COW PERFORMANCE, ADRENAL FUNCTION, AND MILK QUALITY UNDER VARYING LEVELS OF COMPETITION

BY

Theodore Henry Friend

Dissertation submitted to the Graduate Faculty of the
Virginia Polytechnic Institute and State University
in partial fulfillment of the requirements for the degree of
DOCTOR OF PHILOSOPHY
in
Dairy Science

APPROVED:

Dr. C. E. Polan, Chairman

Dr. W. M. Etgen

Dr. P. B. Siegel

Dr. R. G. Cragle

Dr. D. E. Kenyon

February, 1977
Blacksburg, Virginia

ACKNOWLEDGMENTS

The author wishes to express his gratitude to his friend and advisor DR. C. E. Polan for making this dissertation possible.

Assistance and advice provided by the other committee members is also greatly appreciated: Dr. P. B. Siegel for advice on the behavioral aspects of this dissertation; Dr. W. M. Etgen for his expertise in the farm management area; Dr. D. E. Kenyon for service on the committee and making agricultural marketing a stimulating side light during the author's stay at VPI; and Dr. R. G. Cragle for service as a committee member and for creating an excellent atmosphere in the Dairy Science Department in which to learn.

Special thanks go to Dr. M. L. McGilliard and Dr. W. E. Vinson for invaluable advice. The author also expresses his appreciation to the excellent graduate students of the Dairy Science Department whom have been a most important part of his life during the last 6 years.

Finally, but most important, the author wishes to express his love and appreciation to his parents who made all this possible.

TABLE OF CONTENTS

TABLE OF CONTENTS (Continued)

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

The behavior that is manifested in the individual is a result of both endowment from the parents (prepartum) and environmental influences (53). Numerous researchers have ascribed behavioral differences to genetics (35, 46, 64). Nongenetic variations of the parent endowment (eg. stressing the mother) can also effect the behavior of genetically indentical offspring even when reared in the same environment (52). Environmental influences can likewise be illustrated by taking animals that are genetically the same and raising them in different environments (46).

For centuries, man has been selecting cattle that have behavioral traits favorable for domestication. As summarized by Hale (41), these traits include: a large social group (herd) with a hierarchial social structure; promiscuous matings with males dominant over females; precocial young; little disturbed by man or changes in environmental conditions; and general dietary habits.

The traditional dairy cow environment of pasture grazing and stanchion barn housing has given way in recent years to a totally confined group environment: group feeding and free stall housing. Simultaneously the trend has been to assemble larger groups of cows. Managing cows by groups may be more efficient but results in less attention to the individual cow. Therefore, possible effects social behavior may have on the individual has greatly increased. However, little is known about how cows are adjusting behaviorally to this radically different environment.

Scientific information on behavior could be of great economic value to the industry (11), especially in determining optimum stocking rates and minimizing behavioral stress. Resting, feeding, and exercise space requirements and the role that the social hierarchy may play in determining access to these is yet to be determined. Very little is also known about stresses resulting from managing cows by groups. This critical lack of knowledge prompted the following studies.

LITERATURE REVIEW

Behavior of Dairy Cattle in Confinement

Free Stall Behavior

The average amount of time cows spend resting in free stalls appears to be relatively constant, 10.7 hours for 15 cows in 20 stalls (76) and 11.1 hours per day for 21 cows in 20 stalls (27). Cows make maximum use of free stalls between 3 a.m. and 7 a.m. (76), and 1 a.m. to 5 a.m. (58). There is a preference to use certain stalls (27, 39, 76) and social rank appears to effect which stall a cow occupies (27). A cow's successor at a given free stall as well as cows occupying adjacent stalls tended to be of similar social rank, r = .42 and .53, respectively (27).

Recommendations for number of free stalls vary. There also appears to be a discrepancy between current spatial recommendations and practice in the field. One free stall per cow plus up to 10% additional is now recommended (5, 61) while some dairymen are exceeding recommendations by 30% without apparent adverse effect.

Feed Trough Behavior

Dairy cattle are grazing animals. In their natural environment, they do not typically get much feed at any one time nor for any one small cycle of behavior when compared to a carnivore (12). This natural ingestive pattern has been changed in management systems where a cow's total daily nutritional requirements are placed before her in a feed bunk.

Factors controlling intake in ruminants can be broken down into: 1) physical-rate of disappearance of digesta; 2) chemostatic or physiological mechanisms; 3) sensory stimuli such as taste, smell, and; 4) possible psychological factors. The first three are discussed in a review by Jones (50). Possible psychological factors are based on the theory of social facilitation or the presence of other con-specifics causing an increase in feeding activity (40). It has been shown in dairy cattle that group fed cows will consume more total feed then when fed individually in stanchions (19, 49, 59). One investigator (49) attributed the increase to competition while the others favored increased maintenance requirements due to the general increased activity of freedom of movement as the cause.

The amount of time individuals average at the feed truogh has been determined and fluctuates within a narrow range even with different types of forage. The average time a cow spent eating in four studies was 5.2 (58), 4.9 (14), ~4 (76), and 3.7 hours per day (27). A sizeable portion of this variation is probably due to different criterion among the investigators for what constituted eating.

Schein and Fohrman (74) commented that, "There is little doubt that lower order animals would suffer markedly if she were wholly dependent on trough feeding." Apparently animals higher in the social order ate more under group feeding conditions by chasing lower order animals away from the feed. Less dominant cows expend more effort getting feed from a trough (56). McPhee, McBride, and James (60) found that high social strata steers spent more time feeding (611 ± 19.5 vs. 546 ± 1.6 minutes) during a 60-hour period. Lower strata animals ate proportionately more at night when they were less disturbed. Friend and Polan (27) found dominant cows eating when hay, fresh silage, and supplemental concentrate were fed, $r = .40$, .55, and .57, respectively. The above studies indicate that social rank is important in determining how much access a cow will have to feed. They failed however, to measure individual intakes to determine how efficiently cattle use their time at the feed trough.

A view of dominance is that there is one basic social order through which all of a group's resources are regulated (87). Since production in the lactating dairy cow is greatly influenced by nutritional status, a high correlation with milk production would be expected if the social order influenced feed intake. Social orders, derived from measures of agonistic behavior, have been correlated with body weight and/or age but not milk production (2, 8, 17, 22, 23, 27, 34, 74). A possible reason for this lack of association is that access was not limited enough for social dominance to have an effect on intake.

Recommendations for the amount of feed trough space vary greatly, ranging from 15 to 30 inches per cow when feed is continuously available (5, 61). These recommendations have not been based on experimental data, but on custom and successful experience in handling cows. Many farmers are, however, allowing much less feed trough space per cow without apparent adverse effects.

Exercise Lot

The amount of space animals have to interact in can be extremely inportant. Southwick (85) defines density as the number of individuals per unit area or unit space. A simple physical measurement. Crowding, contrastingly, is a product of density, communication, contact and activity. It implies a pressure, a force, and a psychological reaction. It may occur at widely different densities which relate to the tolerance of individuals and of groups to crowding.

Craig, Biswas and Guhl (20) observed that strangeness and crowding in chickens were associated with higher frequencies of social interaction than found in socially undisturbed and uncrowded flocks. Results suggested, however, that individual rates of social interaction were reduced by crowding. Broilers could be reared with similar results at either 464, 696, 929 or 1150 cm^2 of floor space with adequate controls of other environmental factors (81). At the time of that work, 929 cm^2 was standard for many farms. Egg production was depressed with 929 or 1236 cm^2 per bird compared to 1858, 2780, or 3716 cm^2 (26, 82). Adrenal hypertrophy, indicative of milk stress was observed in the 1236 cm^2 groups (82). The 929 cm^2 group adrenals were not examined.

The amount of lot space required per cow is not known. However, Arave (3) noted that restricting cows to a lot size of 2.3 m^2 per cow was actually beneficial. There was less activity, fewer encounters with herd mates, no discernible effect on milk yield, and significantly lower leucocytes than the same cows in a 9.3 m^2 per cow sized lot.

Syme et al. (88) examined behavior of cattle in a lot using a grid system with photography similar to that developed at V.P.I and described in this dissertation. Dropping the most dominant of the seven cows used in the study because she did not conform, high ranking cows moved further (entered more grids) than the low ranking cows (r = .98). Dominant cows also maintained less interanimal distance and there was no noticable tendency for mutual repulsion between higher ranking animals.

Inter-group Movement of Cows

Many dairymen are grouping their cows by production or stage of lactation if compatable with their physical facilities. A separate ration is then formulated for each group based on production. Most grouping schemes require the shifting of individuals from one group to the next as production, breeding status, etc. changes. When a cow is placed in a new group, is she temporarily under stress ? Farmers and researchers (2, 10, 75) have reported a decrease in milk production after regrouping cows. Shifting cows along with dietary changes has caused sharp but temporary reductions in milk production (1, 62). Researchers in two studies (2, 10) however, have observed a 5% decrease in milk production the day after shifting using the same ration indicating the cause of the decrease was behavioral rather than nutritional. Brakel and Leis (10) observed that agonistic encounters increased almost three fold during day 1 after 4 new individuals were introduced to a group of 13. Numbers of encounters as well as production returned to normal levels from day 2 on.

Conclusions

Some descriptive behavioral data has been published on dairy cattle. The average time cows desire to spend in free stalls is relatively constant with cows showing individual preferences for certain stalls. Time cows spend at the feed trough similarly flutuates within a narrow range, with social rank possibly playing a role during highly competitive periods. Exercise space requirements are not known, though there are some indications that reduced space may inhibit aggression. The momentary decrease in milk production observed when cows are moved to a new group is largly due to behavioral stress, not change in diet.

Quantitative data on the behavior of dairy cattle under confinement conditions is lacking. Recommendations for stocking rates of free stalls and feed trough space are not based on experimental data and do not coincide with what some farmers are successfully doing.

Stress in Dairy Cattle

Adrenal Function

Stress is the non-specific response of the body to any demand made upon it (79). The normal response to a wide variety of physical and psychological stressors is the excitation of the hypothalamus. The excitation is transmitted through an adrenocorticotropic hormone (ACTH) releasing factor to the hypophysis, which released ACTH that stimulates the cortical position of the adrenal to produce adrenocorticosteroid hormones. Under chronic stress, hyperplasia and hypertrophy of the a adrenal gland can occur.

In cattle and sheep, corticosteroid concentrations in the blood increased following stressors such as change in environment (71), heat and cold (16, 57, 68, 70, 72), disease (24, 73), the milking stimulus per se (83, 94), extroceptive stimuli (83), forced exercise (4), and at parturition (48).

The adrenal corticoids, mainly cortisol in the cow (93), elicit physiological adjustments enabling the animal to tolerate stress (18). These stimulate many changes in the body such as atrophy of the thymus and lymph glands, inhibition of inflammatory reactions, reduced ability to produce antibody (89), increased resistance to bacteria (30) and greatly influence glucose and general organic metabolism.

Basal corticoid concentrations vary greatly (47, 48, 95). Gwazdauskas (36) found no difference between basal corticoid concentration for heat stressed and control cows. However, corticoid concentrations in response to injection of 200 IU ACTH were different ($P<.10$).

Corticoid output in response to exogenous ACTH provides a measure of the animal's ability to withstand stress (84). Stress-susceptible pigs, unable to maintain homeostasis on exposure to stress, showed a lack of response to exogenous ACTH injection when compared to stress resistant pigs (77). Gwazdauskas et al. (37) documented the effects of an intravenous injection of ACTH on peripheral plasma concentrations of progesterone, cortisol, and corticosterone in lactating dairy cows. Cortisol levels increased greatly, with corticosterone to a lesser extent.

Shayanfar et al. (80) found the mean response to 200 IU ACTH to be negatively influenced by milk production, stage of lactation, age, and temperature.

The effect of forage source on susceptibility to stress in dairy cattle is attracting more attention. It has been suggested that animals fed corn silage as sole source of forage may be less able to tolerate stresses associated with parturition, initiation of lactation, and onset of high milk production (91). However, the balancing of the rations in that work is being questioned. Other workers have reported no adverse effects of the corn silage diet (90, 92) other than increased ketosis (90).

Recently Smith et al. (84) made a study to assess susceptibility to stress in dairy cattle consuming diets in which silages provided the only source of forage by monitoring the plasma corticoid response to exogenous ACTH during early lactation. Each group of 3 dairy cattle were fed large amounts of concentrate and one of the following forage diets: alfalfa-timothy hay plus corn silage; alfalfa-timothy haycrop silage plus corn silage; or corn silage. Plasma corticoid response was 81% greater for the hay and 36% greater for the haycrop silage than that for the corn silage diet. The factors related to the cause of the muted response to ACTH in the animals fed corn sialge remains to be determined.

It is widely accepted that stress, be it psychological or physical, has an effect on the hypothalamic-pituitary-adrenal axis. The hypophyseal response to exogenous ACTH can be used to determine if an animal is under stress. The next question is, what are the effects of elevated glucocorticoids in dairy cattle.

Role of Glucocorticoids

The pattern of cortisol action in the monogastric might be viewed as promoting the conversion of protein to carbohydrate and the storage of carbohydrate in the form of glycogen (13). Skeletal muscle protein is depleted, uptake of amino acids from the plasma by the liver is stimulated and increased liver enzymes prepare amino acids for incorporation into carbohydrates. Cortisol appears to stimulate glycogen synthesis by increasing the conversion of pyruvate to glycogen and inhibiting release of glucose from hepatic cells. Cortisol also promotes the mobilization of fatty acids from peripheral adipose tissues.

Responses of ruminants to pharmacological doses of glucocorticoids include increases in gluconeogenesis, urea synthesis, and blood glucose (6, 54). The role that glucocorticoids play in ruminant metabolism at physiological concentrations appears to be slight. In an extensive study using adrenalectomized lactating ewes, Ely and Baldwin (25) concluded that: 1) Mammary metabolism and milk biosynthesis in ruminants are not strongly dependent on glucocorticoid; 2) Liver gluconeogenesis in ruminants appears to be regulated in part by glucocorticoid(s) but less prominently than in non-ruminants and; 3) Glucocorticoids play a less significant role in the regulation of liver and mammary enzymes and metabolism in sheep than in rats. Ely attributed this relatively minor effect of glucocorticoids in ruminants to their normal dependency upon a high rate of gluconeogenesis.

Head et al. (42) fed 10 ug of flumethesone (a synthetic glucocorticoid) per day to 13 cows from 4 to 44-weeks in lactation and found no effect on milk yield and other parameters studied. Contrastingly, Swanson and Lind (86) found that 5 and 10 ug of flumethasone per day increased milk yield, solids not fat, and fat compared to when 0 or 20 ug were administered. The discrepancy may be that cows in the former study had an average peak daily production of ~22 kg, and the latter ~35 kg.

In summary, the classic stress response via the hypothalamic-pituitary-adrenal axis is present in dairy cattle. Measuring blood corticoids in response to exogenous ACTH is an acceptable means of measuring the existance of a stress under certain conditions. However, the metabolic effects of elevated glucocorticoids may be much less than in monogastrics due to the high rate of gluconeogenesis usually occuring in the ruminant. A milk production response to glucocorticoids has been demonstrated but only in higher producing cows.

