0.34 ng/ml/°. Average serum PRL concentrations in control heifers maintained at 21° during the comparable 12-hr interval randomly fluctuated between 7.8 and 26.1 ng/ml.

Serum PRL concentrations averaged 2.6, 13.0, and 27.7 ng/ml ($P < 0.05$) between Days 2 and 9 when temperatures were maintained at 4.5, 21, and 32°, respectively. These results confirm our previous findings (1), but the more extreme temperatures used in the present study resulted in a greater divergency in serum PRL concentrations. McMurtry *et al.* (7) reported that PRL in cows milk increased as ambient temperature decreased below 5°. To our knowledge there are no reports of changes in serum PRL at temperatures below 4.5°, thus it is not known whether the effects of cold temperatures on PRL in milk were associated with transfer of PRL into milk or an effect on synthesis and/or release of PRL at the anterior pituitary.

Within 5 min of treating heifers with TRH, serum PRL increased ($P < 0.01$) from a pretreatment average of 20.4 ± 4.5 ng/ml at 32° to 109.8 ± 27.1 ng/ml (Fig. 2). At 21° PRL increased ($P < 0.01$) from a pretreatment average of 15.7 ± 0.5 ng/ml to 62.8 ± 27.2 ng/ml 5 min after TRH. Al-

though serum PRL numerically increased from 1.6 ± 0.3 ng/ml before TRH was administered to a peak of 2.5 ± 0.6 ng/ml 5 min after TRH was injected when the ambient temperature was 4.5°, the increase was not significant ($P > 0.10$). Although previous reports (1, 2) suggested a reduced increase in serum PRL after injections of TRH at colder temperatures, we believe the present data are the first to show that release of the hormone may be blocked at temperatures of 4.5°. The concentrations of PRL in serum during the 60 min after TRH was injected were lower ($P < 0.02$) at 4.5° as compared with 21 or 32°, but there was no significant difference between the responses at 21 or 32° ($P > 0.10$).

Serum GH concentrations averaged 14.4 ng/ml for all heifers during the 2-hr interval before the ambient temperature was changed from 21°. During the 12-hr interval on Day 1 of the experiment when temperatures were reduced to 4.5°, maintained at 21°, or increased to 32°, serum GH concentrations randomly fluctuated between 1.9 and 26 ($\bar{x} = 11.0$), 4.6 and 8.9 ($\bar{x} = 7.2$), and 7.1 and 23.3 ($\bar{x} = 13.3$) ng/ml, respectively. These means were not significantly different ($P > 0.10$). Serum GH between Days 2 and 9 averaged 4.0 ± 0.7 , 6.3 ± 1.2 , and 9.4 ± 1.1 ng/ml at 4.5, 21, or 32°, respectively. Although GH tended to increase with increasing temperature, these differences were not significant ($P > 0.10$). These data are in agreement with our (8)

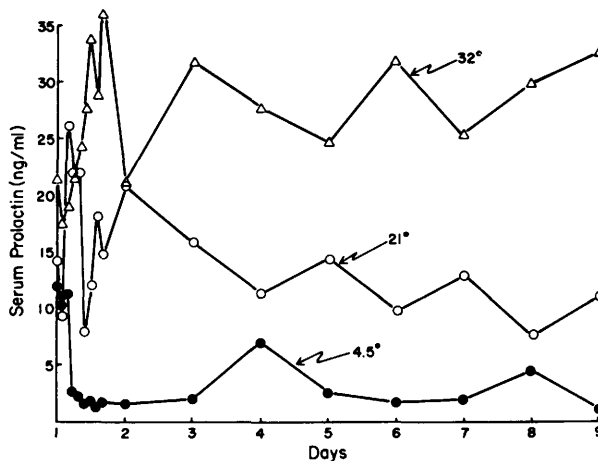


FIG. 1. Serum prolactin concentrations in heifers exposed to ambient temperatures of 4.5, 21, and 32°. Each point for serum prolactin is the mean of four heifers.

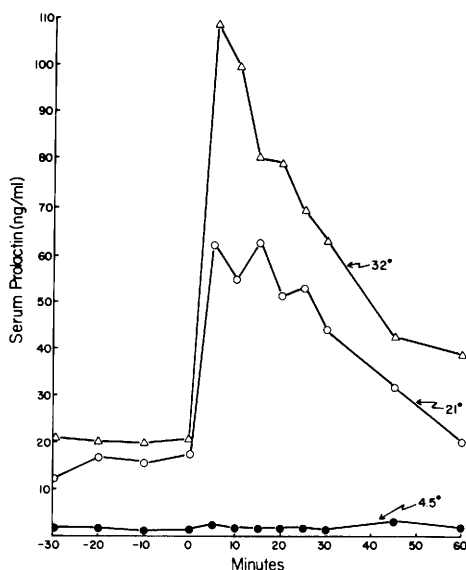


FIG. 2. Serum prolactin response to injection of 10 μ g of thyrotropin-releasing hormone (at 0 min) into each of four heifers at 4.5, 21, and 32°.

previous report that season of the year did not affect serum GH.

