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Effect of Thyrotropin-Releasing Hormone (TRH) on Plasma Glucocorticoids and Thyroxine in Cattle[1] (38869)

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Numerous investigations have accumulated evidence that thyrotropin releasing hormone (TRH) stimulates secretion of thyrotropin (1–3). Convey *et al.* (4) and Vanjonack *et al.* (5) demonstrated a marked elevation of serum and plasma thyroxine concentration with systemic infusion of TRH in cattle. Studies from this laboratory (5) indicate a peak response at 30 min followed by a decline and then a gradual elevation of plasma thyroxine up to at least 6 hr. However, the *in vivo* effects of TRH on plasma glucocorticoids have not been clearly resolved, especially in cattle. Steiner *et al.* (6) reported a rapid transient depression of corticosterone in rats after TRH administration while other studies on cattle (4), humans (7–9), and rats (10) have not observed this effect. Therefore, the objective of this study was to measure the effects of repeated systemic infusions of TRH (200 μg twice daily) on plasma thyroxine and glucocorticoid concentrations in nonlactating cattle and to determine if basal plasma thyroxine concentration could be increased and sustained by repeated infusion of TRH.

Materials and Methods. In the first experiment, two mature nonlactating Holstein cows were acclimated to a constant temperature of 18°, 50% relative humidity for 7 days in the Missouri Climatic Laboratory and prepared with a jugular catheter 24 hr prior to experimentation. The general procedure of each AM (0800–1000 hr) and PM (2000–2200 hr)–sample period was to take a control blood sample; then TRH (200 μg) or saline was infused via jugular catheter (rapidly, approximately 10-sec duration, then flushed with saline) with blood samples taken for approximately 1 hr. Saline infusions were performed every 12 hr (AM–PM) for 48 hr prior to and after the TRH infusion period (every 12 hr, AM–PM, for 72 hr).

A second experiment was performed on two Hereford heifers which were prepared with a jugular catheter 48 hr prior to thermal exposure (35°). After 48 hr of heat exposure, the cattle were infused via jugular catheter with 400 μg TRH and samples were taken 0, 15, 30, 45, and 60 min post-TRH infusion.

All blood samples were treated with 10 units sodium heparin per ml, immediately chilled, plasma separated, and frozen until day of assay.

The infusions and blood sampling were performed remotely to avoid disturbances or excitation. Total plasma thyroxine (T_4) concentrations were determined by a radioimmunoassay procedure (11) with the exception of the charcoal-separation technique of Herbert *et al.* (12) and total glucocorticoids by a competitive protein-binding procedure (13). Studies have shown that the primary glucocorticoid secreted in cattle is cortisol with a cortisol:corticosterone ratio of approximately 3:1 (14–16). All statistical analyses were performed by analysis of variance and protected least significant difference test.

Results. The mean effect (72 hr) of repeated systemic infusion of TRH on plasma thyroxine concentration is shown in Table I. The TRH treatment exhibited an 8% elevation in control values, 41% elevation in mean peak (20–40 min post-TRH) response, and 11% elevation in 1 hr post-TRH infusion values. The general relationship of time and TRH infusion is illustrated in Fig. 1. During saline infusion treatment PM values were slightly increased and both the AM and

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TABLE I. MEAN EFFECT OF REPEATED SYSTEMIC INFUSION OF TRH ON PLASMA THYROXINE AND GLUCOCORTICOIDS CONCENTRATIONS.[a]

Treatment	*N*	Thyroxine (ng/ml)	Glucocorticoids (ng/ml)
Saline treatment	16	84.0±11.8a	24.6±2.2a
TRH-infusion treatment			
1 Control	12	91.0±10.9a	19.8±2.7a
2 TRH peak response (20–40 min post-TRH)	12	118.1±15.9b	6.7±2.1b
3 1 hr post-TRH	12	93.1±12.2a	8.2±1.9b
Post-TRH saline treatment	12	84.1±12.3a	18.7±3.0a

[a] Columns 3 and 4 show SEM. Also in columns 3 and 4 different letter denotes $P < 0.05$).

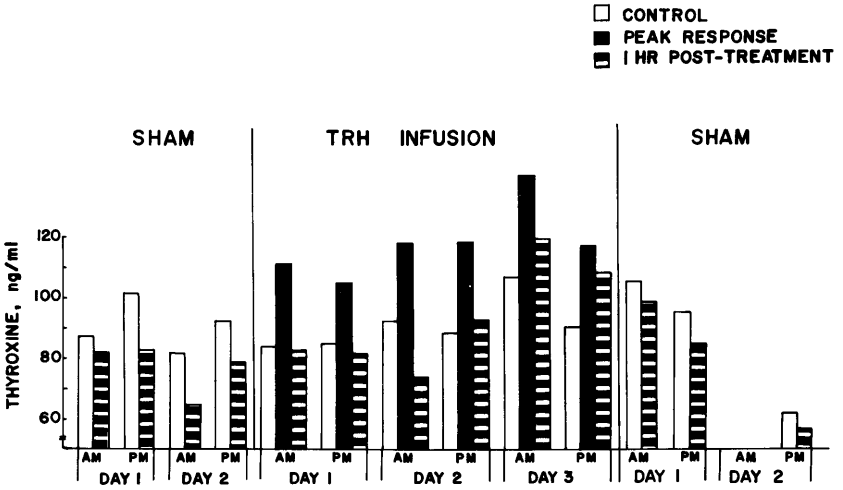

FIG. 1. Plasma thyroxine (ng/ml) response to repeated systemic infusion (twice daily) of 200 μg TRH. Peak response was observed at 20–40 min postinfusion of TRH.