Milk Somatic Cell Count

One possible manifestation of stress in lactating cattle is increased milk somatic cell counts (MSCC). Stressing conditions and increased corticoids have been associated with a blood neutrophylic leucocytosis in dairy cows (29, 67, 96, 97, 98). There is controversy, however, over whether stress can cause elevated MSCC independent of infection.

Corticoids are anti-inflamatory agents and would be expected to inhibit migration of leukocytes to tissues. Paape et al. (66) found no change in milk leukocytes after 3 days of cortisol and then 3 days after ACTH injections. In a subsequent study, Paape et al. (65) supported the inability of ACTH or corticoids to provide increases in leukocyte concentrations of milk from non-infected cows. Similarly, Arave (2) concluded that neither rank nor exchanging cows between groups has a significant effect on leukocyte concentration in milk. One would conclude from these investigations that corticoids have no effect on MSCC.

Complicating the issue, however, Whittlestone et al. (98) found a marked increase in leucocytes in cows with a previous mastitis history following isolation and being chased by a dog. Very recently, Wegner et al. (96) found a highly significant correlation of .86 between blood leucocytes and mean somatic cell count in mastitis-free cows after injection of 250 IU ACTH. A positive correlation also existed between blood neutrophils and somatic cell count in environmental heat stressed cows with no evidence of current mastitis. Mean somatic cell counts in cows with low level mastitis did not change. These investigatiors conclude that adrenal function or corticoids can have an effect of MSCC.

More work is needed in this area to settle the controversy by further elucidating the relationship of corticoids and somatic cell counts. It is important to know if high corticoids or stress can elevate a cow's somatic cell count independent of infection in the gland.

Conclusions

One way dairy cattle respond to stress is through the classic hypothalamic-pituitary-adrenal mechanism. Plasma corticoid concentrations in response to exogenous ACTH has been an effective measure of long term stress. The role that glucocorticoids play in ruminant metabolism, however, appears to be slight due to the ruminant's normal dependency on a high rate of gluconeogenesis. Glucocorticoids are anti-inflamatory agents and would be expected to inhibit migration of leukocytes to tissues. There is controversy however, on the effect of stress and elevated glucocorticoids on milk somatic cell counts.

OBJECTIVES

The review of literature has shown a critical lack of information on cow behavior under varying levels of competition for resources. Current spatial recommendations are not based on research data nor do they reflect practices that appear successful. The advantage cows high in the social hierarchy are thought to have in gaining access to resources is not supported by production data. The objectives of Part I of this dissertation were to determine the effect of varying levels of competition on resting, feeding, social behavior, and individual feed intake in dairy cattle. From these data, spatial requirements can be estimated and the actual role of a cow's dominance determined.

Introducing cows into a new group can result in considerable stress as observed by temporary decreases in milk production and increased agonistic behavior. The influence this stress has on the physiology of dairy cattle must be determined so that corrective recommendations can be made. A means of objectively determining when cattle are under stress would also be of great value. Part II was designed to elucidate the influence of introducing cows into a new group on adrenal function, milk somatic cells and milk production. The use of adrenal response to exogenous adrenocorticotrophic hormone as a measure of stress was also to be evaluated.

PART I: COW PERFORMANCE UNDER VARYING LEVELS OF COMPETITION

Procedure

Experiment I

This experiment examines the effect of varying levels of competition on resting, feeding, and social behavior of dairy cattle.

The herd and husbandry. Twelve Holstein cows from the University herd were selected to represent a cross section of a typical dairy herd in age (4-three, 4-four, 1-five, 2-six, and 1-eight year old) and milk production (7,700 to 10,000 kg per year mature equivalent production). Twelve were chosen because this number seemed necessary to acheive a typical social organization meanwhile facilitating identification and accountability of all cows. Cows ranged from 23 to 79 days in lactation when the experiment commenced. All cows had been dehorned as calves.

The herd was housed in a facility (Fig. 1) designed and built specifically for this type of research, and remained there continuously except when removed for milking twice daily (2.6 hours per day). Cows had continuous access to a complete mixed ration fed twice daily. The ration was formulated to meet the recommended allowances for the mean milk production plus one standard deviation. Group intake was determined as feed offered minus refusals.

Experimental design. On March 14, 1974, after a 4-week adaptation, the free stall treatments commenced. The two most southwestern free stalls were blocked by nailing three planks across the entrance. Subsequently, pairs of free stalls were blocked alternating from each end of the row on March 22, 31, and April 10, when only the middle 4 stalls remained available. All free stalls were then opened on April 13. The last treatment (4 free stalls) was terminated after only 3 days due to the obvious discomfort of the animals.

On April 19, the feed trough treatments commenced with the reduction of the 6.1 m trough to 4.88 m. The feed trough was further reduced to 3.66, 2.44, and 1.22 m on April 26, May 8, and May 17. On May 31, the available feed trough space was restored to the original 6.1 m. Feed

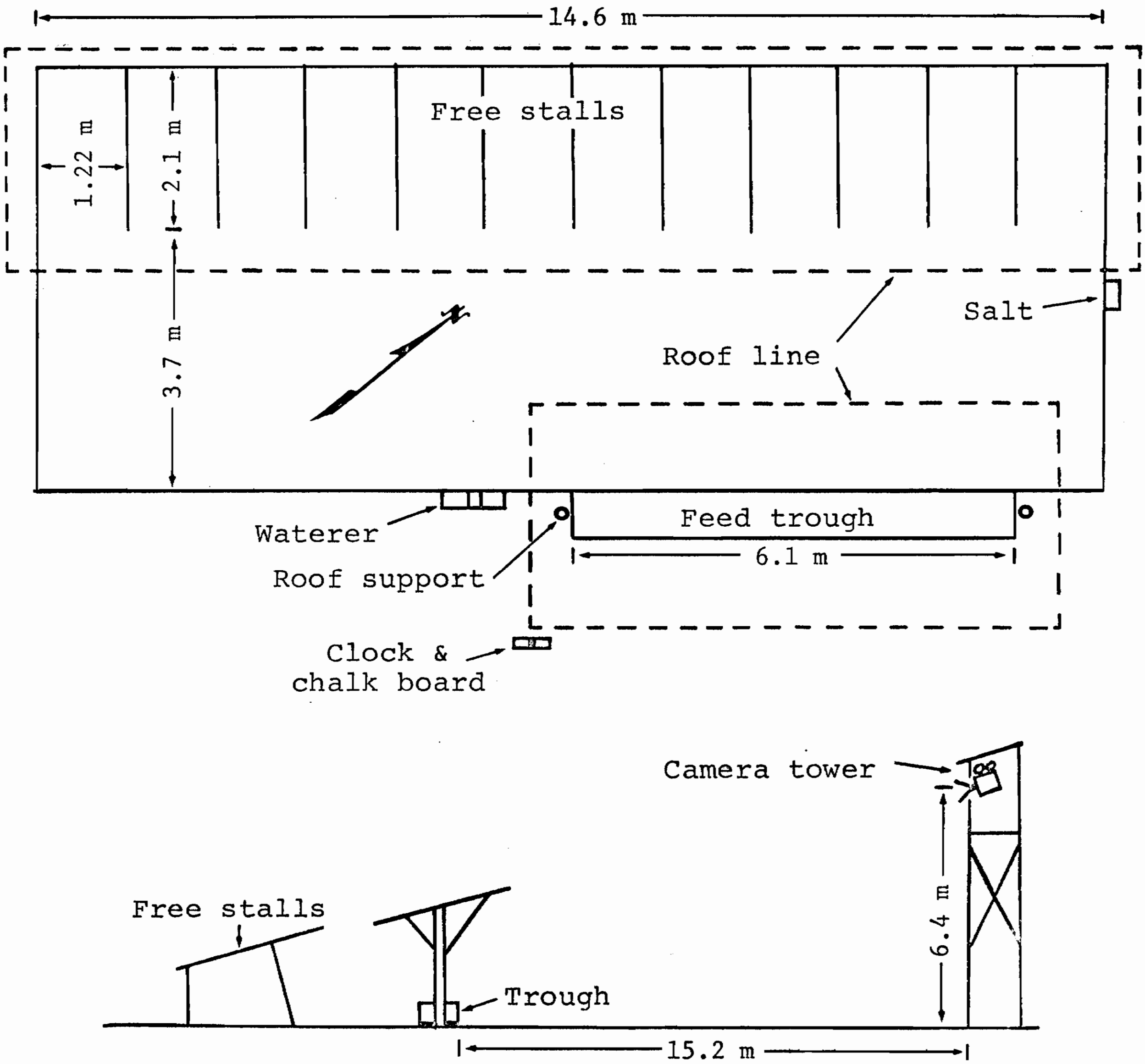


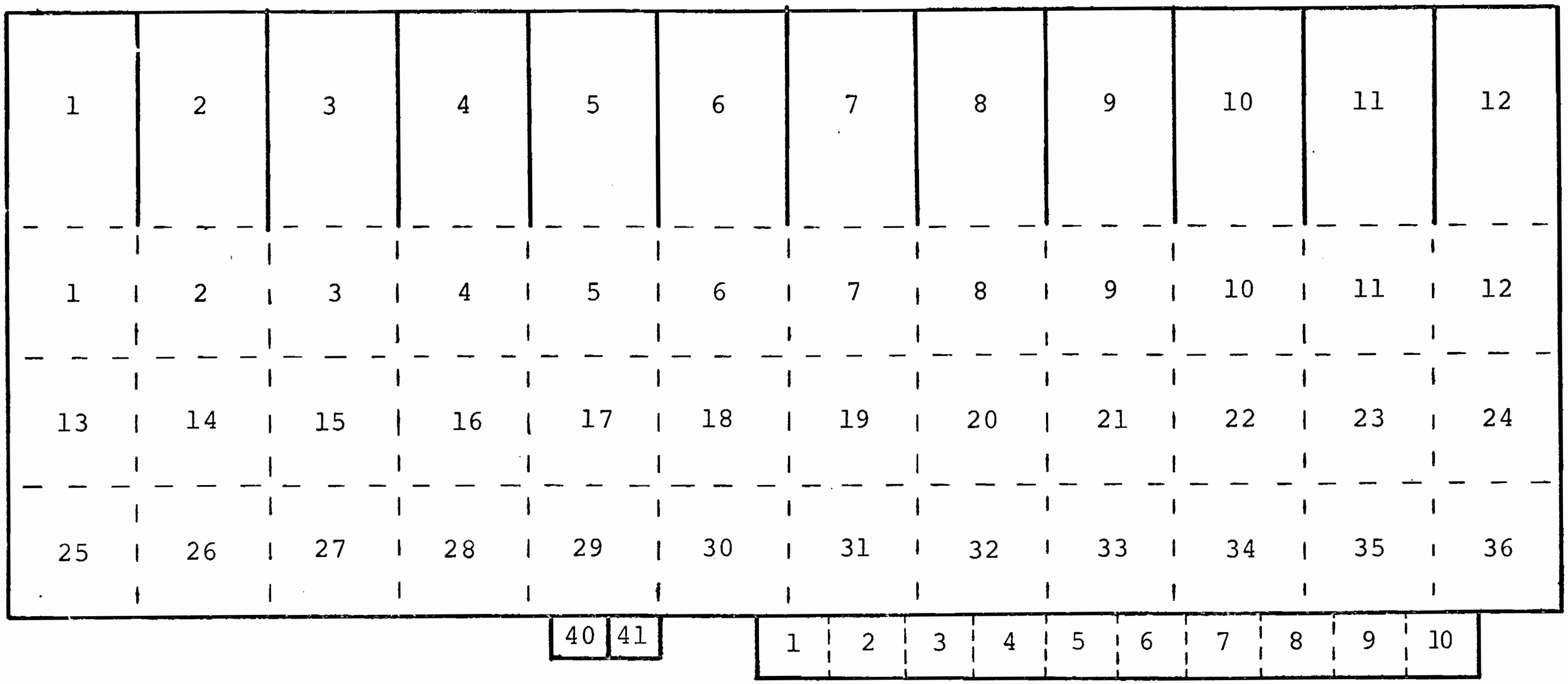
Figure 1. Dairy cattle behavior facility; overhead and lateral views.

was available ad libitum throughout the study despite decreased length of trough.

Behavioral data. Quantitative behavioral data were collected with an 8 mm movie camera (Minolta 8D10) wired to take one frame per minute. This provided 1300 observations or photographs per day excluding milking time. The housing facility was designed to allow unobstructed photographic surveillance of all cows. Each photograph was examined using a Kodak MFS-8 projector. All cows were identified by their natural markings.

The last three days of each treatment were coded for computer analysis except when 4 free stalls were available only 2 days were used. The 6.1 m feed bunk was divided into ten equal sections by painting white lines on the bunk. A cow was coded as eating from a particular section when observed with her head over the bunk. While occupying a free stall, she was recorded as using that stall. The alley was divided into 36 equal quadrants by visually extending each free stall partition across the alley and two lines perpendicular to these the length of the alley (Fig. 2). Cow location was coded as being in the quadrant of their head and shoulders. Four columns of each computer card were assigned to an individual. A typical entry would be FS08. The cow assigned to those columns was in free stall 8 when the photograph was taken. Free stall, feed bunk and alley data were coded simultaneously only during the trials in experiment I when free stalls were being reduced, otherwise only feed trough data were recorded.

A computer program was written in PL1 language to summarize the behavioral data. It would summarize for any coded location or activity such as resting in free stalls, eating, or drinking: total amount of time each individual appeared there; number of occurrances and mean duration with standard deviations for each activity; number of times a cow appeared adjacent to a given individual; and which cow succeeded or was next to occupy the location of a given individual within two minutes. The point of initiation and time interval covered by each summary could be controlled by inserting "flag" cards into the data deck. Summaries produced by the program could be limited by specifying "print codes" in the program.



Codes: FS = Free stalls
FB = Feed bunk
LS = Lot sections
MI = 1 - 36, for cows lying in alley
MI = 40, 41, waterer

Figure 2. Locations and codes for computerizing behavioral data.

Determination of dominance. Personal observation of agonistic behavior in each herd was made periodically during the experiments. These observations were used in the following procedure developed by Dr. M. L. McGilliard of the Department of Dairy Science, V. P. I., to determine where a cow ranked in the social hierarchy.

A dominance value (DV) for each cow was estimated by least squares solutions to the equation $Y_{ij} = G_i + DV_j + e_{ij}$ where Y_{ij} is a observation of cow j in encounter i. An observation was zero when cow j yielded to another cow and one when the other cow yielded. G_i is the value of the i^{th} encounter, an encounter being two cows meeting such that one yields to the other. DV_j is the dominance value of cow j, and e_{ij} is residual. Solutions were not obtained for the G_i's. The G_i's were absorbed into the DV_j equations, and the remaining equations were doubled and restricted by adding five to each diagonal element. Five is approximately twice $\sigma_e^2 / \sigma_{DV}^2$ from Friend and Polan, 1974 (27). This augmentation removes dependencies in the equations and regresses DV_j for numbers of observations on each cow. The sum of DV_j's will be zero. This yields Best Linear Unbiased Predictors or DV_j as described by Henderson (44). Thus, this methodology for computing dominance values will be referred to as BLUP.

