

Adrenal responsiveness in domestic sheep (*Ovis aries*) to acute and chronic stressors as predicted by remote monitoring of cardiac frequency

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The concept of stress and the general adaptive syndrome as advanced by Hans Selye has received considerable attention during the past decade primarily in its interpretation of physiological changes associated with chronic stress. Our work with domestic sheep (*Ovis aries*) habituated to stalls and fitted with halters carrying indwelling electrocardiogram leads and jugular vein cannulas allowed us to remotely test heart rate and blood cortisol responses of these animals to graded stressors. A radioimmunoassay was validated on domestic sheep plasma. We were unable to identify significant alterations of the adrenal response test by sheep exposed to synthetic adrenocorticotrophic hormone after 34 days of chronic stress, suggesting neither adrenal exhaustion nor hypersensitivity. As an indicator of acute stress, we obtained a correlation coefficient of 0.91 between heart rate and blood cortisol, which suggests that heart rate has a strong potential of being a reliable predictor of cortisol values. With a regression equation, the heart rate of observed free-living sheep monitored by telemetry could be used to predict plasma cortisol levels and that, in turn, to predict potential stress-induced changes in animal production, including immunity.

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Le concept, «le stress le syndrome général d'adaptation», avancé par Hans Selye, a fait l'objet d'une grande attention au cours des 10 dernières années, particulièrement sous l'aspect de l'interprétation des changements physiologiques associés à un stress chronique. Nos travaux sur des moutons domestiques (*Ovis aries*) acclimatés à des stalles et munis de licous porteurs d'électrodes ECG (électrocardiogramme) permanentes et de canules intra-jugulaires ont permis d'enregistrer à distance le rythme cardiaque et les changements dans les concentrations sanguines de cortisol en réaction à des facteurs progressifs de stress. Un test radioimmunologique a pu être éprouvé sur le plasma de moutons domestiques. Nous n'avons pu déceler de changements significatifs à la suite des tests sur les réactions adrénériques chez des moutons exposés à une hormone adrénocorticotrope synthétique après 34 jours de stress chronique; il semble donc n'y avoir eu ni épuisement adrénérique, ni hypersensibilité. Comme indicateur de stress grave, nous avons obtenu un coefficient de corrélation de 0,91 entre le rythme cardiaque et le cortisol sanguin; le rythme cardiaque est donc probablement un bon indicateur des concentrations de cortisol. À l'aide d'une équation de régression, le rythme cardiaque chez des moutons libres suivis par télémetrie permet de prédire les concentrations plasmatiques de cortisol et, de là, les fluctuations de production animale causées par le stress, y compris l'immunité.

[Traduit par la revue]

Introduction

Researchers as well as livestock and wildlife managers have been hampered in attempts to measure a stress response of animals exposed to acute and chronic environmental stressors. This is partially the result of a poor understanding of what physiological parameters are altered by stress and, consequently, incomplete knowledge of appropriate parameters to monitor as stress indicators. In addition, most studies have utilized sampling procedures that introduced artificial stress components.

When attempting to evaluate the extent of stress an animal is experiencing, it is important to realize that psychological stressors are as effective as physical stressors in provoking a physiological response (Dantzer and Mormede 1981). For

example, exposure of an animal to a new environment (Dantzer and Mormede 1981) or a "novel stimuli" such as the sight of an unfamiliar object (Stephens 1981) can be as effective as thermal stress in eliciting a stress response.

Conflicting hypotheses exist that attempt to explain the effects of stress on organisms. One of the best known is Selye's (1937, 1971, 1973) nonspecific general adaptive syndrome (GAS). In this scheme, the first stage, called the alarm reaction, occurs when a stressor is initially encountered, and it involves mobilization and buildup of body resources. The second stage is referred to as the resistance stage. Emotional and (or) physical stressors elicit release of corticotropin releasing hormone (CRH) from the hypothalamus, adrenocorticotrophic hormone (ACTH) from the pituitary gland, and glucocorticosteroids from

the adrenal cortex. During this period, glucocorticosteroids tend to abate the effects of traumatic or benign stressors via their antiphlogistic action and control of sugar metabolism (Norris 1980). Continuing exposure to stressors may bring an animal to the final stage, i.e., exhaustion, as characterized by decreased resistance to further perturbations due to exhaustion of most body resources including depressed adrenal glucocorticosteroid production.

