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Effect of Heat Stress on Plasma Concentrations of Prolactin and Luteinizing Hormone in Ewes

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Abstract

Basal concentrations of prolactin but not luteinizing hormone were elevated in ewes by 8–10 h of heat stress given daily during the first 11 days of their oestrous cycle. However, the prolactin and luteinizing hormone responses to thyrotrophin releasing hormone and gonadotrophin releasing hormone were unaffected.

Introduction

The semi-arid tropical area of Australia is characterized by unshaded plains with summer ambient temperatures reaching 46°C, and for 6 months the monthly maximum temperature exceeds 35°C (MacFarlane *et al.* 1956). These conditions require considerable physiological adaptation (Hopkins *et al.* 1978) but the effect of such hot environments on the hormone concentrations of sheep has received little attention.

It has been observed in cattle and rats that circulating prolactin concentrations increase as the ambient temperature increases (Mueller *et al.* 1974; Wettmann and Tucker 1974; Smith *et al.* 1977). Considering prolactin is a hormone connected with both lactation and water regulation (Nicoll and Bern 1972) it was of interest to determine the effect of heat stress on prolactin concentrations in the ewe.

The effect of heat on luteinizing hormone secretion is uncertain. A fall in luteinizing hormone concentration with elevated temperatures has been reported in beef cattle (Madan and Johnson 1973) but a rise in luteinizing hormone concentration has been found in pigs (Riggs *et al.* 1974). To the authors' knowledge, no studies on the effect of heat stress on luteinizing hormone concentrations in ewes have been reported.

Materials and Methods

The experiment was conducted at Julia Creek, Queensland (21°S., 142°E.) during October (i.e. spring). Twelve mature Merino ewes [live weight 34·50 ± 2·18 kg (mean ± s.e.)] were randomly allocated to two equal live weight groups of six ewes. During the first 11 days of their oestrous cycle (day 0 = day of oestrus) group I ewes were exposed to a hot environment (ambient temperature 45–52°C for 8–10 h/day; relative humidity 30–80%) which has been reported sufficient to induce heat stress (Hopkins *et al.* 1978). At the same time group II ewes were housed in a thermoneutral environment where ambient temperature ranged between 30 and 35°C for 8–10 h/day and relative humidity was 30–70%. Water and chopped Flinders grass hay were available *ad libitum* throughout the experiment.

Jugular blood samples were collected from indwelling silastic cannulae at 2-h intervals for 9 h on days 3, 4, 7 and 9 of the oestrous cycle. After 10 days of treatment all ewes were given an

intravenous injection of 20 μg gonadotrophin releasing hormone and on the following day 10 μg thyrotrophin releasing hormone. Blood samples were collected at $\frac{1}{2}$-h intervals for 4 h before and for 6 h after the gonadotrophin releasing hormone injection and the thyrotrophin releasing hormone injection respectively. The resultant plasma was assayed for luteinizing hormone (Goding *et al.* 1969) and prolactin (Hooley *et al.* 1978).

For prolactin the within-assay variation was less than 15% over the range $0{\cdot}6\pm 0{\cdot}1$ to $13{\cdot}6\pm 2{\cdot}2$ ng per tube (mean $\pm$s.e.m.) and the samples were diluted to fit this range. Three plasma pools were assayed repeatedly to measure between-assay variation and these read $90{\cdot}2\pm 29{\cdot}0$ ($n = 14$), $95{\cdot}9\pm 8{\cdot}5$ ($n = 8$) and $61{\cdot}7\pm 3{\cdot}6$ ng/ml ($n = 10$). The sensitivity of the prolactin assay was 0·2 ng/ml (0·02 ng per tube) using NIH–P–S8 standard.

Within-assay variation for luteinizing hormone was less than 20% over the usable portion of the standard curve. Between-assay variation calculated from two plasma pools was $1{\cdot}2\pm 0{\cdot}1$ ($n = 12$) and $6{\cdot}8\pm 0{\cdot}2$ ng/ml ($n = 12$). Assay sensitivity was $0{\cdot}3\pm 0{\cdot}1$ ng/ml ($n = 12$) using LER 1374A standard.