For example, if

Encounter 1: Cow 2 yields to cow 1

Encounter 2: Cow 4 yields to cow 3

Encounter 3: Cow 3 yields to cow 2

Encounter 4: Cow 1 yields to cow 2

then the equations before absorption are

$$\begin{bmatrix} 1 \\ 0 \\ 1 \\ 0 \\ 1 \\ 0 \\ 1 \\ 0 \end{bmatrix} = \begin{bmatrix} 1 & 0 & 0 & 0 & 1 & 0 & 0 & 0 \\ 1 & 0 & 0 & 0 & 0 & 1 & 0 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 1 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 & 1 \\ 0 & 0 & 1 & 0 & 0 & 1 & 0 & 0 \\ 0 & 0 & 1 & 0 & 0 & 0 & 1 & 0 \\ 0 & 0 & 0 & 1 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 1 & 1 & 0 & 0 & 0 \end{bmatrix} \times \begin{bmatrix} G_1 \\ G_2 \\ G_3 \\ G_4 \\ DV_1 \\ DV_2 \\ DV_3 \\ DV_4 \end{bmatrix}$$

$$Y = X \cdot \beta$$

The first two equations taken out of matrix form are $1 = G_1 + DV_1$ and $0 = G_1 + DV_2$.

$$\begin{bmatrix} 2 & 0 & 0 & 0 & 1 & 1 & 0 & 0 \\ 0 & 2 & 0 & 0 & 0 & 0 & 1 & 1 \\ 0 & 0 & 2 & 0 & 0 & 1 & 1 & 0 \\ 0 & 0 & 0 & 2 & 1 & 1 & 0 & 0 \\ 1 & 0 & 0 & 1 & 2 & 0 & 0 & 0 \\ 1 & 0 & 1 & 1 & 0 & 3 & 0 & 0 \\ 0 & 1 & 1 & 0 & 0 & 0 & 2 & 0 \\ 0 & 1 & 0 & 0 & 0 & 0 & 0 & 1 \end{bmatrix} \times \begin{bmatrix} G_1 \\ G_2 \\ G_3 \\ G_4 \\ DV_1 \\ DV_2 \\ DV_3 \\ DV_4 \end{bmatrix} = \begin{bmatrix} 1 \\ 1 \\ 1 \\ 1 \\ 1 \\ 2 \\ 1 \\ 0 \end{bmatrix}$$

$$X'X \qquad \beta \qquad X'Y$$

After absorption, doubling and adding 5 to diagonal elements, the equations with resulting dominance values become

$$\begin{bmatrix} 7 & -2 & 0 & 0 \\ -2 & 8 & -1 & 0 \\ 0 & -1 & 7 & -1 \\ 0 & 0 & -1 & 6 \end{bmatrix} \times \begin{bmatrix} DV_1 \\ DV_2 \\ DV_3 \\ DV_4 \end{bmatrix} = \begin{bmatrix} 0 \\ 1 \\ 0 \\ -1 \end{bmatrix} \quad \text{and} \quad \begin{bmatrix} DV_1 \\ DV_2 \\ DV_3 \\ DV_4 \end{bmatrix} = \begin{bmatrix} .038 \\ .134 \\ -.004 \\ -.168 \end{bmatrix}$$

This reduced augmented X'X matrix can be formed easily by making each nondiagonal element the number of encounters of a pair times -1, diagonal elements the total number of encounters of a cow plus $2\ \sigma_e^2 / \sigma_{DV}^2$ (5 in this example), and X'Y the number of wins minus losses for each cow.

BLUP regresses DV's simultaneously toward the mean. We believe this gives a more equitable ranking than the procedure proposed by Kaiser (51). Kaiser does not discuss the situation of multiple encounters between two cows. Kaiser's measure of internal consistency (R^2) for Experiments I and II was .58. The similar measure: $R^2 = (\frac{1}{2}\ \text{No. of Enc.} - \frac{1}{2}\ \Sigma(DV_i \times X'Y_i))$ / No. of Enc. from BLUP for Exp. I and ii was .67 and .64. Kaiser's method compared favorably with angular dominance values and weighted dominance values (8) and was used by Dickson et al. (22), Friend and Polan (27), and Brakel and Leis (10).

Regression analysis. Multiple regressions were used to determine the relative importance in predicting access to feed, free stalls or individual feed intake of DV, age, body weight, daily milk production during each treatment, mature equivalent (ME) milk production (corrected for stage of lactation and age) and percent milk fat. The best 10-variable model using the Maximum R^2 Improvement procedure of the Statistical Analysis System (7) was determined for linear and quadratic combinations of the above independent variables. The partial regression coefficients for each regression were standardized for variation and then converted to percent of their sum disregarding signs (Tables 2, 4, 6, & 7). On a percentage basis, the relative importance of independent variables can be compared within and across treatments. The actual β values for the regressions are in Appendix Tables 1 - 4. The depletion of degrees of freedom of error (12 observations for 10 variables, Tables 2, 4, & 6) should be considered in interpretation of the R^2 for each regression.

Experiment II

This experiment was designed to compare estimates of individual feed intake of group fed dairy cows at two levels of competition. Intake is also related to behavior patterns, dominance, and other parameters.

The herd and husbandry. Twelve Holstein cows were used. They were selected, as in Exp. I, to represent a cross section of a typical dairy herd. There were 4-three, 5-four, 1-five, 1-six, and 1-eight year old with milk production ranging from 7,200 to 10,000 kg ME production. Cows ranged from 61 to 122 days post partum. They were housed in the same facility (Fig. 1) as in Exp. I with access to all free stalls and feed trough. Alley size was reduced however, by moving the free stalls to within 2.4 m of the trough. DV's for each cow as well as quantitative behavioral data were determined using the procedures described in Exp. I.

Feed intake determination. Individual feed intake was estimated using dual dietary indicators (21). Cows were allowed 2 weeks for adjustment to a complete corn silage based ration containing 25% ground orchard grass hay to assure a minimum of 3.5% lignin in the ration. Each cow was then given 10 g of encapsulated Cr_2O_3 commencing November 11, 1974 (day 1) to estimate fecal output. Feed intake was estimated by relative concentration of dietary lignin in the fecal samples. Individual fecal samples were obtained on day 6 (of the Cr_2O_3 administration) at 1100, day 7 at 300, day 7 at 1900, day 8 at 700, day 8 at 2300, and day 9 at 1500 hours. Immediately after the final fecal sampling, feed trough space was reduced from .5 to .25 m per cow (treatment II). The fecal sampling sequence was then repeated beginning on day 13 at 1100 hours. Fecal samples were composited for each cow by treatments, dried at 60 C and ground for analysis. Cr_2O_3 concentration was determined by a modification of the procedure of Hill and Anderson (45) and acid detergent lignin was determined by the Van Soest procedure (28).

Results and Discussion

Experiment I

Changes in behavior patterns and usage occurred in free stalls, exercise lot, and at the feed trough over the five levels of competition. Results from each location and their inter-relationships will be covered separately.

Distribution of dominance values (DV) for the twelve cows is shown in Figure 3. BLUP DV's were more evenly distributed when compared to those calculated by Kaiser's procedure (51). The shifts in order are due to BLUP's regressing for numbers of observations. BLUP values are used in all subsequent discussion of dominance.

Free Stalls. The effects of reducing number of free stalls is summarized in Table 1. Cows averaged 14.2 hours resting when there was low competition for free stalls, 3 hours longer than found previously (27). In that study, the exercise space was approximately 8 times larger and cows had access to a dirt paddock for about 2.5 hours per day in which they spent considerable time lying. Resting time was not reduced significantly from control (1 stall per cow) until there were six or fewer free stalls available. Meanwhile, the coefficient of variation increased probably due to a combination of desire for resting counterbalanced with social pressure for stall turnover. Percent utilization (use / time available x 100) increased with competition, reaching a plateau of maximal utilization between .5 and .67 free stalls per cow. The average number

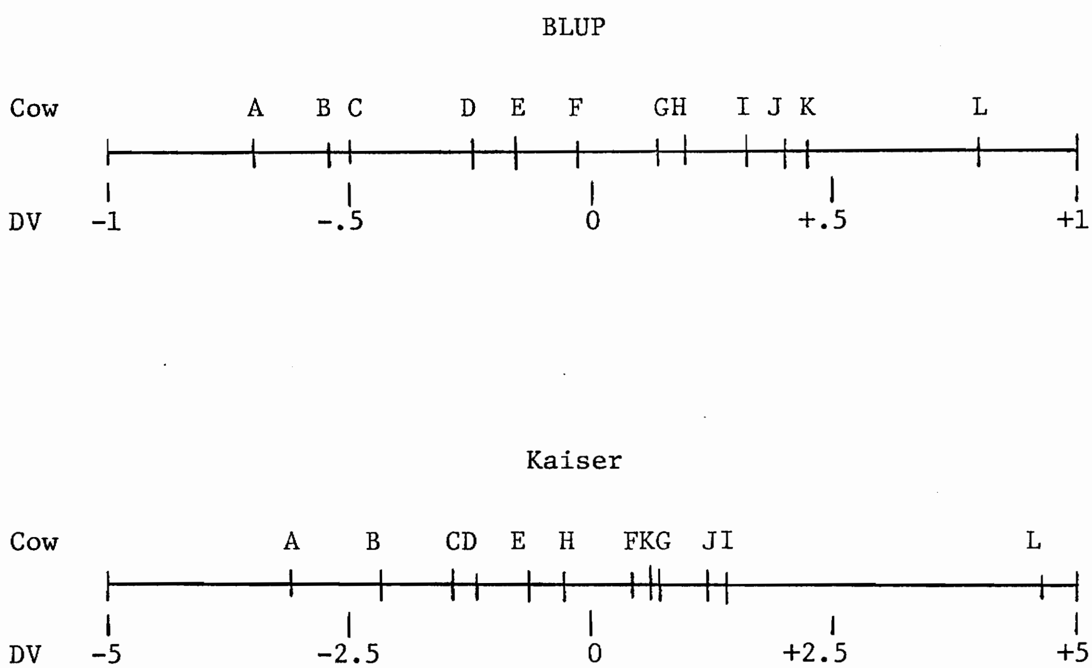


Figure 3. Distribution of dominance values (DV) calculated by BLUP and Kaiser's procedure (51) for cows in Experiment I.

Table 1. Effect of reducing number of free stalls on daily free stall utilization per cow.

	Free stalls per cow				
	1.0	.83	.67	.50	.33
Avg. time resting in stalls (hr±SD)	14.2±1.5	14.2±1.9	13.2±2.0	10.4±2.0[D]	6.9±2.0[D]
Correlation with dominance value	-.26	.15	.18	.17	.23
Coefficient of variation for time resting	10.9%	13.5%	15.3%	20.0%	28.5%
Percent utilization[1]	66.9%	80.3%	93.2%	97.8%	98.1%
Avg. no. of resting periods ±SD	10.7±2.6	11.3±2.7	10.5±2.1	7.9±2.6[D]	6.4±1.8[D]
Correlation with dominance value	-.37	.11	.09	.65*	.43
Avg. duration of resting periods (hr±SD)	1.34±.64	1.25±.64	1.25±.69	1.22±.61	1.11±.42
Correlation with dominance value	.21	.06	.07	-.48	-.48
Correlation of dominance value with avg. dominance value of neighbors while resting[2]	-.69*	.12	.42	.04	.30
Correlation of dominance value with avg. dominance value of successors[2]	-.13	-.14	-.20	-.18	-.51

[1]Use / time available X 100.

[2]Weighted by time.

[D]Differs from 1.0 stalls per cow (dunnett's test).

*Differs from zero (P<.05).

of resting periods per day did not decrease until .5 stalls per cow or fewer were available. Average duration of resting periods changed very little. Average feed intake per cow of total mixed ration did not change with competition for stalls. Correlations of dominance value with time in free stalls (Table 1) became larger with more competition. When one stall per cow was available, dominant cows had fewer numbers of resting periods, but of a longer duration compared to subordinates. With increased competition, a reversal occurred at .5 or fewer stalls per cow. Average rank of a cow's neighbors while resting was correlated (r = -.69) with DV and cows occupied stalls adjacent to opposites in DV only when there was 1 stall per cow. Average DV of a cow's successors (the next cow to occupy the free stall) showed a slight negative correlation with the successee's DV.

Some cows appeared to evict a stall occupant, subsequently occupying the space themselves. Several evictors were more forceful and apparently more impatient than others. Number of evictions observed per day were 3, 3, 4, 6, and 7 for 1.0, .83, .67, .50, and .33 stalls per cow. Some cows evicted were higher in DV than the evictor, however, there was no trend associated with number of available stalls relative to DV.

When .50 or .33 stalls were available, cows were observed lying an average of 18.2 and 20.1 minutes per day in the alley. This was not correlated with DV, age, weight, or production.

The relative effect of 12 variables on time spent in stalls is shown in Table 2. Through examination of the percentage figures the relative importance of the independent variables can be judged across rows and columns. When 1 stall per cow was available, DV and the quadratic or square of DV contributed little to the model relative to % milk fat and square of % milk fat, which contributed a total of 52% of the explained variation. Production variables were especially indicative of time spent in stalls even as competition increased. When the other independent variables were held constant, ME milk production was related to time in free stalls in a positive direction for .83 through .33 stalls per cow. Percent milk fat however, had a neutral effect at 1.0 stalls and was negative across all other levels of competition. Percent milk fat was uncorrelated with ME production or daily production in these cows.

Table 2. Percent contribution of independent variables to variation accounted for by regression of time spent in free stalls during increased competition.

Independent variable	No. free stalls per cow 1.0	.83	.67	.50	.33
DV	1	1		1	
DV^2	1	1	0	1	
Age	11	8	6	8	4
Age^2	11	7	3	7	2
Body weight	6	6	6	7	7
Body $weight^2$	6	7	8	8	8
Daily prod.	6		6		4
Daily $prod.^2$	6		5		3
ME milk prod.		20	32	20	30
ME milk $prod.^2$		21	33	21	30
% milk fat	26	15		14	6
% milk fat^2	26	14	1	13	6
Total	100	100	100	100	100
R^{2a}	.86	.99	.99	.99	.96

$^aR^2$ = coefficient of multiple determination, depletion of degrees of freedom must be considered in interpretation (page 18).

^{2}Is the quadratic component.

Alley. Behavior of cows in the alley (concrete area between free stalls and feed trough) was recorded only during the free stall trials. If cows were not in stalls, they were in the alley or eating. Time in the alley was correlated negatively with time in stalls, except for the .33 stalls per cow treatment. There was no association between time in the alley and time at the trough. Cows with high and low DV spent the most time in the alley. Number of times a cow appeared adjacent to others in the alley (on neighboring quadrants) was quadratic with DV, R^2 = .62, .44, .10, .66, .17 for 1., .83, .67, .50, and .33 stalls per cow. Cows with high and low DV had the greatest number of neighbors. DV had little relationship to preference of neighbors in the alley. There was also no association between number of neighbors and DV in stalls or at the trough. Work by Syme et al. (88) indicated an inverse relationship between social status and spatial proximity (number of neighbors) and found submissive animals did not show marked avoidance of more dominant cows in a lot.