It is the exhaustion stage of stress that most researchers attempt to use as an indicator of chronic stress exposure and physiological condition of an animal. However, there is some disagreement as to what actually happens during prolonged stress exposure and how to measure it. For example, Sakellaris and Vernikos-Daniellis (1975) observed not a decreased responsiveness but a markedly elevated responsiveness of the adrenal cortex after presentation of a novel acute stressor to chronically stressed rats. Therefore, although Selye (1950) described this last stage of stress as adrenal "exhaustion" Sakellaris and Vernikos-Daniellis (1975) hypothesized that it actually represents a period of adrenal "sensitization" with elevated glucocorticosteroid release. If chronic exposure to stressors is associated with sustained elevated glucocorticosteroid levels, a pathological condition may result due to impairment of immunodefensive mechanisms (Jensen and Rasmussen 1970; Pappe et al. 1973; Hartmen et al. 1976; Stein et al. 1976).

The adrenal response test (ART), consisting of exogenously administered synthetic ACTH both before and after chronic stress, has been used to assess adrenal cortical activity in cattle (Friend et al. 1977, 1979; Gwazdauskas et al. 1972). One objective of our study was to employ this test on chronically stressed domestic sheep to assess adrenal exhaustion or hypersensitization and to determine if the ART is a reliable method for assessing adaptation to stress in sheep.

As an alternative to monitoring adrenal gland responsiveness to synthetic ACTH, many attempts have been made to directly monitor blood cortisol levels of animals under various stressful conditions. When an animal experiences an acute stressor, cortisol levels in the blood peak within 30 min following the event and then decline (Bassett and Hinks 1969) but with continuous stress, cortisol levels persist at the high levels (Burchfield et al. 1980). Therefore, a determination of blood cortisol levels during acute and chronic stress of free-ranging animals would be an excellent measure of the stress response. However, it is difficult to accurately measure resting or base-line concentrations of blood cortisol because the act of sampling in itself causes sharp elevations of cortisol within 15 min of initial handling for sampling (Bassett and Hinks 1969; Sapolsky 1983).

It was therefore a second objective of our study to expose animals to stressors of varying severity and determine if there is a correlation between the blood cortisol response and an accompanying physiological change, such as heart rate, that can be remotely monitored. Other investigators (Thompson et al. 1968; Candland et al. 1969; Cherkovich and Tatoyan 1973; Moen et al. 1978) have revealed that heart rate is a sensitive physiological indicator of excitement or arousal in birds and mammals. Consequently, if a cortisol - heart rate regression could be established, the blood cortisol level could be predicted from an elevated heart rate response to a stressor perceived by a free-ranging animal fitted with a heart rate telemetry transmitter.

Methods

Five adult mixed-breed range ewes were acclimated to halter

restraints over 20 days and confined separately in adjoining 1.5 × 3 m stalls. Halter ropes were of sufficient length to allow access to *ad libitum* food and water and allow the animals to turn around and lie down. Each animal was anesthetized with xylazine hydrochloride (Rompun®, Bayvet division, Cutter Laboratories, Inc., Shawnee, KS 66201) for surgical placement of jugular cannulas and electrocardiogram (EKG) leads. A 2-mm i.d. Silastic tube cannula was inserted into a jugular vein and coursed underneath the wool where it was held in place by gluing the wool together over it and exteriorizing it at the back of the neck. The use of indwelling cannulas was essential to eliminate the increase in blood cortisol that has been reported to occur as a result of repeated venipuncture (Bassett and Hinks 1969). Three 8-mm disc electrodes were implanted subcutaneously in the chest region at an orientation that provided a clear EKG signal with only minimal muscle artifact. These leads were also passed under the wool and exteriorized in the dorsal neck region. The cannula and three EKG leads were threaded through Tygon tubing which was secured to the halter rope and then passed through a 2.5 cm diameter hole in a 25 cm thick cinder block wall to an adjacent, isolated room. The EKG leads were connected to Grass polygraphs where heart rate, in beats per minute (bpm), was monitored and the cannulas were connected to three-way Teflon stopcocks for blood sampling. EKGs were continuously monitored on all five sheep during trials. In addition, 5-mL blood samples were taken simultaneously on all animals during the testing period by technicians sitting at a table in the isolated room adjacent to the sheep room. During test trials heparinized blood samples were drawn from the cannulas before and at 5, 10, 15, 20, 30, 45, 60, 80, and 100 min after administration of the stressor. An equal volume of sterile physiological saline was replaced into the animals via the jugular cannula; this was followed by volumes of sterile heparinized saline sufficient to backfill the cannulas. Each blood sample was immediately placed on ice and centrifuged under refrigeration, and the plasma frozen for later radioimmunoassay.