PM values were slightly increased and both the AM and PM 1 hr postsaline values were decreased from control. The effect of repeated TRH infusion was an increase of plasma thyroxine within 20–40 min of TRH infusion during the 3 experimental days. On day 3 (AM) of the TRH treatment, the control, mean peak response (20–40 min), and 1 hr post-TRH values were all at their highest levels with day 3, 1 hr post-TRH values being higher ($P < 0.05$) than comparable day 1 and 2 values. During the post-TRH infusion (saline) period a depression ($P < 0.05$) of plasma thyroxine levels was observed on day 2 (Fig. 1).

The mean effect of repeated systemic infusion of TRH (iv) on plasma glucocorticoid concentrations is shown in Table I. The TRH treatment period exhibited a 19.5% (NS) depression in control values, 73.2% ($P < 0.01$) depression in 20- to 40-min post-TRH values, and a 66.6% ($P < 0.01$) depression in 1 hr post-TRH infusion values. The general relationship of time and TRH infusion is illustrated in Fig. 2. Clearly is shown a depression of plasma corticoid concentration within the first hour of TRH infusion. In the post-TRH saline period day 1 values were somewhat depressed (NS) as compared to pre-TRH saline period; but on day 2 of post-TRH saline period, the values were clearly in the range of pre-TRH saline values.

Results of the second experiment are shown in Table II. The effect of exposure of Hereford heifers to 35° for 48 hr was a 13.5%

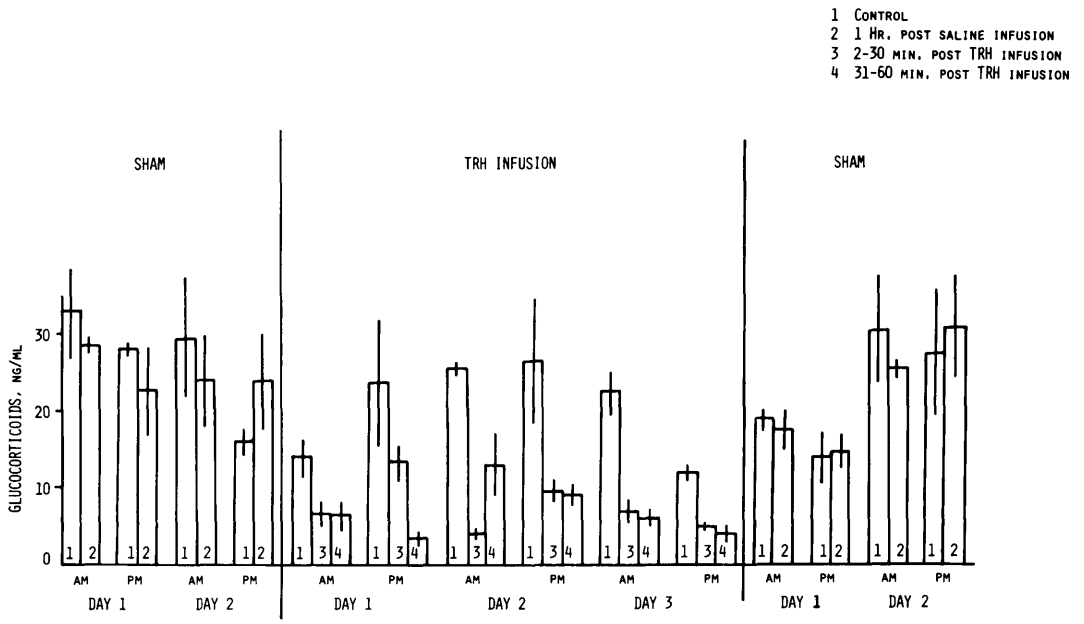

FIG. 2. Plasma glucocorticoid (ng/ml) response to repeated systemic infusion (twice daily) of 200 μg TRH.

and 50.2% depression of plasma thyroxine and glucocorticoids concentrations, respectively. The effect of 400 μg (iv) of TRH was 21.9% elevation ($P < 0.05$) of thyroxine values at 45 min postadministration and a depression ($P < 0.05$) of glucocorticoid levels at 15 and 90 min, with 45-min sample being maximally depressed ($P < 0.01$). At 60 min glucocorticoid values show evidence of recovery but are still significantly depressed. Responses of thyroxine and corticoids to TRH during high thermal exposures were similar in direction but less in magnitude as compared to the thermoneutral study values although the relationship of elevated thyroxine and depressed glucocorticoid concentrations existed between experiments.