Results

Respiratory rates and rectal temperatures were consistently elevated during the period when the ewes in group I were exposed to hot conditions (temperature 45°C). During the heat treatment period the mean ($\pm$s.e.) respiratory rate of the group I ewes was $164{\cdot}7\pm 4{\cdot}5$ per min, which was significantly ($P < 0{\cdot}001$) greater than that for group II ewes ($61{\cdot}6\pm 5{\cdot}3$ per min). Rectal temperatures were higher in group I ewes ($40{\cdot}7\pm 0{\cdot}1$°C, mean $\pm$ s.e.) than in group II ewes ($40{\cdot}4\pm 0{\cdot}2$°C) but not significantly different ($P > 0{\cdot}05$). The ewes in group I were considered heat stressed (HS) and group II ewes non-heat stressed (NHS).

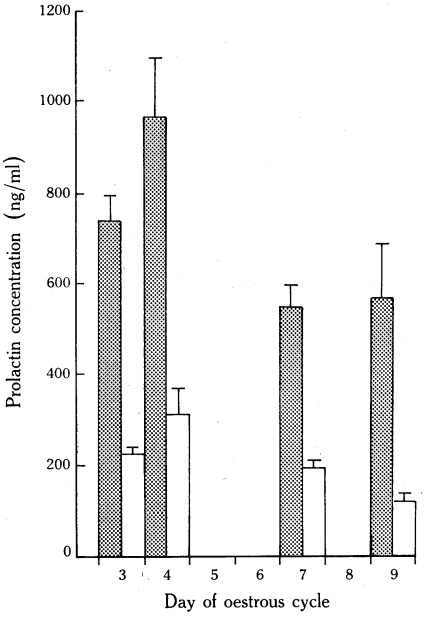

Fig. 1. Effect of heat stress on mean basal prolactin levels in ewes during the oestrous cycle. Group I ewes (shaded histograms) were heat stressed for 8–10 h/day beginning on day 1 of the oestrous cycle. Group II ewes (unshaded histograms) were not heat stressed. Standard errors are shown by vertical bars.

Prolactin levels were consistently ($P < 0{\cdot}05$) higher in the HS group on all days measured (Fig. 1). On the day of gonadotrophin releasing hormone treatment, prolactin levels in the HS ewes were higher during the first few hours of sampling than during the remaining period. In contrast prolactin levels in the NHS ewes remained low. Both groups showed a slight increase after gonadotrophin releasing

hormone injection (Fig. 2*a*) which was probably due to the stress of increased handling, etc. at the time. All ewes in both groups had a large release of prolactin within 15 min of thyrotrophin releasing hormone injection (Fig. 2*a*). Mean prolactin levels of the HS ewes were significantly ($P < 0{\cdot}05$) higher than in the NHS ewes on all occasions except during the 45 min after thyrotrophin releasing hormone injection