The sheep were simultaneously exposed to one of three acute stressors which differed in the extent of their severity. Animals were exposed to the stressor only once during the midmorning of a test day. The first stressor was considered to be mild and consisted of the sudden appearance into the sheep room by one of the researchers who shouted, turned, and left the room within 10 s. After a minimum of 1 day recovery from the mild stressor, a moderate stressor was presented to the sheep; this consisted of yelling and banging on the sheep stalls for 2 min by one of the researchers. The severe stressor was a 5-min exposure to a working sheep dog. The behavioral state at rest and excitement during each trial was filmed and recorded on a closed circuit video camera while simultaneous EKG and blood samples were taken in the adjacent room.

After completion of the acute stress trials sheep were exposed to a chronic stress for 35 days which consisted of short bursts of loud noise occurring at random intervals throughout the day and night. Smoke alarm buzzers were fitted into each sheep stall. An electrical random sequencer was designed and constructed to provide current to the buzzers for 3 s at irregular intervals. Each interval (between 15 and 160 s) was selected at random out of 32 possible choices by the sequence generator. In this manner, the animals would be less prone to habituate to the chronic noise stressor than if it was a rhythmic event. Blood samples for cortisol analysis were taken between 10:00 and 12:00 in the room adjacent to the sheep room via the indwelling jugular cannulas.

Adrenal response tests were conducted before and at the termination of 35 days of stress exposure by injecting 20 IU of ACTH via the jugular cannula and taking blood samples periodically over the next 4 h in a manner similar to that in the acute stress protocol.

Plasma cortisol titer was determined using radioimmunoassay. To assay cortisol, individual 10- μ L aliquots of plasma were added to 12 × 75 mm glass tubes each containing 500 μ L of 0.01 M phosphate-buffered saline (PBS) with 0.1% gelatin (pH 7.0); 3.0 mL of nanograde methylene chloride (Burdick Jackson Laboratories) was added and the tubes were vortex-mixed for 30 s. Aqueous layers (extracted plasma) were removed and the tubes were brought to a volume of 2.5 mL by aspiration. The methylene chloride extracts were placed in a 37°C water bath and brought to dryness using gentle streams of air.

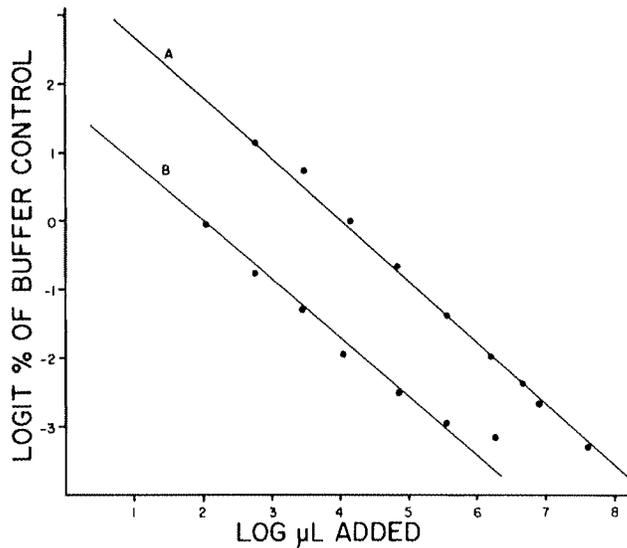


FIG. 1. Logit-transformation of data obtained from the radioimmunoassay of a cortisol dilution series resulted in a line (A) with a slope of -0.95 . Line B (slope -0.87) represents assayed extracted sheep plasma. The x-axis is the natural log of the amount of cortisol added (μL) and the y-axis is the logit transformation of percentage of buffer control binding.