Feed Trough. The average time spent at the feed trough was uneffected by linear space until only .1 m of space per cow remained (Table 3). Coefficient of variation for use increased at this point. Prior to this, cows used the trough 3.7 hours per day, identical with earlier work (27, 76). Average daily feed intake appeared to drop at .1 m of trough per cow. Percent utilization was calculated on the assumption that two cows could eat from every 1.22 m of bunk space. For this reason, percent utilization did not approach 100% since some cows prevented others from eating near them. However, utilization efficiency increased markedly as competition increased at the feed bunk. Correlation of dominance with average duration of eating periods increased with less feed trough (Table 3). The correlation with average DV of a cow's neighbors while eating changed from .61 to -.52 with increased competition. During periods of low competition, cows similar in DV ate together. No changes in milk production were evident due to increased competition at the trough during this brief period.

Importance of 12 variables on time spent at the trough during increased competition is shown in Table 4. Production variables had the most influence on the time a cow spent eating as competition for feed increased.

Table 3. Effect of varying length of feed trough on daily utilization per cow.

	Feed trough length per cow				
	.5 m	.4 m	.3 m	.2 m	.1 m
Time spent at feed trough (hr) ± SD	3.82±.97	3.73±.79	3.73±.73	3.76±.87	2.57±.80[D]
Correlation with dominance value	.46	.32	.30	.67*	.71**
Coefficient of Variation for time at feed trough	25.3%	21.1%	19.5%	23.2%	31.0%
Percent utilization[1]	21.5	26.9	34.6	51.9	70.6
Avg. daily feed intake (kg)[2]	37.3	37.4	37.8	36.0	33.2
Correlation of dominance with avg. dominance of neighbors while eating	.61*	.37	.27	-.10	-.52
Correlation of dominance with avg. dominance of successors	.21	.35	.72**	-.21	-.05

[1]Use / time available X 100.

[2]Based on herd refusals ∿47% dry matter.

[D]Differs from .5 m feed trough per cow (Dunnett's Test).

*Differs from zero (P<.05).

**Differs from zero (P<.01).

Table 4. Percent contribution of independent variables to variation accounted for by regression of time spent at feed trough during increased competition.

Independent variable	Meters of feed trough per cow				
	.5	.4	.3	.2	.1
DV	2	0	1	2	
DV^2	1	0	1	0	1
Age	9		2	19	1
Age^2	8	1		17	1
Body weight		12	1		0
Body $weight^2$		13			
Daily prod.	9	8	10	11	7
Daily $prod.^2$	9	9	8	13	9
ME milk prod.	6	28	22	12	31
ME milk $prod.^2$	5	29	22	12	33
% milk fat	25	0	16	7	9
% milk fat^2	26		17	7	8
Total	100	100	100	100	100
R^{2a}	.99	.99	.99	.99	.99

[a] R^2 = coefficient of multiple determination, depletion of degrees of freedom must be considered in interpretation (page 18).

[2] Is the quadratic component.

When the other independent variables were held constant, ME production and percent milk fat (except for .1 m) positively effected amount of time at the trough. Daily milk production however, had a negative influence on time spent eating. The significant correlation between dominance and time spent eating (Table 3) are due to the relationship of dominance with production oriented variables and not dominance per se.

Relationships. Time spent in free stalls as the number of stalls per cow were reduced 1.0, .83, .67, .50, .33 was correlated ($P<.05$) with time at the feed trough -.85, -.84, -.72, -.61, -.77. Time spent standing in front of the feed bunk but with head not over the feed was correlated ($P<.05$) with age -.69, body weight -.62, ME milk production -.57, and daily milk production -.63. Young, lightweight and lower producing cows used their time in front of the trough inefficiently. Cows were observed most often in this position during periods of low competition when only one or two cows were eating. The amount of time cows spent on the periphery of the group eating at the trough during peak periods of use (eg. return from milking, or post-feeding) was correlated with DV -.40, age -.31, body weight -.28, ME milk production -.16, and daily milk production -.51. Submissive, young, light, low producing cows stood on the periphery.

Activity was measured as number of times a cow changed location in free stalls, alley, and feed trough during the free stall trials. Number of changes had no linear association with DV, but a trend was evident when quadratic effects were tested, R^2 = .35, .24, .23, .52, .52, the latter two differ from zero ($P<.05$), for 1.0, .83, .67, .50, and .33 stalls per cow. Dominant cows were active, possibly chasing cows while low DV cows may have actively avoided superiors during the 6 and 4 stall treatments. Syme et al. (88) found that apart from the most "dominant" animal, their high ranking cows moved further than the low ranking cows using a grid system similar to that used in this study. However, they deleted a cow because she did not conform and had a low number of cows for observations.

Experiment II

This experiment was designed to examine the association of dominance and other variables with individual dry matter intake at two levels of competition in twelve group fed cows. The distribution of dominance values for the cows is shown in Figure 4. BLUP dominance values were more evenly distributed when compared to those calculated by Kaiser's procedure (51). The shifts in order are due to BLUP's regressing for numbers of observations. BLUP values are used in all subsequent discussion of dominance. Dominance values were correlated with age, body weight, and ME milk production, .43, .55, .12, respectively. Age was correlated with body weight, .72 ($P<.01$).

Table 5 summarizes ingestive data collected during Experiment II. Estimated average daily dry matter intake, was 13 and 22 percent below the actual intake based on refusals for the group when .5 m and .25 m of trough space were available per cow. Estimated individual dry matter intakes are in Appendix Table 5.

Time cows spent at the feed trough was slightly longer than in Exp. I. During this study, temperatures averaged 5 C, except during part of the .25 m feed trough treatment when temperatures dropped to a daily average of -4 C but with little wind chill. This could help account for the increased intake during that treatment.

Relative importance of 12 variables in describing time at the feed trough and individual dry matter intake is shown in Table 6. Trends in the relative importance of variables describing time spent eating at .5 m and .25 m are similar to those in Exp. I. Individual dry matter (DM) intake was positively affected by percent milk fat. Body weight and daily production also increased dry matter intake at .5 and .25 m. Chandler and Walker (15) found DM intake of cows fed individually to be significantly affected by season, body weight, crude fiber, daily milk production and percent milk fat.

The 12 cows in this study were relatively homogeneous, days in lactation (± SD) were 115.8 ± 20.2 and daily production (kg ±SD) during .5 m and .25 m treatments was 29.25 ± 4.8 and 28.1 ± 5.8. Daily production may have contributed more to variation in intake if there had been greater diversity in production. The relationship was quadratic (R^2 = .52) for time at the trough with individual dry matter intake during the .25 m trough per cow treatment. Cows who spent intermediate time at the trough

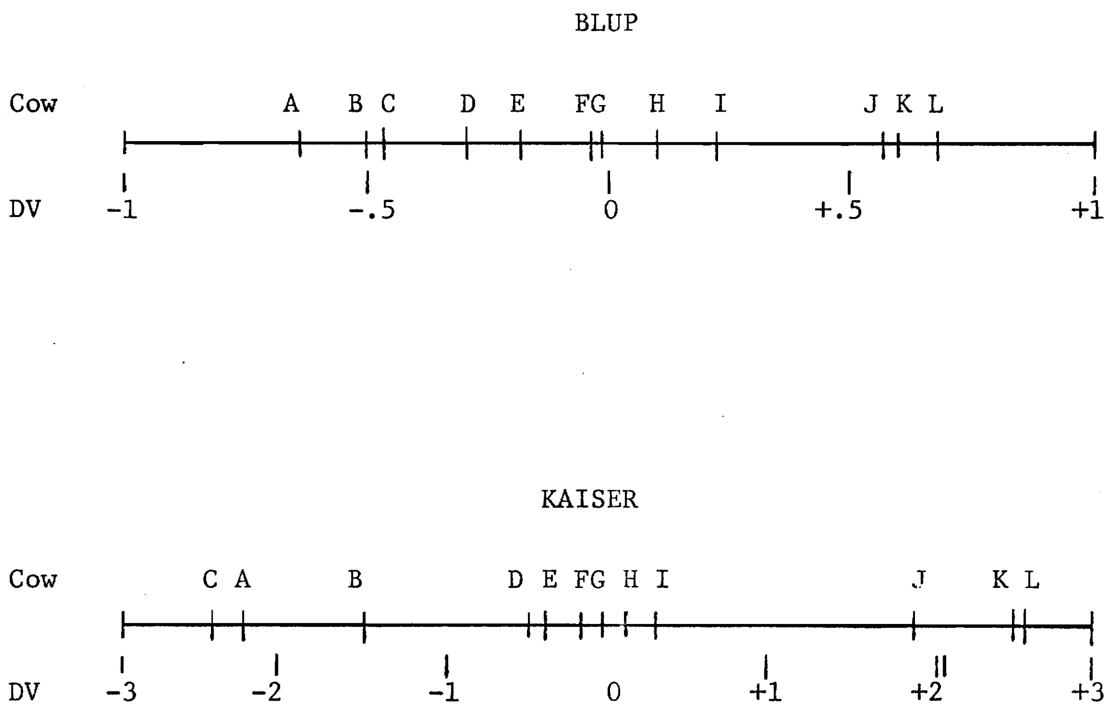


Figure 4. Distribution of dominance values (DV) calcualted by BLUP and Kaiser's procedure (51) for cows in Experiment II.

Table 5. Dry matter (DM) intake and feed trough use during Experiment II.

	Length of feed trough per cow	
	.5 m	.25 m
Estimated avg. daily DM intake based on individual estimates (kg) ± SD	18.9±2.6	19.7±2.8
Actual avg. daily DM intake based on group refusals (kg)	21.8	25.2
Avg. use (hr) ± SD	4.54±.79	3.96±.66
Percent utilization[a]	27.2%	49.7%

[a]use / time available X 100.

Table 6. Percent contribution of independent variables to variation accounted for by regression of time spent at feed trough and individual dry matter (DM) intake at two levels of competition.

Independent variable	Time at trough		D.M. intake	
	.5	.25	.5	.25
DV	1	0	0	
DV^2		0	3	0
Age	5	7	9	9
Age^2	6	7	7	13
Body weight	12	10	14	
Body $weight^2$	13	9	12	1
Daily prod.			3	8
Daily $prod.^2$	1			10
ME milk prod.	12	21		12
ME milk $prod.^2$	12	22	1	11
% milk fat	19	13	25	19
% milk fat^2	18	12	26	17
Total	100	100	100	100
R^{2a}	.99	.99	.99	.99

[a]R^2 = coefficient of multiple determination, depletion of degrees of freedom must be considered in interpretation (page 18).

[2]Is the quadratic component.

apparently consumed the most.

When .5 m of trough was available per cow, cows tended to eat beside and succeed cows of opposite DV (r = -.17 and -.50) commensurate with Exp. I. Altering of behavior did not occur until .2 m or less was available.

General Discussion

Spatial Requirements of Dairy Cattle

Time spent resting, number of resting periods, and percent utilization changed drastically indicating a crowding situation when less than .67 free stalls per cow were available. The relationship of DV to free stall use changed also. Based on these data, minimum stalls needed per cow without altering daily free stall usage = 14.2 / (.93 x H) where 14.2 is the average desired resting time in hours (h), .93 is maximum efficiency of use precrowding, and H is hours per day that free stalls are available to the herd. Therefore, .71 stalls is the calculated minimum in this study for 21.4 h. One stall per cow is the maximum needed if readily accessible. To assure reaching a plateaued resting time, each cow must have access to a free stall 15.3 h per day. The minimum number of stalls needed could well vary depending on the design of the facility and season. If empty free stalls are obstructed visually it is less likely cows will use them efficiently. The effect of herd size on social stability and associated behavior is not known.

There was a marked change in ingestive behavior when trough length was restricted to .1 m per cow. Dominance value played a more important role in time spent eating based on correlation. It appears .2 m of trough per cow is adequate provided individuals have access to feed at least 21 hours per day.

Based on Exp. II, time spent at the feed trough is not predictive of DM intake when an abundance of feed trough space is available. Some cows utilize time at the trough more efficiently than other cows.

Current recommended stocking rates are more conservative than necessary. In general, dairymen could have up to 30% more cows than free stalls and increase the number of cows per unit of feed trough without altering behavior of production. This means it is possible to increase herd size without increasing facilities within limits. A dairyman with an 80 cow free stall barn could increase his herd to 104 cows in his persent facility. This would mean a savings of at least $7,166.00 with a cold type barn (9), by not building the 24 additional free stalls called for by current recommendations (5, 61).

Importance of Dominance Values in Resting and Feeding

To evaluate the importance of DV on access to limited amounts of feed, data from an earlier study (27) was subjected to regression analysis. There were 21 cows housed in a totally different facility from Exp. I and II (Fig. 5). Approximately 45 kg of concentrate (16% crude protein) was spread over the silage remaining from the previous night when the cows were removed from the lot for the morning milking. At 1100 hours two bales of alfalfa hay were offered in the trough. During the afternoon milking, the residual silage was removed and the trough refilled with fresh silage (∿40 kg per cow per day).

A highly competitive situation arose at the feed trough upon the herd's return from milking and when hay was fed since about 66% of the herd could eat from the trough simultaneously. The supplemental concentrate poured on the silage was consumed in 15 minutes with the correlation between time spent eating during that 15 minute period and DV being .60 ($P<.01$). The correlation (.29) was not different from zero when the period was the 30 minutes that was needed for the hay to be consumed.