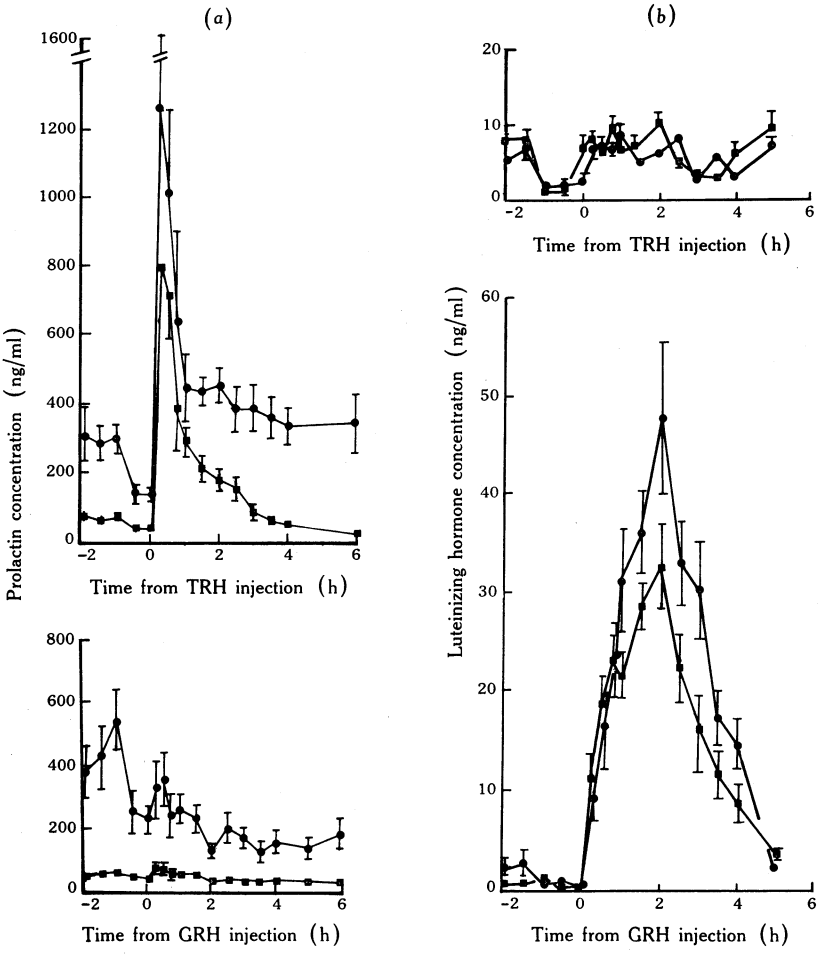

Fig. 2. Effect of 10 μg of thyrotrophin releasing hormone (TRH) and 20 μg gonadotrophin releasing hormone (GRH) injected intravenously on (*a*) mean prolactin levels and (*b*) mean luteinizing hormone levels in HS (●) and NHS (■) ewes. Standard errors are shown by vertical bars.

when the standard error was very high. The area under the curve for the 4-h period after the thyrotrophin releasing hormone injection (i.e. a measure of prolactin response) was significantly ($P < 0{\cdot}05$) greater in HS ewes (2712 ± 405 *v.* 1380 ± 278 area units, mean ± s.e.). However, if baseline prolactin levels were subtracted the areas under the curves were not significantly ($P > 0{\cdot}20$) different [952 ± 255 (HS) *v.* 752 ± 215 area units (NHS), mean ± s.e.].

Luteinizing hormone levels were never significantly ($P > 0{\cdot}20$) different between the two groups and all ewes had a rapid luteinizing hormone release following gonadotrophin releasing hormone injection but not after thyrotrophin releasing hormone injection (Fig. 2*b*).

Discussion

Prolactin concentrations in the ewe were increased by heat stress but prolactin sensitivity to thyrotrophin releasing hormone was not altered. Luteinizing hormone concentrations before and after gonadotrophin releasing hormone were similar in HS and NHS ewes, indicating that heat stress specifically affected prolactin secretion rather than the secretion of all pituitary hormones. Since thyrotrophin releasing hormone is thought to act directly upon the pituitary (Kann *et al.* 1973; Vale *et al.* 1973) the failure of heat stress to affect the pituitary prolactin response to thyrotrophin releasing hormone suggests that heat stress alters prolactin secretion at some site other than the pituitary. This could be at the hypothalamic level although the metabolic clearance rate of prolactin could also be affected. Evidence in the cow suggests that both the secretion rate and the clearance rate of prolactin are affected by heat stress (Smith *et al.* 1977).

The prolactin response to heat suggests that the seasonal variation in prolactin concentrations (Schams 1972; Walton *et al.* 1977) could be due to seasonal changes in ambient temperature. However, this seems unlikely since Ravault and Ortavant (1977) and Hart (1975) have been able to alter prolactin concentrations in sheep and goats by modifying the day length, presumably without changing ambient temperature. These 'seasonal' changes can be induced by changes in lighting regimes and can occur during constant temperature conditions (Ravault and Ortavant 1977). It is unlikely that the rise in prolactin concentration following heat treatment is merely a stress effect since Smith *et al.* (1977) and Mueller *et al.* (1974) found a depression in prolactin concentration during exposure to cold stress.

The physiological role (if any) of this rise in prolactin concentration with temperature is unknown although it may be important for water homeostasis (Lockett and Niall 1965; Horrobin *et al.* 1971; Labella *et al.* 1975).

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