A standard solution of 2000 pg cortisol/100 μL methanol was prepared and 5 mL of this solution was added to 5 mL methanol creating a solution of 1000 pg/100 μL . This doubling dilution series continued resulting in an eight-point standard dilution series from 1000 to 7.8 pg/100 μL methanol. From this series, duplicate 100- μL aliquots were removed and placed into the bottom of 12 \times 75 glass tubes with the methanol in each tube subsequently blown to dryness using gentle streams of air. To the tubes representing the standard curve 500 μL of the methylene chloride extracts of plasma, 500 μL of PBS-gel were added. All tubes were gently vortex-mixed and allowed to stand overnight to allow for resuspension. The following day, 100 μL of [^3H]cortisol was added to each tube and after gentle mixing, 100 μL of cortisol antibody (1:3000) was added to all tubes except those used to determine total [^3H]cortisol added (these contained 100 μL [^3H]cortisol and 1.5 mL PBS-gel) and nonspecific binding (these contained 100 μL [^3H]cortisol and 0.6 mL PBS-gel). Anticortisol antibody (A-155) was obtained from Western Chemical, Fort Collins, CO. The antigen was a conjugate made by linking bovine serum albumin to carbon-3 of the cortisol molecule. This antigen displayed cross reactivities of 1.4% for corticosterone, 1.6% for cortisone, 1.3% for 11-deoxycorticosterone, 1.0% for progesterone, and less than 0.1% for estradiol, estriol, and estrone as tested by Western Chemical. Additional tubes used to determine the amount of [^3H]cortisol bound to antibody without competition from unlabeled cortisol (to be called buffer control tubes) were placed into the assay. All tubes in the assay were gently vortex-mixed and allowed to incubate overnight at 4°C. To remove unbound [^3H]cortisol from that remaining bound, assay tubes were placed in an ice-water slurry and 1 mL of dextran-coated charcoal (0.02 g Dextran (70 000 MW) was added to 100 mL PBS (without gel)) then 0.25 g acid-washed, neutralized charcoal were added. After gentle vortex-mixing, the tubes were incubated for 15 min; the charcoal was pelleted by centrifugation (1500 \times g, 10 min, 4°C). The supernatant fraction from each tube was poured into individual miniscintillation vials, scintillant was added, and the amount of ^3H activity in each sample was determined by liquid scintillation spectrometry. The amount of cortisol present in the samples was determined by computer program using logit-transformed data.

The cortisol radioimmunoassay (RIA) used in these experiments was validated using two procedures, parallelism and quantitative recovery. To determine parallelism, a seven-point, two-fold dilution series from 125 to 1.95 μL of domestic sheep plasma in PBS-gel was constructed in triplicate. A second dilution series similar to the first was constructed

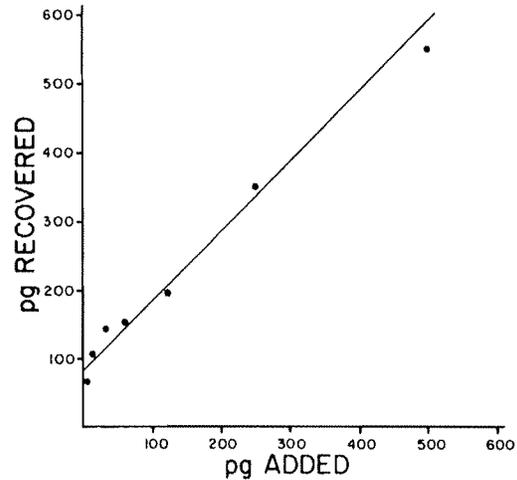


FIG. 2. When increasing concentrations of exogenous cortisol were added to a constant volume of sheep plasma (from a pooled sample) and then subjected to heating, extraction, and radioimmunoassay, a line with a slope of 0.95 with a correlation coefficient of $r = 0.99$ was generated. The x-axis is the amount of cortisol added (pg) at each point while the y-axis is the amount assayed (pg) at each point.

using a cortisol standard of 1000 pg cortisol/500 μL PBS-gel in triplicate. Following construction, both dilution series (plasma and standard) were extracted and assayed as previously explained. To analyze these data, the amount of binding at each point was expressed as a percentage of buffer control binding, then logit transformed. Logit percentage of buffer control binding for the lines generated by assaying diluted domestic sheep plasma and the standard solution of cortisol were plotted against the natural log of the amount (μL) added.

A quantitative recovery procedure was used to establish the accuracy and limits of the radioimmunoassay. A seven-point, two-fold dilution series from 500 to 7.8 pg/mL of cortisol in methanol with four replicates at each point was constructed. The methanol in each tube was removed with gentle streams of air; the cortisol was resuspended in 490 μL of PBS-gel. To each tube, 10 μL of domestic sheep plasma was added; the tubes were gently vortex-mixed and allowed to resuspend overnight. Cortisol was extracted and assayed as outlined in the general procedures section. Following computer determination of the amount of cortisol assayed per millilitre, a plot of picograms cortisol added versus picograms cortisol assayed was made and regression statistics were applied to the line so generated.