When six characteristics of the cows are evaluated simultaneously in a model (Table 7), DV played little if any role in determining access to resources. ME milk production and ME milk production squared contributed 60% of the explained variation while DV was of negligible importance in describing the amount of time cows had access to the concentrate. Therefore, the high correlation DV has was due to its interrelationships with other more important variables. Cows with higher ME production consumed the concentrate. Daily milk production and percent milk fat were most important in predicting (positively) access to the hay. The total amount of time at the trough per day was influenced (negatively)

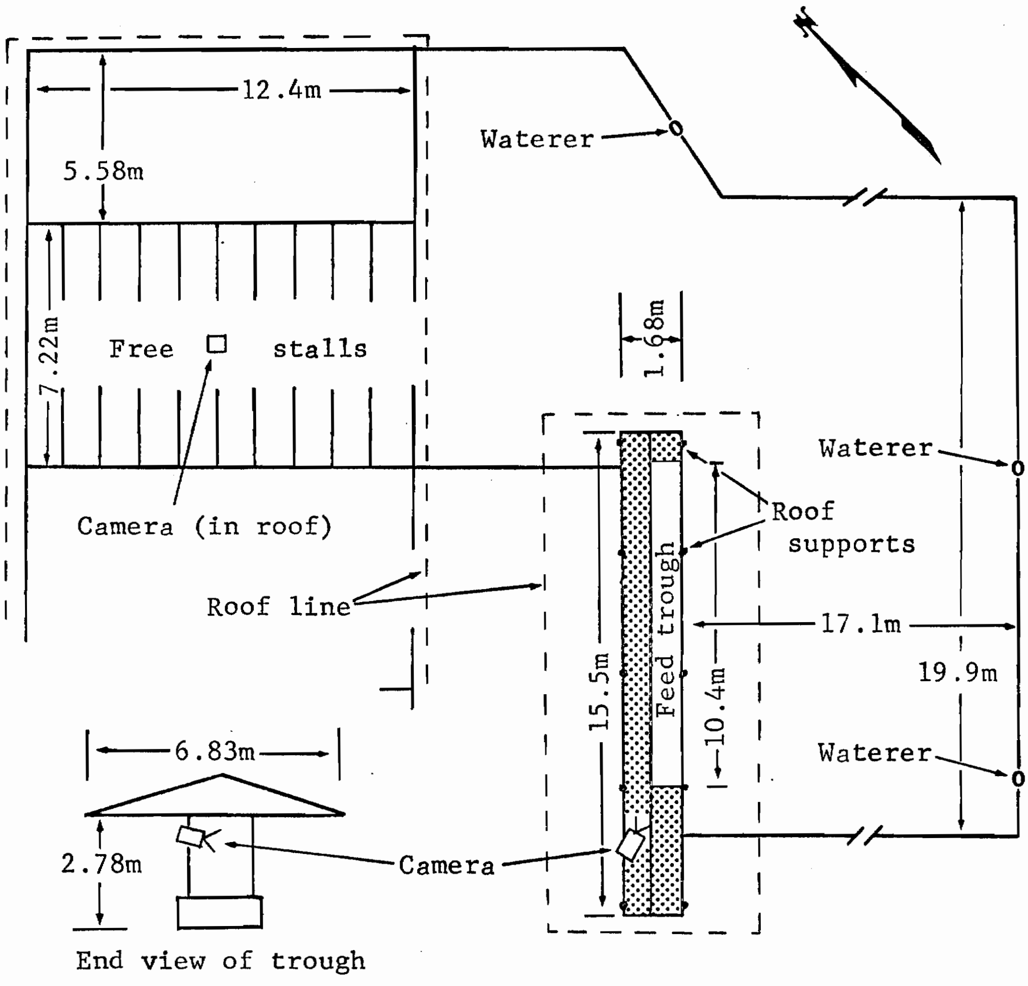


Figure 5. Overhead view of dairy cattle housing for earlier study by Friend and Polan (27).

Table 7. Percent contribution of independent variables to variation accounted for by regression of time spent at feed trough or in free stalls.

Independent variable	First 15 min post-feeding concentrate	First 30 min post-feeding hay	At trough per day	In stalls per day
DV	1			0
DV^2	1		1	1
Age	7	3	11	
Age^2	7	3	9	
Body weight	1	11	8	15
Body $weight^2$		8	8	16
Daily prod.	11	15	10	11
Daily $prod.^2$	11	16	11	12
ME milk prod.	30	4	21	10
ME milk $prod.^2$	30	5	20	8
% milk fat		17	1	14
% milk fat^2	1	18		13
Total	100	100	100	100
R^{2b}	.83	.85	.70	.64

[a]Ten variables were run so that this table would be consistent with Tables 2 - 4. Blank variables are essentially zero.

[b]R^2= coefficient of multiple determination.

[2]Is the quadratic component.

by age and production. Time spent in free stalls was best described by body weight (positively) and production oriented variables (negatively). It should be noted, however, that the only competition in this experiment was for the concentrate and hay which were limited in both access and quantity. The insignificant effect DV had in this study supports data from Exp. I and II.

In conclusion, the DV's used in these studies failed to be adequate measures of a socially mediated priority of access (87). This occurred for two different behaviors, eating and resting. What we have determined was an aggressive order rather than dominance. From these data, the existance of a classical dominance hierarchy in dairy cows is doubtful. It appeared that physiological drives are the prime mover in these cows, though one might suspect dominance may be more important in feral cattle. Production variables were most important and positively affected access to limited amounts of feed, dry matter intake, and time at the feed trough. Access to free stalls was best described by production oriented variables which had a positive effect. The high producing dairy cow is the product of many generations of selection for milk production and behaviors compatible with domestication. Cows operating close to their physiological peak apparently gain long term access to resources through persistency rather than aggression.

PART II: INFLUENCE OF BEHAVIORAL STRESS ON ADRENAL FUNCTION, MILK SOMATIC CELLS, AND MILK PRODUCTION

Procedure

Experiment I

The influence of behavioral stress on adrenal function and milk production was investigated in this experiment.

The herd and husbandry. Nine lactating Holstein cows between 15 to 72 days postpartum were used in each of 2 replicates (test cows). Test cows of each replicate were randomly assigned by age to 3 groups either tested as controls or at 2 or 9 days after initiation of stress to determine plasma corticoid response to 200 IU ACTH (Porcine, Sigma Chemical co., St. Louis, Mo.). Each treatment group contained 1-first, 1-second or third, and 1-fourth or more lactation cows. Animals were stressed by moving them into an established group of twelve cows (base) occupying the facility in Figure 1 (free stalls were moved 4.9 m from the feed trough), and the resulting high stocking rate. Each cow had access to .67 free stalls, .34 m linear feed trough, and 3.96 m^2 concrete lot space. Based on Part I, Exp. I, the feed trough space is more than adequate while .67 free stalls per cow is marginally adequate for cows in this facility. The control cows remained in the approximately 20 cow herd from which all test cows originated. Test cows had a minimum of 2 weeks for acclimation to the herd before 2 and 9 day cows were moved in with with the base cows initiating stress. This herd had 1 free stall, .5 m feed trough and approximately 15 m^2 concrete lot per cow.

Adrenal function determination. Cows were fitted with indwelling jugular catheters 14 hours prior to ACTH injection. Catheters were filled with heparinized (30 IU/ml) saline, then tucked in a pouch glued to the neck. Cows then were returned to their respective groups until testing. Total plasma corticoids were determined by competitive protein binding assay (38) from blood samples collected at hours 2, 1, 0 pre-injection; .25, .50, .75, 1, 2, 3, 4, 5, 6, 7, 8, 9, and 10 post-ACTH injection. Blood samples (30 ml) were drawn into heparinized syringes, immediately placed into an ice bath, centrifuged at 4 C, and then frozen until analyzed.

The first replicate was tested in January; the second in March, 1976. Cows were restrained in tie stalls during sampling with access to feed and water.

Experiment II

The influcenc of 1, 2, and 3 days of behavioral stress on adrenal function and milk production was investigated in this esperiment.

The herd and husbandry. Adrenal responsiveness to 200 IU ACTH was determined on 16 cows (test) ranging from 32 to 327 days postpartum in July, 1976. All were first lactation cows to minimize effect of age on response found in Exp. I and reported by Shayangar et al.(80). Test cows were randomly assigned to 4 groups to be tested either as non-stressed controls or at 1, 2, or 3 days after initiation of stress. Introduction of the 12 into an already established group of 12 cows (base) occupying the facility in Fig. 1 and the resulting crowding constituted the stress. Each of the 24 cows had access to .5 free stalls, .25 m feed trough space and 2.97 m^2 of lot space. Based on Part I, Exp. I, at .5 stalls per cow, a crosding situation esists, the amount of time cows normally rest is restricted. Available lot space was restricted. However, Arave et al. (3) found restricting cows to 2.3m^2 was actually beneficial. Control cows remained in the herd from which the test cows originated (described in Exp. I) and each randomly assigned to have their response to ACTH tested with the stressed cows. Relative aggressiveness values for the 16 test cows were determined from direct observations of agonistic behavior in their original herd and the least squares ranking procedure BLUP described in Part I of this dissertation.

Adrenal function determination. Cows were fitted with jugular catheters and handled as in Exp. I. Total plasma corticoids were similarly determined from blood samples collected at hours 2, 1, 0 pre-ACTH injection; .25, .50, .75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.25, 2.5, 3.0, 3.5, 4, 5, and 6 hours post-ACTH. Cows were restrained in tie stalls during blood sampling with access to feed and water. When cows were removed for testing, 4 substitutes were temporarily added to ensure there were always 24 animals in the facility. Substitute cows came from the same herd as the cows tested; thus the effect of their presence on the tested cows adrenal response was minimal.

Milk somatic cell counts. Quarter milk samples were taken at evening milkings on tested and base cows -6, -2, and +4 days of stress. These milk samples were evaluated for possible mastitis causing bacteria (63) and milk somatic cell counts (MSCC) were made using the Fossomatic. Fossomatic MSCC's were also made on composite samples for each cow -5, -4, -3, -1, +1, +2, and +3 days of stress. The Fossomatic compares with direct microscopic counting (43).

Statistical Analysis.

Least squares analysis of variance (nested design) and multiple regression analyses were used to evaluate effects of treatments (days of stress) and other variables on response to ACTH. Dunnett's procedure was used to compare treatment means with controls. Corticoid response (area under the curve) was determined for each cow by $\Sigma(C_{s+1}/2 \times I)$, where C is the total plasma corticoids in ng/ml for sample s and the next sample s + 1 with an interval of I hours between them. Response was integrated from time 0 to 6 hours post-ACTH, by which time corticoids were back to pre-ACTH concentrations. To illustrate plasma corticoid concentrations in response to ACTH, regression equations through the fifth order were calculated. Only the highest order equation significant was used.

Results

Experiment I

All cows adjusted to the sampling routine with no apparent nervousness after second or third blood samples. Mean basal plasma corticoid concentration before ACTH (-2, -1, and 0 hours, Appendix Table 6) was 7.4±5.0 ng/ml (±SD, n=54). Basal cortiocids varied greatly over time and were not affected by treatments, time, age, daily milk production, percent milk fat, days in lactation or body weight. Mean basal corticoid concentration for each cow was not correlated with response to ACTH.

Mean area (ng/ml ± SD) under the corticoid response to ACTH curves were 200.4 ± 28.0, 229.8 ± 33.5, 221.4 ± 35.2 for zero, 2 and 9 days of stress. Days of stress, body weight, days in lactation, daily milk production, and percent milk fat had no effect on corticoid response following ACTH. Age and mature equivalent milk production were the closest ($P<.12$ and .11) to having an effect. Stressed cows decreased slightly in milk production (Table 8), however the non-stressed controls also declined similarly. Corticoid reponse curves from individual data are plotted in Figure 6. Time by treatment interaction indicated that the curves were not heterogeneous. A trend for the stress cows to respond with greater corticoid output than controls is visible. Since there was an apparent change in responsiveness after 2 days, it appeared an early response was possible and merited further experimentation. The lack of significance was attributed to having only the mild stress of adjustment to a new environment and the older cows knowing each other. Exp. II was then designed to provide a stocking rate known to crowd the available free stalls as well as disturb the hierarchial organization.

Experiment II

All cows, except one, adjusted to the blood sampling routine. A control cow (944) remained extremely nervous, cowering and greatly resisting whenever approached for a blood sample. Because of this, it seemed justified to delete her from subsequent statistical analysis. Her response to ACTH was 113 ng/ml higher than the mean of the other controls. When she remained in the analysis, treatments were significant at $P<.10$.

Basal corticoids -2, -1, and 0 hours, Appendix Table 7, were not significantly affected by treatment, time, age, daily milk production, percent milk fat, days in lactation or body weight. Mean basal corticoid concentration was 7.19 ± 6.4 ng/ml (± SD, n = 46). Mean basal corticoids were correlated with response to ACTH, $r = .61$ ($P<.03$).

Analysis of variance showed treatments effected ($P<.025$) adrenal response to ACTH. Mean area (ng/ml±SD) under the corticoid response curves were 161.6±12.6, 158.2±28.8, 227.7±32.2, and 229.9±40.3 for zero, 1, 2, and 3 days of stress. Corticoids from 1 day under stress did not change while 2 and 3 day treatments resulted in greater corticoid output than controls (Dunnett's $P<.05$). Individual response was not affected by days in lactation, age, weight, milk production, percent milk fat or

Table 8. Daily milk production of control and stressed cows from both replicates in Experiment I.

	Control	Stressed	
	Adrenal response tested		Base[a]
No. of observations	6	12	24
Avg. daily prod. -3, -2, -1 days of stress (kg)	33.0±7.1[b]	31.4±6.8	25.1±5.6
Avg. daily prod. day 1 of stress (kg)	32.2±7.7	31.1±5.7	24.7±5.8
Avg. daily prod. day 2, 3, 4 of stress (kg)	32.2±8.4	30.7±6.4	24.6±5.2

[a]The same 12 base cows were used in both replicates.

[b]Standard deviation of observations.

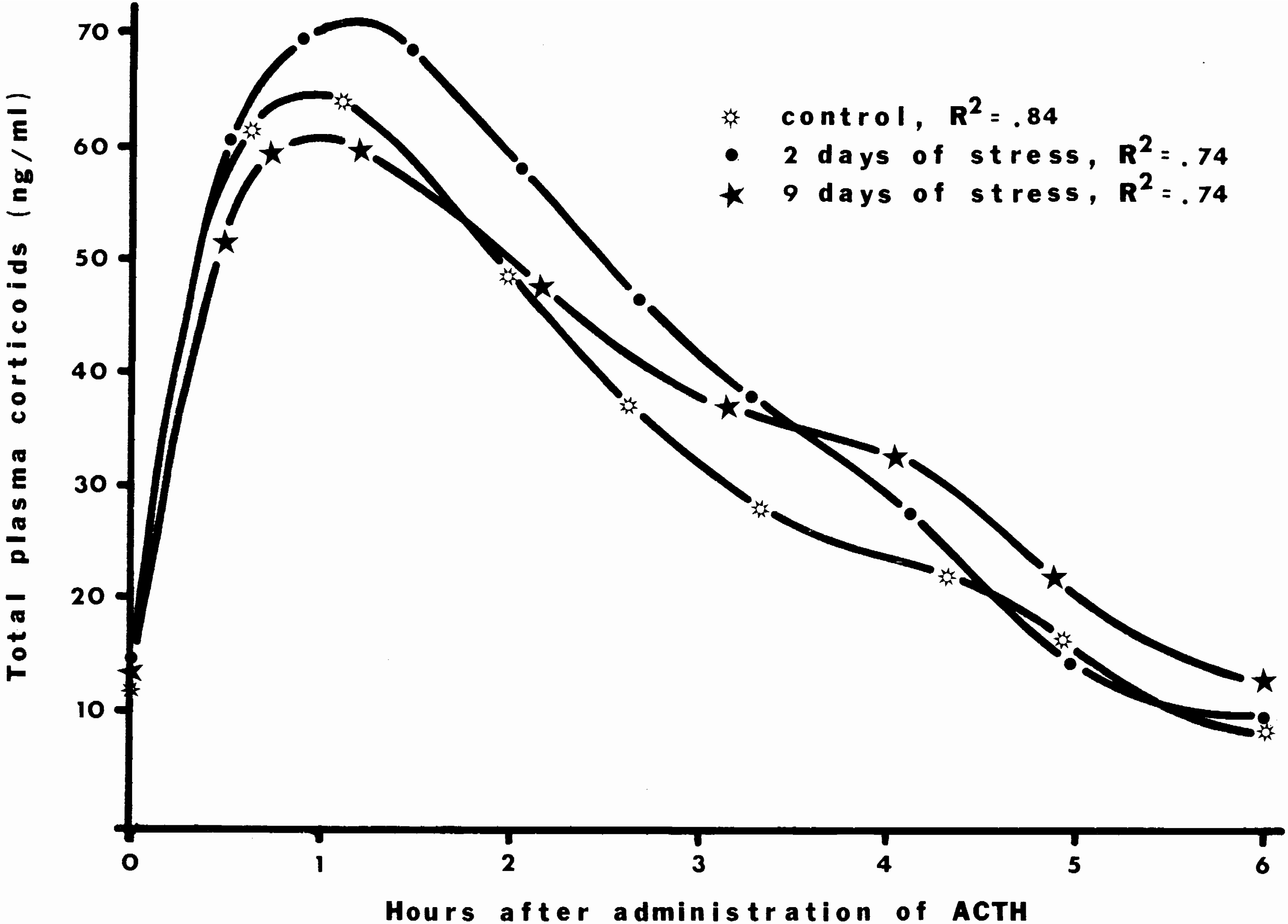


Figure 6. Total plasma corticoid concentrations in response to injection of 200 IU ACTH, Experiment I.

aggressiveness (DV). Response curves based on individual data are illustrated in Figure 7. Time by treatment interaction revealed no evidence that these curves were not parallel.