TABLE II. EFFECT OF TRH ON HEREFORD PLASMA GLUCOCORTICOIDS AND THYROXINE LEVELS AFTER 48-HR PREEXPOSURE TO 35°.

Time (min) post-TRH administration	Thyroxine		Glucocorticoids	
	Ng/ml	% Change from 48hr at 35°	Ng/ml	% Change from 48 hr at 35°
0 (18.5°)	121.5	—	27.9	—
0 (35°)	105.0	—	13.9	—
15 (35°)	106.5	↑ 1.4%	7.3	↓47.4%
30 (35°)	115.0	↑ 9.5%	6.7	↓51.8%
45 (35°)	128.0	↑21.9%	2.2	↓84.2%
60 (35°)	116.0	↑10.5%	7.2	↓48.2%

Discussion. The results of this study indicate that bovine plasma thyroxine levels can be repeatedly elevated (20–40 min post-TRH) by systemic infusion of TRH during the 3 experimental days and that plasma thyroxine levels show a depression ($P < 0.05$) 48 hr post-TRH treatment. Previous studies (4, 5) have demonstrated that the elevation of T_4 levels are observed for 5–6 hr post-TRH administration. The peak response observed in this study and a previous study (5) may represent the direct action of TRH and/or TSH with the decline from 20–40 min to 1 hr post-TRH infusion values representing distribution of the hormone. The depressed post-TRH saline period levels may reflect a lack of responsiveness to endogenous TRH stimulation and/or lower TSH levels (17) due to the previous high thyroxine levels having a suppressive effect (17, 18) on the hypothalamo–pituitary–thyroid axis.

The results of this study also indicate that

TRH either directly or indirectly may cause a relatively rapid but transient depression (within 1 hr) of plasma glucocorticoids. This rapid depression suggests that TRH in abnormally high concentrations throughout the body (due to peripheral infusion) has a direct or indirect peripheral effect on glucocorticoid secretion, utilization, or excretion. Another possibility is that the hypothalamo–pituitary–thyroxine axis may exert some transient inhibitory effect on the hypothalamo–pituitary–adrenal axis via TRH.

Other studies have not observed this effect of TRH [cattle (4), humans, (7–9), rats (10)] on plasma glucocorticoid concentration or ACTH, although it is not clear whether these differing reports are due to a direct effect of TRH or to some "stress" associated with its administration (10). In this study administration of TRH and blood samples were obtained remotely to avoid disturbance or excitation to the animals. Steiner *et al.* (6), on the other hand, reported a rapid corticosterone depression after TRH administration and suggested that either the corticosterone is rapidly transformed into an unassayable form or rapidly eliminated from circulation; also Sakiz and Guillemin (19) Reported that when the pituitary is induced to secrete TSH by TRH, it secretes less ACTH in response to stress.

The results of this study also indicate diminished effectiveness of TRH to elevate plasma thyroxine concentrations under imposed conditions of high thermal exposure. These findings in cattle are consistent with a study of Vanjonack and Johnson (20) which reported diminished effectiveness of TRH (im administration) to sustain plasma thyroxine concentration with chronic high thermal exposure. A factor that may also be considered in responsiveness to TRH is basal thyroxine concentrations. The heifers' basal thyroxine concentrations (105 ng/ml at 35°) were higher than the dairy cows' (84 ng/ml), with Tal and Sulman (21) showing normal responsiveness to TRH with thermal exposure in rats although the effect of high thermal exposure was to decrease basal serum thyroxine concentrations.

In conclusion, the results of this study indicate repeated systemic infusion of TRH is effective in repeatedly elevating plasma thyroxine concentrations which is associated with a relatively rapidly depression of plasma glucocorticoids. In view of these findings, further studies are warranted to determine the possible effects of TRH on the hypothalamo–pituitary–adrenal axis with attention to the effects of stress on glucocorticoid response to TRH. Because of the rapid depression of plasma glucocorticoids and thyroxine values nearly returning to baseline after 1 hr, further studies should be undertaken to determine the effects of TRH on glucocorticoid and thyroxine secretion and metabolic clearance rates. Also, because TRH has been shown to be a stimulus to lactation in humans (22) and cattle (23) and the results of this study indicate that TRH is effective in repeatedly elevating T_4 levels, further studies should be undertaken to determine effective dose, method of administration, and duration of treatment for its applicability to animal agriculture.

Summary. The repeated systemic infusion of TRH (200 μg, twice daily) was effective in repeatedly elevating plasma T_4 levels (20–40 min postinfusion) for the 3 experimental days. The post-TRH treatment (saline infusion) exhibited a depression of plasma thyroxine levels, possibly explained by previous high thyroxine levels exerting a suppressive effect on the hypothalamic–pituitary–thyroid axis. A depression of plasma glucocorticoid levels was shown within 1 hr in both experiments after TRH administration. The results indicate that TRH administration (systemic) is effective in temporarily elevating thyroxine levels and simultaneously lowering plasma glucocorticoids in cattle.

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