Regression lines were calculated by the method of least squares. Differences between means were examined using a repeated measures design (Nie et al. 1975) followed by a post-hoc correlated paired *t*-test of samples taken at pre-stress and at various post-stress sampling intervals as well as analysis between peak responses for the various stress intensities (Zar 1984; Nie et al. 1975).

Results

The parallelism experiment for validation of the cortisol RIA demonstrated that when a serial dilution of domestic sheep plasma was extracted and assayed, the resulting line was parallel to the line generated by the assay of a standard solution of cortisol. The slope of the line generated by assaying sheep plasma was -0.87 while that for the standard solution was -0.95 (Fig. 1). Exogenous cortisol added to domestic sheep plasma before extraction and assay was quantitatively recoverable. Regression analysis of picograms cortisol added versus picograms cortisol assayed resulted in a line with a slope of 0.95 and a correlation coefficient of 0.99, indicating that the extraction would recover known amounts of cortisol added to samples and the radioimmunoassay procedures would accurately measure the amount of added cortisol (Fig. 2). The intra-assay variance for plasma cortisol was 5.3% with inter-assay variance of 10.1%.

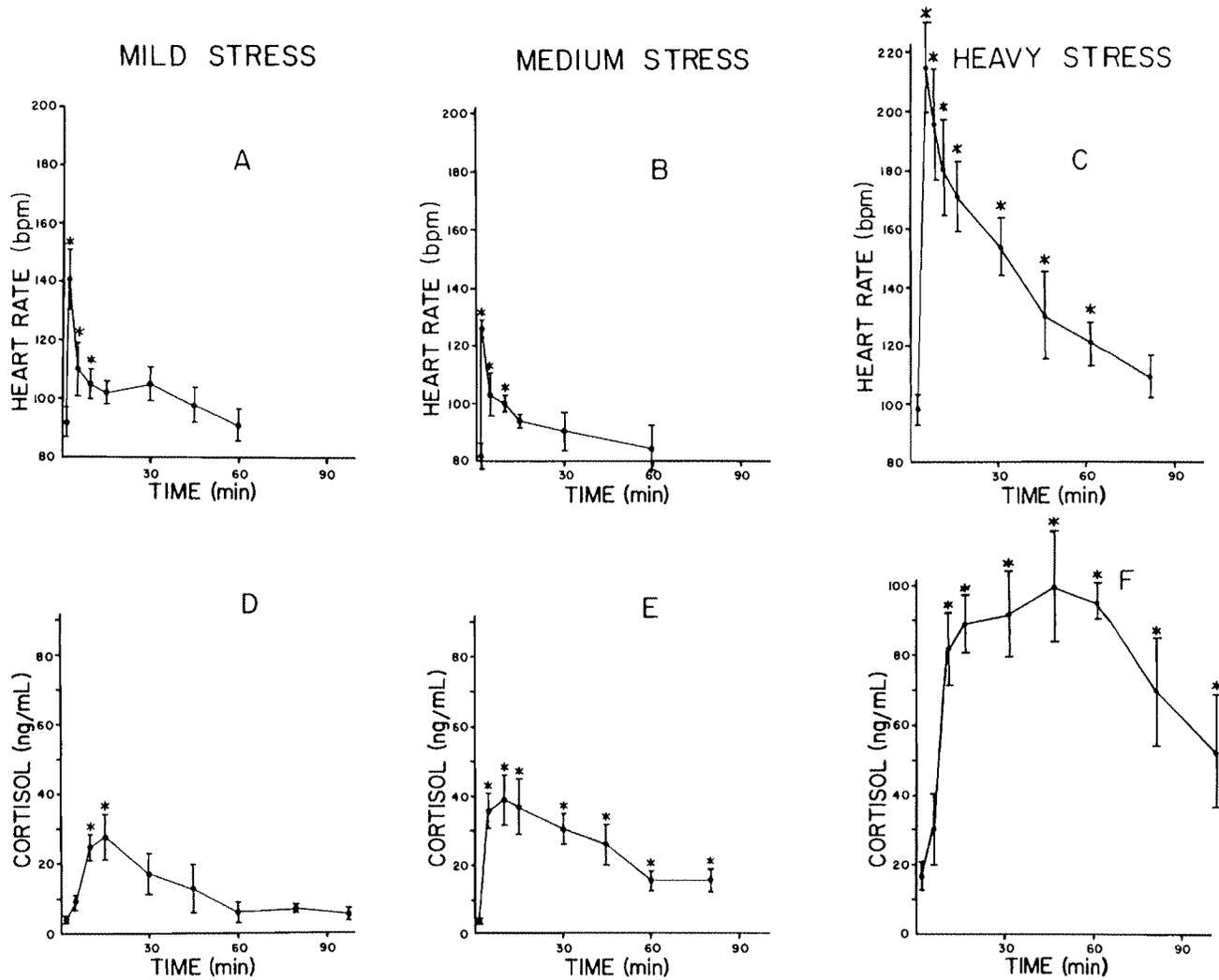


FIG. 3. Heart rate (bpm) and plasma cortisol levels (ng/mL) of sheep after exposure to acute mild ($n = 5$), medium ($n = 4$), and heavy ($n = 4$) stressors. Vertical lines represent \pm SEM. Asterisks identify mean values that are significantly elevated from pre-stress levels.

In all cases, the resting heart rate (90 ± 9.5 bpm) was significantly ($P < 0.001$) elevated immediately after exposure to the acute stressors. There was no significant difference between peak elevated heart rate caused by mild and medium stress. However, heart rate during mild stress returned to resting values by 10 min post-stress, while during medium stress, it remained significantly ($P < 0.05$) elevated until 20 min post-stress (Figs. 3A, 3B). The heart rate induced by the severe stressor was significantly ($P < 0.05$) higher than that induced by mild or medium stress and remained ($P < 0.05$) elevated from resting values for 60 min (Fig. 3C).