Time required for change in adrenal responsiveness to ACTH coincides with Selye's General Adaptation Syndrome (78). The second stage, or adaptation, begins 48 hours after an injury and is characterized by adrenal enlargement and increased production and storage of corticoids.

Stress appeared to have little effect on milk production in this experiment (Table 9). The tested group increased production while the base group decreased about the same amount during the first day of stress. The non-stressed controls similarily decreased in production during the same period.

Stress from introduction and subsequent crowding did not alter the bacteriological status of quarters from the 24 cows after 3 days of stress. There was one new bacteriologically positive quarter while all infections detected prior to stress (present both at -2 and -6 days) were present on day 4. Mean composite cell counts for 3 days prior and 3 days under stress in cows bacteriologically negative -6, -2, and +4 days of stress increased above 400,000 cells per ml for one cow and decreased in one. The composition of milk begins to alter when there are more than 150,000 somatic cells per ml in quarter samples (69). Counts increased above 150,000 cells in 8 and decreased in 5 quarters (bacteriologically negative) after 3 days of stress when compared to means of -2 and -6 days. In the 12 stressed adrenal response tested cows, 3 noninfected quarters had reductions while 4 increased above 150,000 cells per ml. Three quarters increased while 1 dropped below 150,000 cells per ml among the non-stressed controls. Only one of seven infected (Staphylococcus aureus) quarters had a change in somatic cell count. It decreased to less than 150,000 cells day 4 of stress. No trends were evident between somatic cell counts and corticoid release.

Discussion

One problem inherent in using research herds for behavioral stress work is that these cattle may be more accustomed to change than cattle

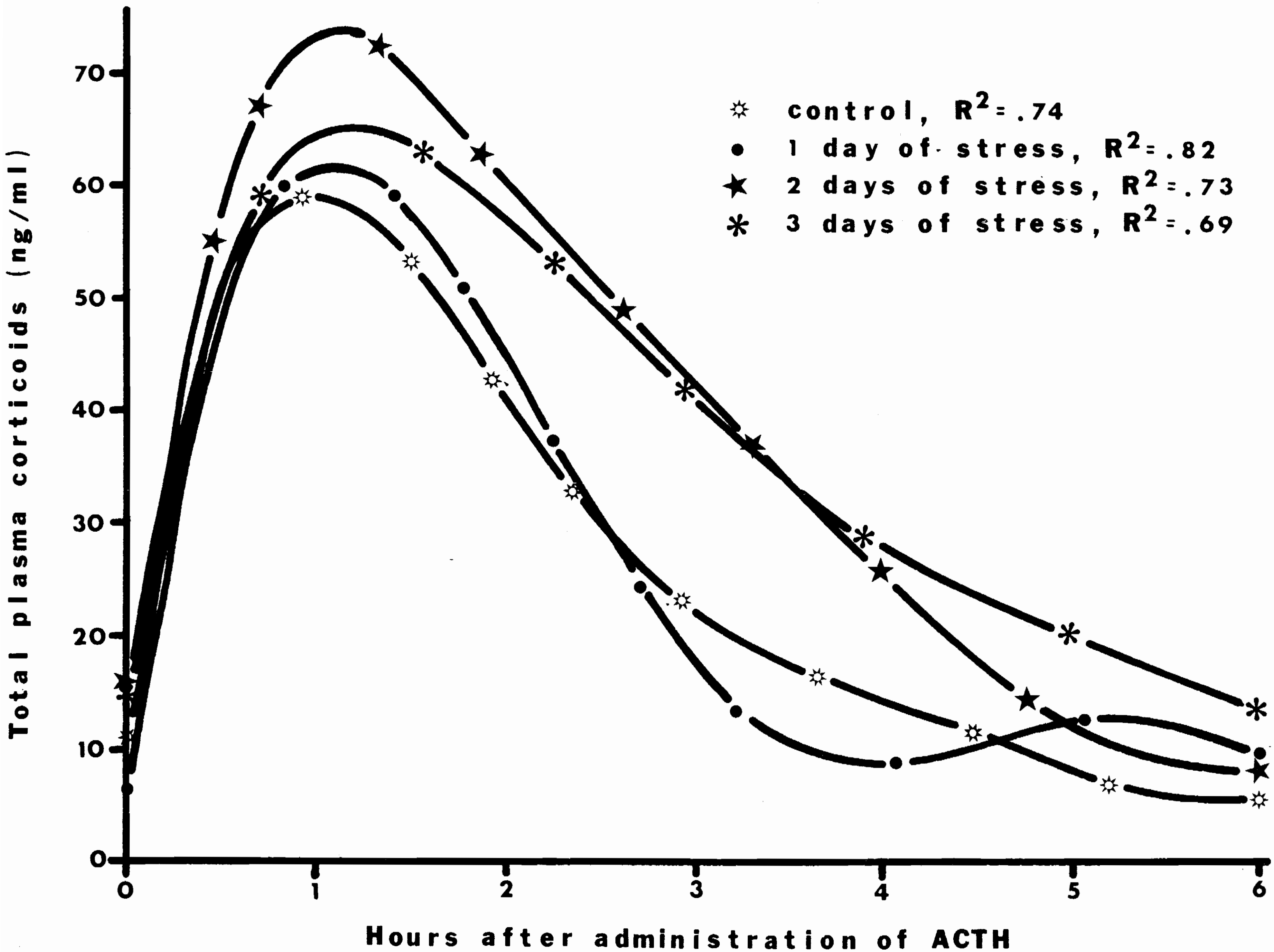


Figure 7. Total plasma corticoid concentrations in response to injection of 200 IU ACTH, Experiment II.

Table 9. Daily milk production of control and stressed cows in Experiment II.

	Control	Stressed	
	Adrenal response tested		Base
No. of observations	4	12	12
Avg. daily prod. -3, -2, -1, days of stress	24.6±3.2[a]	21.3±3.2	29.1±6.7
Avg. daily prod. day 1 of stress (kg)	24.1±4.3	21.8±2.8	28.7±7.0
Avg. daily prod. day 2, 3, 4 of stress (kg)	24.2±4.6	21.8±3.6	28.9±6.8

[a]Standard deviation of observations.

in commercial herds. Research cattle are frequently reshuffled from one study to the next and have considerable human contact. The older cows in the test group of Exp. I were probably well acquainted with many cows in the base group. Test cows in Exp. II, being first lactation, had not had the opportunity to interact with any of the base cows as adults. This probably contributed to greater behavioral stress.

Corticoid response to ACTH has been used in studies with long term stressors. These data, however, indicate that only 48 hours are required for the adrenals of dairy cattle to mobilize for a great increase in corticoid synthesis. Stress from shifting cows probably elicited the trend for increased adrenal response in Exp. I since these cows were only mildly crowded for space. Basal concentrations of corticoids (pre-ACTH) varied greatly and were not adequate measures of changes in the adrenal. Sampling pre-ACTH in these experiments however, was instituted primarily to allow cows to acclimate to the sampling procedure. These samples may reflect the handling required to prepare cows for sampling.

Failure of the somatic cell and microbial data to be meaningfully altered supports the view that stress per se does not alter somatic cell counts. This does not mean that stress may not be a factor in the etiology of mastitis. Increased social competition among chickens increases resistance to Staphylococcus aureus (31), Streptococcus faecalis (32) and Escherichia coli (33). All three organisms can cause elevated somatic cell counts, though Staph. aureus is the major cause of mastitis in many herds (63).

We would expect a 5 percent decrease in milk production after combining cows to form a new group (2, 10, 74). A similar decrease in milk production in both experiments would have been reported had the control cows not also decreased in production.

SUMMARY AND CONCLUSIONS

Twelve dairy cows were used to determine behavior with varying numbers of free stalls and length of feed trough. Available free stalls were 1, .83, .67, .50, .33 per cow. Linear feed trough was .5, .4, .3, .2, .1 m per cow changed at 7-day intervals maintaining 1 free stall per cow. Cow behavior and locations were quantified by time-lapse photography at 1-minute intervals during the last 3-days of each treatment. Current spatial recommendations for dairy cattle can be greatly reduced. Behavior was altered only when less than .67 free stalls or .2 m of linear feed trough was available per cow. Minimum stalls needed per cow without altering daily free stall usage = [14.2 hours (average use)] / [hours per day that free stalls are available to the herd X .93 (maximum efficiency before crowding)]. In this study, .71 stalls per cow is the calculated minimum. The efficiency of use in other facilities could decline depending on total exercise area, season, and physical layout of free stalls. Linear feed trough of .2 m appears adequate to ensure desired amount of eating time when individuals have access to food in the trough 21 hours per day. Estimated individual dry matter intakes were the same at .5 m and .2 m of feed trough per cow.

It has long been accepted that social status plays an important role in activity and performance of individual cows (55). This notion persists even though social status, as determined by observations of agonistic behavior, is not correlated with production in dairy cattle. The findings of this dissertation refute the importance of a cow's dominant status. Data from three experiments were used to examine dominance values as measures of a socially mediated priority of access to resources. A least squares procedure, which regressed for numbers of observations, was used for obtaining dominance values from observations of agonistic behavior. Behavioral data were quantified by time-lapse photography at 1-minute intervals. Multiple regressions were used to determine the importance of dominance value, age, body weight, daily milk production, mature equivalent milk production and percentage milk fat, with their quadratic combinations, in predicting access to resources. Dominance was of little importance relative to production variables in describing access to a limited amount of grain, concentrate, or hay. Likewise, production oriented variables were more important in describing access to free stalls or the feed trough at 5 levels of competition. Individual feed intakes of group fed dairy cattle at 2 levels of competition were described predominantly by production variables. It appears that the aggressive behavior one observes has little utility in domestic dairy cattle. The momentary increase in conflict observed when strange cows are introduced into a group may not be due to establishing a dominance hierarchy, but more to individuals learning who the aggressive cows are by trial and then avoiding them.

The literature and dairymen have reported temporary drops in milk production when cows are moved to a strange group. The effects on adrenal activity and production of the physical and psychological stress resulting from the disturbed aggressive order were examined. Adrenal response to 200 IU adrenocorticotrophic hormone (ACTH) was determined by quantifying plasma corticosteroids in two groups of lactating Holstein cows. One group of 18 cows received ACTH via jugular catheter 0, 2, or 9 days after introduction to an established group in restricted space. Differences in total plasma corticoids (area under curve) in response to ACTH were not statistically different although corticoid response 2 and 9 days of stress tended to be greater than day 0. A second group of 16 cows received ACTH at 0, 1, 2, or 3 days after introduction to a new group and crowding. Mean corticoid responses to ACTH (area under the curve, ng/ml ± SD) were 161.6 ± 12.6, 158.2 ± 28.2, 227.7 ± 32.2, and 229.9 ± 40.3 for cows injected days 0, 1, 2, and 3, respectively, days 2 and 3 being greater ($P<.05$) than day 0. Plasma corticoid concentration is an effective means of detecting adrenal alteration in response to stress. Due to the large number of CPB assays that must be made to document a response curve however, its' use is limited. Bacteriological status of quarter milk samples were not changed by stress. In non-infected quarters of 24 stressed cows, 8 quarters increased above 150,000 cells per ml while 5 quarters decreased below 150,000 cells per ml of milk after 4 days of stress when compared to 2 and 6 days prior stress. Milk production did not change relative to controls.

REFERENCES

1 Akinyele, I. O., and S. L. Spahr. 1975. Stage of lactation as a criterion for switching cows from one complete feed to another during early lactation. J. Dairy Sci. 58:917.

2 Arave, C. W. and J. L. Albright. 1976. Social rank and physiological traits of dairy cows as influenced by changing group membership. J. Dairy Sci. 59:974.

3 Arave, C. W., J. L. Albright, and C. L. Sinclair. 1974. Behavior, milk yield, and leucocytes of dairy cows in reduced space and isolation. J. Dairy Sci. 57:1497.

4 Arave, C. W., J. L. Walters, C. H. Mickelson and R. C. Lamb. 1976. The effect of prepartum exercise on five blood traits of cows. Paper 101, presented at 71st annual meeting ADSA. North Carolina State Univ., Raleigh.

5 Babson Bros. Dairy Research Service. 1970. The way cows will be milked on your dairy tomorrow. 7th ed. Babson Bros. Co., Oak Brook, Il.

6 Baird, G. D. and R. J. Heitzman. 1970. Gluconeogenesis in the cow: The effects of a glucocorticoid on hepatic intermediary metabolism. Biochem. J. 116:865.

7 Barr, A. J. and J. H. Goodnight. 1972. A user's guide to the statistical analysis system. Students supply stores, North Carolina State Univ., Raleigh.

8 Beilharz, R. G., D. F. Butcher and A. E. Freeman. 1966. Social dominance and milk prodcution in Holsteins. J. Dairy Sci. 49:887.

9 Boehlje, M. and R. Herr. 1976. What a new dairy barn would cost you. Hoard's Dairyman, May 25:639.

10 Brakel, W. J. and R. A. Leis. 1976. Impact of social disorganization on behavior, milk yield, and body weight of diary cows. J. Dairy Sci. 59:716.

11 Brambell, F. W. R. 1965. Report of the technical committee to inquire into the welfare of animals kept under the intensive livestock husbandry system. Command paper 2836. London: H. M. S. O. p. 10.

12 Breland, K. and M. Breland. 1966. Animal behavior. Macmillian Company, N. Y.

13 Brobeck, J. R. Ed. 1973. Best & Taylor's Physiological Basis of Medical Practice. The Williams and Wilkins Co., Baltimore.

14 Burghardt, W. G. and C. E. Coppock. 1974. Behavioral and feeding responses of lactating dairy cows fed a complete ration in free stalls. Unpublished mimeo. Cornell Univ., Ithaca, N.Y.