During the 30 min before TRH was injected on Day 9, the mean serum GH concentration of 1.8 ± 0.3 ng/ml at 4.5° was lower ($P < 0.05$) than the pre-TRH concentrations at 21° (4.7 ± 0.4 ng/ml) or 32° (7.4 ± 0.9 ng/ml). Serum concentrations during the 60 min after TRH increased ($P < 0.01$) to peak values of 8.1 ± 4.0 , 19.6 ± 4.4 , and 16.4 ± 2.6 ng/ml at 4.5, 21, and 32°, respectively (Fig. 3). Ambient temperature did not affect ($P > 0.10$) the increase serum GH concentrations after TRH. From these data, we conclude that the effects of ambient temperature on serum PRL after TRH do not apply to serum GH.

Serum PRL averaged 12.9 ± 1.1 and 21.7 ± 7.6 ng/ml at 50 and 90% relative humidity, respectively, but these means were not significantly different ($P > 0.10$). However, serum GH averaged 6.2 ± 0.8 and 10.2 ± 1.5 ng/ml at 50 and 90% relative humidity, respectively; and these differences approached significance ($P \approx 0.10$).

Although relative humidity did not significantly affect serum PRL in our preliminary experiment, the mean at 90% was almost double that at 50% relative humidity. Thus, whether or not relative humidity contributes

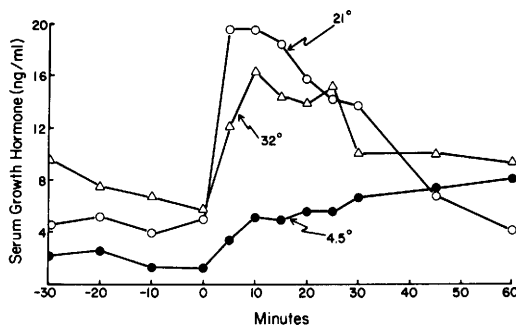


FIG. 3. Serum growth hormone response to injection of 10 μ g of thyrotropin-releasing hormone (at 0 min) into each of four heifers at 4.5, 21, and 32°.

to the rise in serum PRL observed during the warmer seasons of the year in cattle (9, 10) will require additional experimentation. Furthermore, we (11) have observed that long daylength increases serum PRL. It is now apparent that several environmental stimuli are associated with the seasonal changes in serum PRL concentrations. However, the physiological significance of these changes in serum PRL remain to be elucidated.

Summary. Twelve heifers were exposed to 21° ambient temperature for 10 days, and then subjected to 4.5, 21, or 32° for 9 days in controlled environmental chambers. Serum prolactin (PRL) decreased linearly ($P < 0.01$; 0.6 ng/ml/°) as the temperature was reduced during the first day from 21 to 4.5°; serum PRL increased linearly ($P < 0.05$; 1.17 ng/ml/°) as the temperature was increased from 21 to 32°. Between Days 2 and 9 serum PRL averaged 2.6, 13.0, and 27.7 ng/ml ($P < 0.05$) at 4.5, 21, and 32°, respectively. Injection of thyrotropin-releasing hormone (TRH) caused serum PRL to increase within 5 min from 20.4 to 109.8 ng/ml at 32°; at 21° serum PRL increased from 15.7 to 62.8 ng/ml, whereas at 4.5° serum PRL did not respond to TRH. Serum growth hormone (GH) averaged 4.0, 6.3, and 9.4 ng/ml at 4.5, 21, and 32°, respectively, but these means were not different ($P > 0.10$). TRH released GH at all temperatures tested, but the quantity released was unaffected by ambient temperature. Relative humidities of 50 and 90% did not significantly alter ($P > 0.05$) serum PRL or GH. We conclude that ambient temperature

affects basal and TRH-stimulated concentrations of serum PRL but not GH in heifers.

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1. Wettemann, R. P., and Tucker, H. A., *Proc. Soc. Exp. Biol. Med.* **146**, 908 (1974).
 2. Mueller, G. P., Chen, H. T., Dibbet, J. A., Chen, H. J., and Meites, J., *Proc. Soc. Exp. Biol. Med.* **147**, 698 (1974).
 3. Cochran, W. G., and Cox, G. M. "Experimental Designs," p. 376. John Wiley and Sons, New York (1957).
 4. Convey, E. M., Tucker, H. A., Smith, V. G., and Zolman, J., *Endocrinology* **92**, 471 (1973).
 5. Koprowski, J. A., and Tucker, H. A., *J. Dairy Sci.* **54**, 1675 (1971).
 6. Purchas, R. W., Macmillan, K. L., and Hafs, H. D., *J. Anim. Sci.* **31**, 358 (1970).
 7. McMurtry, J. P., Malven, P. V., Arave, C. W., Erb, R. E., and Harrington, R. B., *J. Dairy Sci.* **58**, 181 (1975).
 8. Koprowski, J. A., and Tucker, H. A., *Endocrinology* **93**, 645 (1973).
 9. Koprowski, J. A., and Tucker, H. A., *Endocrinology* **92**, 1480 (1973).
 10. Schams, D., *Acta Endocrinol.* **71**, 684 (1972).
 11. Bourne, R. A., and Tucker, H. A., *Endocrinology* **97**, 473 (1975).
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