Plasma cortisol levels were significantly elevated ($P < 0.001$) in all cases above resting values (9 ± 2.3 ng/mL) within minutes post-stress. Cortisol levels returned to pre-stress levels 30 min after removal of the mild stressor (Fig. 3D) as contrasted to continuously elevated cortisol levels through 90 min (and as long as 180 min) of post-stress monitoring for both the medium and severe stressors (Figs. 3E, 3F). Cortisol values induced by severe stress were significantly ($P < 0.05$) higher than those induced by medium and mild stress (Figs. 3D, 3E, 3F). There was a good correlation ($r = 0.91$) between heart rate and plasma cortisol concentrations when peak heart rate and peak cortisol concentrations for all sheep exposed to acute stressors were plotted (Fig. 4).

During chronic stress, plasma cortisol levels of sheep were significantly ($P < 0.05$) elevated on the 5th day and remained elevated until day 24 when the random noise generator failed. Once the noise generator was operational (2-day delay), plasma cortisol levels again increased to previous chronic stress values (Fig. 5).

Administration of ACTH resulted in a significant elevation of plasma cortisol within 15 min after intravenous infusion via the jugular cannula. There was no significant difference in the ART before the onset of stress and at the termination of 35 days of chronic stress (Fig. 6).

Discussion

The interrelationship of the pituitary-adrenal axis in the stress response still remains uncertain. Selye (1950) suggested that an exhaustion of the adrenal cortex occurs during prolonged stress exposure but others believe that adrenal sensitivity to acute stress declines following prolonged exposure to chronic stress (McNulty and Thurley 1973; Ader 1975). Alternatively, Sapolsky's (1983) work on baboons suggested that chronic stress may influence a decline in cortisol production, not via exhaustion of the adrenal cortex but by impairment of pituitary ACTH production. On the other hand, Friend et al. (1977, 1979) and others (Paul et al. 1971; Barrett and Stockham 1963)

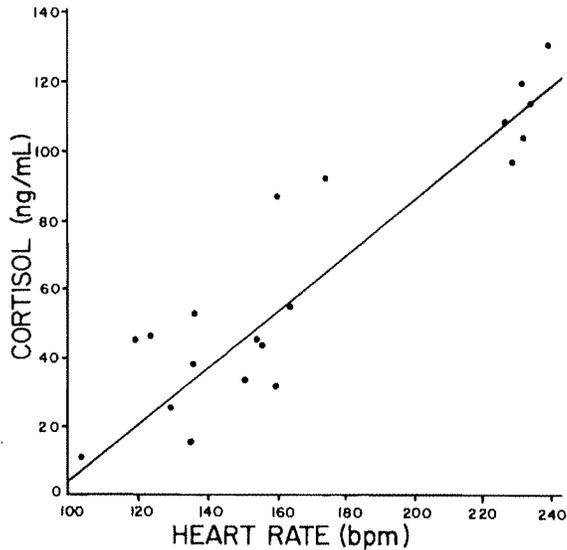


FIG. 4. Regression of peak heart rate (bpm) and peak plasma cortisol (ng/mL) levels for all sheep during exposure to mild, medium, and heavy stressors.