15 Chandler, P. T. and H. W. Walker. 1972. Generation of nutrient specifications for dairy cattle for computerized least cost ration formulation. J. Dairy Sci. 55:1741.

16 Christison, G. I. and H. D. Johnsor. 1972. Cortisol turnover in heat stressed cattle. J. Anim. Sci. 35:1005.

17 Collis, K. A. 1976. An invertigation of factors related to the dominance order of a herd of dairy cows of similar age and breed. App. Anim. Ethology 2:167.

18 Cope, C. L. 1972. Adrenal steroids and disease. J. P. Lippincott Co., Philadelphia.

19 Coppock, C. E., C. H. Noller, B. W. Crowl, C. D. McLellon and C. L. Rhykerd. 1972. Effect of group versus individual feeding of complete rations on feed intake of lactating cows. J. Dairy Sci. 55:325.

20 Craig, J. V., D. K. Biswas and A. M. Guhl. 1969. Agonistic behaviour influenced by strangeness, crowding and heredity in female domestic fowl (Gallus Gallus). Anim. Behav. 17:498.

21 Crampton, E. W. and L. E. Harris. 1969. Applied Animal Nutrition, 2nd Ed. W. H. Freeman and Co., San Francisco. p. 119.

22 Dickson, D. P., G. R. Barr, L. P. Johnson and D. A. Wieckert. 1970. Social dominance and temperament of Holstein cows. J. Dairy Sci. 53:904.

23 Dickson, D. P., G. R. Barr and D. A. Wieckert. 1967. Social relationships of dairy cows in a feed lot. Behav. 29:195.

24 Dvorak, M. 1971. Plasma 17-hydroxycorticosteroid levels in healthy and diarrhoeic calves. Brit. Vet. J. 127:372.

25 Ely, L. O. and R. L. Baldwin. 1976. Effects of adrenalectomy upon ruminant liver and mammary function during lactation. J. Dairy Sci. 59:491.

26 Fox, T. W. and J. T. Clayton. 1960. Population size and density as related to laying house performance. Poult. Sci. 39:896.

27 Friend, T. H. and C. E. Polan. 1974. Social rank, feeding behavior, and free stall utilization by dairy cattle. J. Dairy Sci. 57:1214.

28 Goering, H. K. and P. J. Van Soest. 1970. Forage Fiber Analyses. Ag. Handbook No. 379, A. R. S., U. S. D. A.

29 Goetsch, D.D., L. E. McDonald and G. Odell. 1957. The effect of four synthetic corticosteroids on leukocytes, blood glucose, and plasma sodium and potassium in the cow. Amer. J. Vet. Res. 20:697.

30 Gross, W. B. 1970. Corticosterone and ACTH as treatments for Escherichia coli infection in chickens. Poult. Sci. 49:1256.

31 Gross, W. B. and G. Colmano. 1967. Further studies on the effects of social stress on the resistance to infection with Escherichia coli. Poul. Sci. 46:41.

32 Gross, W. B. and G. Colmano. 1969. The effect of social isolation on resistance to some infectious diseases. Poul. Sci. 48:514.

33 Gross, W. B. and H. S. Siegel. 1965. The effect of social stress on resistance to infection with Escherichia coli and Mycoplasma gallisepticum. Poul. Sci. 44:998.

34 Guhl, A. M. and F. W. Atkeson. 1959. Social orgainzation in a herd of dairy cows. Trans. Kansas Academy of Science 62:80.

35 Guhl, A. M., J. V. Craig and C. D. Mueller. 1960. Selective breeding for aggressiveness in chickens. Poul. Sci. 39:970.

36 Gwazdauskas, F. C. 1974. Interrelationships of certain thermal and endocrine phenomena and reproductive function in the female bovine. Ph. D. Dissertation. Univ. of Florida, Gainesville.

37 Gwazdauskas, F. C., W. W. Thatcher and C. J. Wilcox. 1972. Adrenocorticotropin alteration of bovine peripheral plasma concentrations of cortisol, corticosterone, and progesterone. J. Dairy Sci. 55:1165.

38 Gwazdauskas, F. C., W. W. Thatcher and C. J. Wilcox. 1973. Physiological, environmental, and hormonal factors at insemination which may affect conception. J. Dairy Sci. 56:873.

39 Hacker, R. R., J. L. Albright, R. W. Taylor and D. L. Hill. 1969. Cow preferences for permanent bedding materials supported by different foundations in a free stall slotted floor barn. J. Dairy Sci. 52:918 (Abstr.).

40 Hafez, E. S. E. (Ed.). 1962. The Behavior of Domestic Animals. Williams and Wilkins Co., Baltimore.

41 Hale, E. B. 1969. Domestication and evolution of behavior. In E. S. E. Hafez (Ed.). The Behavior of Domestic Animals. Williams and Wilkins Co., Baltimore.

42 Head, H. H., W. W. Thatcher, C. J. Wilcox and K. C. Bachman. 1976. Effect of a synthetic corticoid on milk yield and composition and on blood metabolites and hormones in dairy cows. J. Dairy Sci. 59:880.

43 Heald, C. W., A. M. Lineen, M. A. Remsburg, G. M. Jones and W. N. Patterson. 1976. Use of the Fossomatic somatic cell counter. Paper 60, presented at 71st annual meeting ADSA. North Carolina State Univ., Raleigh.

44 Henderson, C. R. 1973. Sire evaluation and genetic trends. Page 10 in Proceedings of the Animal Breeding and Genetics Symposium in honor of Dr. Jay L. Lush. ASAS, ASDA, Chanpaign, Il.

45 Hill, F. W. and D. L. Anderson. 1958. Comparison of metabolizable energy and productive energy determinations with growing chicks. J. Nutr. 64:587.

46 Hinde, R. A. 1970. Animal Behavior: A Synthesis of Ethology and Comparative Psychology. 2nd Ed. McGraw Hill, New York.

47 Hudson, S., M. Mullord, W. G. Whittlestone and E. Payne. 1975. Diurnal variations in blood cortisol in the dairy cow. J. Dairy Sci. 58:30.

48 Hudson, S., M. Mullford, W. G. Whittlestone and E. Payne. 1976. Bovine plasma corticoids during parturition. J. Dairy Sci. 59:744.

49 Hyppola, K. and O. Hasunen. 1970. Dry matter and energy standards for dairy cows. Acth Agralia Fennica, 116:1.

50 Jones, G. M. 1972. Chemical factors and their relation to feed intake regulation in ruminants: A review. Canadian J. of Anim. Sci. 52:207.

51 Kaiser, H. F. 1959. Contributions to the method of paired comparisons. (Mimeo.). Bureau of Educational Research, Univ. of Il., Champaign-Urbana.

52 Keeley, K. 1962. Prenatal influence on behavior of offspring of crowded mice. Science, 135:44.

53 Klopter, P. H. and J. H. Hailman. 1967. An Introduction to Animal Behavior. Prenticw-Hall, Englewood Cliffs, N. J.

54 Kronfeld, D. D. and P. E. Hartmann. 1973. Glucose redistribution in lactating cows given dexamethasone. J. Dairy Sci. 56:903.

55 Lamb, R. C. 1976. Relationship between cow behavior patterns and management systems to reduce stress. J. Dairy Sci. 59:1630.

56 Larsen, H. J. 1963. Feeding habits of grazing and green feeding cows. J. Anim. Sci. 22:1134.

57 Lee, J. A., J. D. Roussel and J. F. Beatty. 1976. Effect of temperature - season on bovine adrenal cortical function, blood cell profile, and milk production. J. Dairy Sci. 59:104.

58 Lewis, R. C. and J. D. Johnson. 1954. Observations of dairy cow activities in loose-housing. J. Dairy Sci. 37:269.

59 McCullough, M. W. 1962. Use of digestible dry matter by dairy cows group-fed silage. J. Dairy Sci. 45:1107.

60 McPhee, C. P., F. McBride and J. W. James. 1964. Social behavior of domestic animals. II. Steers in small yards. Anim. Prod. 6:9.

61 Midwest Plan Service. 1971. Dairy Housing and Equipment Handbook. Iowa State Univ., Ames.

62 Moseley, J. E., C. E. Coppock and G. B. Lake. 1976. Abrupt changes in the forage-concentrate ratios of complete feed fed ad lititum to dairy cows. J. Diary Sci. (In Press).

63 National Mastitis Council, Inc. 1969. Microbiological procedures for the diagnosis of bovine mastitis. University of New Hampshire Press, Durham.

64 Ortman, L. L. and J. V. Craig. 1968. Social dominance in chickens modified by genetic selection - physiological mechanisms. Anim. Behav. 16:33.

65 Paape, M. J., D. W. Carroll, A. J. Kral, R. H. Miller and C. J. Desjardins. 1974. Corticosteroids, circulating leucocytes, and erythrocytes in cattle: Diurnal changes and effects of bacteriologic status, stage of lactation, and milk yield on response to adrenocorticotropin. Amer. J. Vet. Res. 35:355.

66 Paape, J. J., A. J. Kral, C. Desjardins, W. D. Schultze and R. H. Miller. 1973. Failure of either corticosteroids, or ACTH to increase the leukocyte concentration in milk. Amer. J. Vet. Res. 34:353.

67 Paape, M. M., W. D. Schultz and R. H. Miller. 1973. Leukocytic response to adrenocorticotrophic hormone as influenced by the infectious history of the mammary gland. J. Dairy Sci. 56:733.

68 Panaretto, B. A, and M. R. Vickery. 1970. The rates of cortisol entry and clearance in sheep before and during their exposure to a cold, wet environment. J. Endocrinol. 47:273.

69 Reichmuth, J. 1975. Somatic cell counting - interpretation of results. Proc. I. D. F. Seminar on mastitis control, Reading Univ., Eng. April, 11:93.

70 Reid, R. L. 1962. Studies on the carbohydrate metabolism of sheep. XV. The adrenal response to climatic stress of cold, wind and rain. Aust. J. Agr. Res. 13:269.

71 Reid, R. L. and S. C. Mills. 1962. Studies of the carbohydrate metabolism of sheep. XIV. The adrenal response to psychological stress. Aust. J. Agr. Res. 13:282.

72 Rhynes, W. E. and L. L. Ewing. 1973. Plasma corticosteroids in Hereford bulls exposed to high ambient trmperatures. J. Anim. Sci. 36:369.

73 Robertson, W. G., J. P. Mixner, W. W. Bailey and H. D. Lennon, Jr. 1958. Effects of certain acute stress conditions on the plasma levels of 17-hydroxycorticosteroids and protein-bound iodine in dairy dattle. J. Dairy Sci. 41:302.

74 Schein, M. W. and M. H. Fohrman. 1955. Social dominance relationships in a herd of dairy cattle. Anim. Behav. 3:45.

75 Schein, M. W., C. E. Hyde and M. H. Fohrman. 1955. The effect of physiological disturbances on milk production in dairy cattle. Page 79 in Proc. Ass. Southern Agr. Workers 52nd Convention, Louisville, K. Y. (Abstr.).

76 Schmisseur, W. E., J. L. Albright, W. M. Dillon, E. W. Kehrederg and W. H. M. Morris. 1966. Animal behavior responses to loose and free stall housing. J. Dairy Sci. 49:102.

77 Sebranek, J. G., D. N. Marple, R. G. Cassens, E. J. Briskey and L. L. Katenschmidt. 1973. Adrenal response to ACTH in the pig. J. Anim. Sci. 36:41.

78 Selye, H. 1963. A syndrome produced by diverse nocuous agents. Nature 138:32.

79 Selye, H. 1973. The evolution of the stress concept. Am. Scientist. 61:692.

80 Shayanfar, F., H. H. Head, C. J. Wilcox and W. W. Thatcher, 1975. Adrenal responsiveness in lactating Holstein cows. J. Dairy Sci. 58:870.

81 Siegel, P. B. and R. H. Coles. 1958. Effects of floor space on broiler performance. Poultry Sci. 37:1243.

82 Siegel, H. S. 1959. Egg production characteristics and adrenal function in White Leghorns confined at different floor space levels. Poult. Sci. 38:893.

83 Smith. V. G., E. M. Convey and L. A. Edgerton. 1972. Bovine serum corticoid response to milking and extroceptive stimuli. J. Dairy Sci. 55:1170.

84 Smith, R. D., W. Hansel and C. E. Coppock. 1975. Plasma adrenocorticoid response to corticotropin in dairy cattle fed high silage diets. J. Dairy Sci. 58:1708.

85 Southwick, C. H. 1971. The biology and psychology of crowding in man and animals. The Ohio J. of Sci. 71:65.

86 Swanson, L. V. and R. E. Lind. 1976. Lactational response of dairy cows to oral administration of a synthetic glucocorticoid. J. Dairy Sci. 59:614.

87 Syme, G. J. 1974. Competitive orders as measures of social dominance. Anim. Behav. 22:931.

88 Syme, Lesley A., G. J. Syme, T. G. Waite and A. J. Pearson. 1975. Spatial distribution and social status in a small herd of dairy cows. Anim. Behav. 23:609.

89 Thaxton, P., C. R. Sadler and Bruce Glick. 1968. Immune response of chickens following heat exposure or injections with ACTH. Poul. Sci. 47:264.

90 Thomas, J. W., L. D. Brown and R. W. Emery. 1970. Corn silage compared to alfalfa hay for milking cows fed various levels of grain. J. Dairy Sci. 53:342.

91 Trimberger, G. W., H. F. Tyrrell,D. A. Morrow, J. T. Reid, M. J. Wright, W. F. Shipe, W. G. Merrill, J. K. Loosli, C. E. Coppock, L. A. Moore and C. H. Gordon. 1972. Effects of liberal grain feeding on health, reproductive efficiency, economy of milk production and other related responses in dairy cows. New York Food and Life Sci. Bull. #8.

92 Vandersall, J. H. and R. W. Hemken. 1970. All corn silage forage program: Effect on consecutive lactations and dry matter consumption. Proc. Md. Nutr. Conf., p. 90.

93 Venkataseshu, G. K. and V. L. Estergreen, Jr. 1970. Cortisol and corticosterone in bovine plasma and the effect of adrenocorticotropin. J. Dairy Sci. 53:480.

94 Wagner, W. C. 1969. Suckling and ovarian activity in the cow. Amer. J. Vet. Med Assn. 154:1395.

95 Wagner, W. C. and S. L. Oxenreider. 1972. Adrenal function in the cow. Diurnal changes and the effects of lactation and neurohypophyseal hormones. J. Anim. Sci. 34:630.

96 Wegner, T. N., J. D. Schuh, F. E. Nelson and G. H. Stott. 1976. Effect of stress on blood leukocyte and milk somatic cell counts in dairy cows. J. Dairy Sci. 59:949.

97 Wegner, T. N. and G. H. Stott. 1972. Serum minerals, leukocyte profiles, and plasma corticoids in dairy heifers after an injection of corticotropin. J. Dairy Sci. 55:1464.

98 Whittlestone, W. G., R. Kilgour, H. deLangen and G. Duirs. 1970. Behavioral stress and the cell count of bovine milk. J. Milk Food Technol. 33:217.