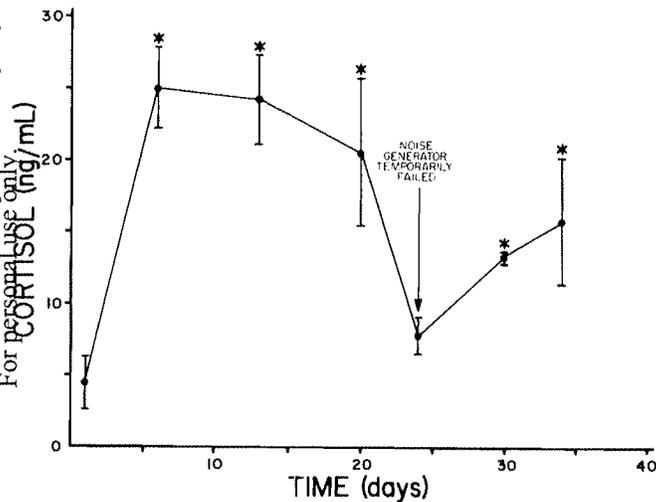


FIG. 5. Plasma cortisol levels (ng/mL) of sheep chronically stressed for 24 days (auditory stimuli at random intervals) followed by 2 days without stress (days 25–27) and 8 additional days exposure to chronic auditory stressor. Vertical lines represent \pm SEM. Asterisks identify mean values that are significantly elevated from pre-stress levels.

present data that support the opposite view, that stress tends to increase adrenal sensitivity to an acute stressor. However, our study presents no evidence of exhaustion or impairment of either pituitary or adrenal control of cortisol release in sheep. For example, cortisol levels were consistently elevated during chronic stress and rebounded to similar elevated levels when the stressor was temporarily removed and then reinstated. In addition, the ART response to ACTH administration on sheep before and after 35 days of stress was virtually unaltered, thereby not supporting the concept of adrenal exhaustion or hypersensitization.

It has often been reported in studies on chronic stress that animals habituate to the stressor as reflected by diminished blood cortisol levels (Mason et al. 1957). In our study, cortisol concentrations did not decrease with continuous daily exposure to stressor, perhaps because of the irregular unpredictable

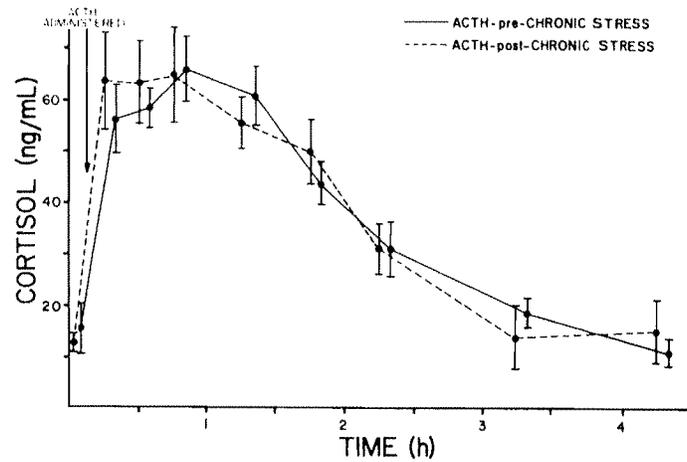


FIG. 6. Average cortisol values (ng/mL) of four sheep after receiving ACTH via jugular cannulas on three occasions before chronic stress exposure and average plasma cortisol (ng/mL) of the same four sheep receiving ACTH via jugular cannulas after 35 days of chronic auditory stress exposure. Vertical bars represent \pm SEM.

interval of the noise stimuli. Chronically elevated blood cortisol could have an important effect on the efficiency of animal production as well as a potential pathologic effect on the immune system. For example, high plasma cortisol levels can enhance protein catabolism by increasing mobilization of amino acids for gluconeogenesis (Burke 1973; Baird and Heitzmann 1970). This, in turn, may decrease weight gain and be a significant problem for animals exposed to a continuous set of stressors such as an unfamiliar arena, overcrowding, and transport (Van Mourik and Stelmasiak 1984; Van Mourik et al. 1985).

In contrast to Selye's concept of glucocorticoid-enhanced resistance to stress, it is becoming more evident that stress-elevated glucocorticoids suppress the synthesis, secretion, and action of intracellular mediators such as established hormones, prostaglandins and other arachidonic metabolites, certain secreted neutral proteinases, lymphokines, and a variety of bioactive peptides (see Munch et al. (1984) for review). For example, elevated cortisol tends to decrease circulating lymphocytes and cause degeneration of lymph and thymic tissue as well as decreasing antibody production, thereby inhibiting the body's ability to resist disease (Jansen and Rasmussen 1970; Paape et al. 1973; Hartman et al. 1976; Stein et al. 1976). As recently suggested for cattle (Blecha and Baker 1986), plasma cortisol may impair interleukin 2 production and thereby decrease immune competency. Cortisol also acts to suppress the exudative phase of inflammation by inhibiting the movement of white blood cells and plasma proteins to the site where microorganisms and their products accumulate (Fruhman 1962; Ivanov 1960). Therefore, chronic exposure of an animal to stressors and elevated cortisol levels could produce an immunosuppression as assessed by immunologic reactivity (Huber and Douglas 1971; Revillard 1971; Roth 1984). It is believed by Munch et al. (1984) that the elevated glucocorticoids function to prevent the immune system from overreacting during stress exposure thereby preventing the generation of autoimmunity.

Previous attempts to assess a reaction to stress in animals by associating behavioral changes with fluctuations in blood cortisol levels have met with problems of altered behavior resulting from the sampling procedure (Bassett and Hinks

1969). In addition, even though overt changes in behavior have traditionally been used to assess the extent of stress in various species of mammals, it had been established that physiological parameters such as cardiac response to disturbing visual or auditory stimuli can be elicited in the absence of any behavioral changes (Thompson et al. 1968; Cherkovbick and Tatoyan 1973; Moen et al. 1978). In our study using indwelling cannulas and EKG leads on animals habituated to their environment, it was possible to remain independent of behavioral changes and rely upon a correlation between heart rate and blood cortisol levels as the animals responded to a series of graded stressors. Comparison of heart rate responses during mild and heavy stress reflects the significant changes in peak cortisol levels occurring during these states. But the differences in peak cortisol elicited by a mild and medium stressor were not directly reflected by the heart rate change. The lack of difference in heart rate response between these two conditions may be because the degree of severity between our mild and medium stressors was relatively small and may not have been sufficient to elicit a differential peak heart rate response under these specific conditions. However, the fact that the heart rate remained elevated longer in response to the medium compared with the mild stressor supports our claim that there is a correlation between different stressors and the heart rate – blood cortisol response. This is evident by the high correlation coefficient (0.91) of the regression line for the heart rate and blood cortisol values representing responses of all animals exposed to the three different stressors.

Studies of free-living birds (Kanwisher et al. 1978) and ungulates (Ward et al. 1976; MacArthur et al. 1979; Stemp 1982) have revealed that telemetered heart rate is a sensitive indicator of arousal (Jenkins and Kruger 1975). Through our correlation between heart rate and changes in blood cortisol, a predicted value of cortisol can now be assigned to these aroused states for sheep. There also are many aspects of an animal's physiology and, consequently, its productivity and state of health that can be predicted as a result of knowing cortisol changes related to heart rate changes. Through a cardiac-related cortisol increase, the extent of protein breakdown due to cortisol-induced gluconeogenesis could be estimated (Burke 1973; Baird and Heitzmann 1970; Van Mourik and Stelmasiak 1984). This is critical information to managers trying to maximize growth and productivity of animals while at the same time exposing them to changing environments, minimal space restrictions, and high densities (Van Mourik et al. 1985; Van Mourik and Stelmasiak 1984). In addition, impaired immune competency of an animal may potentially be predicted under stressful conditions based upon a heart rate signal representing blood cortisol changes. For example, heart rate telemetry can demonstrate if an animal perceived any combination of heavy, medium, or mild stressors during a day, and could be used to predict if the animal sustained chronically elevated blood cortisol which might influence its immune competency (Hudson 1973).

Although there are variables not included in this system, such as the relationship between heart rate and nonstress induced changes in metabolism (e.g., running or eating) we feel that heart rate telemetry may provide a new dimension in assessing the response of animals to environmental stressors.

It was the purpose of a subsequent study on bighorn sheep (Harlow et al. 1987) to determine if the promising results from this study on domestic sheep could also be obtained on a wild species of sheep.

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