APPENDIX

Appendix Table 1. β values for regression equations with time in free stalls at various levels of competition as the dependent variable.

Independent variable	No. of free stalls per cow					Units
	1.0	.83	.67	.50	.33	
Intercept	26.73	-22.95	-120.6	-22.55	-78.62	
DV	-.7468	-.6812		-.6989		
DV^2	.9840	1.157	.6682	.9429		
Age	.1139	.1143	.1620	.1110	.0710	months
Age^2	-.00095	.00077	-.00082	-.00076	-.00029	
Body weight	-.0744	.1063	.2131	.1091	.1490	lbs X 10
Body $weight^2$	.00029	-.00051	-.0010	-.00051	-.0007	
Daily prod.	.0809		-.0240		-.1046	lbs
Daily $prod.^2$	-.00056		.000013		.00056	
ME milk prod.		.3960	1.220	.3624	.6766	lbs X 100
ME milk $prod.^2$		-.0010	-.00315	-.00096	-.00175	
% milk fat	-15.14	-11.97		-10.55	5.398	%
% milk fat^2	2.084	1.577	-.1451	1.377	-.8372	

Appendix Table 2. β values for regression equations with time at the feed trough at various levels of competition as the dependent variable.

Independent variable	Meters of trough per cow					Units
	.5	.4	.3	.2	.1	
Intercept	-13.06	-79.73	-19.60	-2.260	-5.087	
DV	.415	-.1600	.2861	.202		
DV^2	-.722	.555	-.352	-.144	.134	
Age	-.0426		.0087	-.0405	.00148	months
Age^2	.00035	.00022		.0003	-.000018	
Body weight		.2969	-.00685		-.00076	lbs X 10
Body $weight^2$		-.00133				
Daily prod.	-.0825	-.2662	-.0774	-.0313	-.0227	lbs
Daily $prod.^2$	.00055	.00208	.00045	.00033	.00023	
ME milk prod.	.0399	.7759	.1519	.0328	.08198	lbs X 100
ME milk $prod.^2$	-.000095	-.00205	-.00038	-.000094	-.00022	
% milk fat	7.310	-.4347	4.635	.7920	-.938	%
% milk fat^2	-.996		-6.403	-.1143	.1200	

Appendix Table 3. β values for regression equations with time at the feed trough or dry matter (DM) intake at two levels of competition as the dependent variable.

Independent variable	Time at trough for meters of trough/cow		DM intake for meters of trough/cow		Units
	.5	.25	.5	.25	
Intercept	28850.	26450.	-520.4	-73.06	
DV	-164.3	54.78	-2.111		
DV^2		-49.53	-35.86	2.3219	
Age	29.73	34.90	.9699	-.4464	months
Age^2	-.2737	-.3166	-.00675	.00531	
Body weight	-108.19	-76.59	2.4187		lbs X 10
Body $weight^2$	.4885	.3226	-.00825	.00028	
Daily prod.			-.7188	.6561	lbs
Daily $prod.^2$	-.0879			-.00588	
ME milk prod.	-77.71	-123.8		-.6714	lbs X 100
ME milk $prod.^2$	.2276	.3383	.000227	.00159	
% milk fat	-8036.	-5262.	208.94	70.131	%
% milk fat^2	1007.1	631.05	-27.866	-8.138	

Appendix Table 4. β values for regression equations with time at the feed trough or in free stalls as the dependent variable.

Independent variable	First 15 min post-feeding concentrate	First 30 min post-feeding hay	At trough per day	In stalls per day	Units
Intercept	-895.3	-1914.9	5951.	6207.5	
DV	17.87			52.61	
DV^2	20.67		243.7	-322.9	
Age	19.00	-16.78	-183.9		years
Age^2	-1.5199	1.3369	12.37		
Body weight	.0329	.8103	-1.657	6.040	lbs
Body weight^2		-.00024	.00074	-.00249	
Daily prod.	6.599	19.76	-38.78	-74.374	lbs
Daily prod.^{2}	-.06681	-.2022	.4442	.8418	
ME milk prod.	.07568	.0216	-.3395	-.2909	lbs
ME milk prod.^{2}	-.0000021	-.00000071	.0000091	.0000067	
% milk fat		448.8	151.16	-1980.6	%
% milk fat^2	1.8429	-65.38		272.64	

Appendix Table 5. Chromic oxide and acid detergent lignin(ADL) determinations used to estimate individual dry matter intake.

Cow	Cr_2O_3 in feces (Mg/gm)[a]	% ADL	Kg dry matter intake
	.5 meters feed trough per cow[b]		
457	2.40	10.39	23.3
569	2.33	8.53	19.8
680	2.48	8.13	17.6
719	2.38	8.53	19.3
732	3.54	9.45	14.3
735	2.43	8.43	18.7
744	2.32	8.72	20.3
749	2.25	8.87	21.2
829	3.29	8.78	14.4
840	3.02	10.62	19.0
842	3.30	10.88	17.8
857	2.35	9.19	21.1
	.25 meters feed trough per cow[c]		
457	2.05	8.67	23.5
569	2.11	8.47	22.3
680	2.17	8.00	20.5
719	2.23	8.13	20.3
732	2.18	8.40	21.4
735	2.55	7.77	16.9
744	2.35	8.04	19.0
749	1.85	7.25	21.7
829	3.68	8.49	12.8
840	3.04	10.45	19.1
842	2.46	8.67	19.6
857	2.46	8.47	19.2

[a]Ten grams administered orally twice daily.

[b]Feed contained 3.71% ADL, 57.6% DM, 14.7% CP, 17.4% CF.

[c]Feed contained 3.60% ADL, 58.7% DM, 13.2% CP, 19.5% CF.

Appendix Table 6. Plasma total corticoids[a] in response to 200 IU ACTH administration, Experiment I.

Cow	Days of stress	Hours post-ACTH administration -2	-1	0	.25	.5	.75	1	2	3	4	5	6	7	8	9	10	11	12	Area of curve[b]
									Replicate I											
505	2	10.6	9.7	6.5	41.2	57.9	55.4	57.7	59.8	39.8	24.8	9.4	3.3	1.9	4.7	5.9	4.7	9.1	21.2	211.0
755	2	10.8	15.6	4.9	47.0	74.5	84.1	71.4	57.9	49.9	29.5	15.6	4.9	3.3	5.9	1.9	2.6	6.8	8.5	251.8
955	2	5.6	2.1	8.2	53.1	71.6	85.7	74.5	59.8	70.1	24.7	18.8	6.3	5.1	6.4	3.0	2.8	8.1	2.3	276.7
588	9	17.8	2.2	10.9	18.5	53.5	55.3	67.6	51.5	35.9	21.8	8.2	3.8	2.5	3.0	1.8	1.7	2.4	3.7	194.7
829	9	11.8	6.1	9.1	66.4	66.3	76.9	66.7	55.7	43.7	29.0	15.9	6.3	4.2	6.0	2.8	1.3	1.5	2.3	242.7
958	9	11.5	4.3	13.6	52.4	58.2	58.0	51.2	46.8	45.1	30.9	21.5	12.6	5.8	4.6	2.6	1.9	2.2	1.2	226.4
400	0	2.4	10.8	22.9	37.6	51.4	51.4	49.2	51.9	29.7	22.2	9.8	7.8	3.2	5.7	3.1	4.1	1.4	1.0	186.3
660	0	2.6	9.5	2.6	41.7	78.8	48.9	52.1	49.4	27.1	33.8	9.1	3.9	2.1	5.6	3.6	1.0	2.2	12.9	196.6
941	0	4.6	12.2	1.7	45.2	72.6	58.5	70.5	63.0	32.8	16.5	12.8	2.6	4.0	3.2	3.0	1.8	1.4	3.6	214.7
									Replicate II											
667	2	12.2	2.8	8.0	108.7	59.9	68.3	71.1	59.1	40.8	21.1	14.4	3.5	7.7	4.2	2.0	2.0	2.5	1.7	241.8
867	2	12.5	3.8	3.0	23.1	19.6	48.4	53.4	83.7	27.8	21.6	15.6	18.5	5.8	2.8	1.2	3.1	2.3	1.2	183.1
973	2	2.2	1.6	3.0	47.6	42.1	48.7	42.4	51.6	31.4	16.5	15.9	12.5	5.1	3.9	2.2	1.4			183.1
661	9	11.6	2.8		49.5	50.3	39.5	31.2	27.6	29.9	18.0	23.9	19.1	13.2	7.3	4.4	1.8	2.0	1.1	164.2
830	9	16.1	3.1	5.6	42.0	37.1	52.7	32.4	70.2	38.5	33.7	34.5	24.3	7.9	8.8	5.8	4.4	1.1	.8	242.9
954	9	14.2	2.6	12.6	52.4	49.1	68.9	59.3	54.8	56.3	29.2	28.7	13.7	11.0	4.8	3.0	1.6		1.3	257.1
617	0	6.9	2.4	8.4	56.9	42.9	47.6	54.2	50.3	33.7	26.7	21.6	6.4	5.1	5.2	2.6	2.8	2.8	.7	207.3
884	0	5.0	2.4	12.2	52.2	78.1	80.7	85.3	44.9	34.9	29.5	18.7	9.2	9.8	2.9	3.1	1.3	.6		240.2
946	0	8.2	2.3	3.2	28.8	42.1	47.9	58.8	42.7	15.5	19.2	12.9	8.9	7.7	4.4	3.2	2.2	.7		157.1

[a]Mean of duplicates.

[b]Hours 0 through 6.

Appendix Table 7. Plasma total corticoids[a] in response to 200 IU ACTH administration, Experiment II.

		Hours post-ACTH administration																	
Cow	Days of stress	-2	-1	0	.25	.5	.75	1	1.25	1.50	1.75	2	2.25	2.50	3	3.50	4	5	6	Area of curve[b]
922	1		1.0	5.8	35.5	44.9	49.1	69.1	50.4	54.6	53.7	30.9	24.9	22.8	6.8	6.3		2.7		126.3
949	1	1.9	3.9	7.9	54.9	54.4	69.5	67.8	37.0	52.9	70.2	53.6	68.8	13.0	21.6	23.0	19.6	14.2	11.6	194.4
969	1	5.7	1.9		32.9	53.5	51.6	46.4	69.3	66.3	62.9	35.6	37.7	12.7	16.2	14.0	13.8	11.9	7.9	160.3
913	1	4.7	4.6	4.1	51.8	41.6	38.8	58.9	87.1	63.6	58.1	49.8	29.7	20.3	7.8	8.3	4.9	9.2	6.2	152.0
908	2	5.4	5.5	7.2	55.0	54.8	62.2	78.8	48.8	73.3	70.1	45.9	27.6	19.5	62.5	18.3	13.8	9.3	7.5	201.2
933	2	2.3	5.1	5.5	51.8	60.4	73.8	78.9	51.8	67.1	34.9	49.5	23.0	58.6	41.9	15.6	14.4	9.1	9.0	198.6
956	2	38.9	6.6	1.4	57.2	81.5	31.2	62.2	56.0	77.3	77.9	68.6	96.1	55.1	58.3	31.6	25.9	16.2	10.1	258.5
989	2	4.9	22.1	7.9	57.4	69.7	75.1	71.1	68.9	76.5	74.5	67.8	76.6	57.0	48.2	26.3	17.5	15.4	10.1	252.6
939	3	8.1	6.7	6.7	81.7	67.7	61.4	81.7	72.9	69.5	72.7	66.7	75.5	62.1	60.9	24.8	21.4	28.0	10.7	278.8
961	3	10.7	8.9	12.3	74.4	55.0	42.8	58.5	104.6	53.6	57.7	48.0	56.6	42.8	14.5	12.8	14.9	11.4	8.5	195.9
979	3		1.8	8.6	19.6	42.8	54.3	52.0	37.4	64.8	46.6	39.2	46.5	46.5	36.9	30.4	30.6	13.4	17.1	197.9
981	3	3.8	14.8	8.2	51.7	30.4	60.9	45.3	54.2	70.3	59.5	62.4	56.5	59.0	38.7	48.6	28.3	29.6	13.4	247.1
932	0	10.4	2.8	2.8	38.8	75.5	63.1	62.2	74.4	66.1	60.1	47.9	52.9	14.9	8.6	15.4	8.7	11.6	3.9	173.3
944	0	11.7	12.6	4.9	146.2	86.6	58.8	76.6	86.7	81.2	56.2	66.7	81.4	49.0	44.3	34.0	17.2	13.1	9.7	274.1
970	0	25.5	1.9	12.2	33.9	27.9	58.3	52.9	36.4	36.7	42.8	36.9	46.5	34.3	14.7	11.0	17.6	10.2	9.0	148.2
986	0	3.6	2.6	6.2	42.8	81.5	34.1	46.2	41.3	48.6	49.5	44.5	24.1	39.2	30.0	23.9	12.6	5.1	6.5	163.4

[a]Mean of duplicates.

[b]Hours 0 through 6.

VITA

Name: Theodore Henry Friend IV

Birth: Yonkers, New York May 4, 1948

High School Education: Lincoln High School
Yonkers, New York 1964-1966

College Education: Westminster College
Fulton, Missouri 1966-1967

Cornell University
Ithaca, New York 1967-1970
Bachelor of Science in Animal Science, 1970

Virginia Polytechnic Institute and State Univ.
Blacksburg, Virginia 1970-1977
Master of Science in Dairy Science, 1973
Doctor of Philosophy in Dairy Science, 1977

Background: The author was raised in Yonkers, New York, spending summers and weekends in rural New York. When an undergraduate, the author worked on various New York dairy farms as assistant herdsman during summer vacations. He served as Communications/Electronics Material Officer on a U. S. Navy destroyer on deployment Viet Nam from 1970-1971.

Honorary Organization: Phi Sigma

Professional Organizations: American Dairy Science Association
American Society of Animal Science
Animal Behavior Society

Theodore H. Friend

COW PERFORMANCE, ADRENAL FUNCTION, AND MILK QUALITY UNDER VARYING LEVELS OF COMPETITION

by

Theodore H. Friend

(ABSTRACT)

Twelve dairy cows were used to determine behavior with varying numbers of free stalls and length of feed trough. A least squares procedure, which regressed for numbers of observations, was adapted for obtaining dominance values. Available free stalls were 1.0, .83, .67, .50, .33 per cow. With 1.0 free stalls, linear feed trough was .5, .4, .3, .2, .1 m per cow changed at 7-day intervals. Cow behavior and locations were quantified by time-lapse photography at 1-minute intervals during the last 3-days of each treatment. Spatial recommendations for dairy cattle can be greatly reduced. Behavior was altered only when less than .67 free stalls or .2 m of linear trough was available per cow. Minimum stalls needed per cow without altering daily free stall usage = [14.2 hours (average use)]/[hours per day free stalls are available to the herd X .93 (maximum efficiency before crowding)]. Linear feed trough of .2 m appears adequate to ensure desired amount of eating time when individuals have access to feed in trough 21 hours per day. Estimated individual dry matter intakes were the same at .5 m and .25 m of trough per cow. Intake was affected by time spent eating for .25 m. In 10-variable models for various levels of competition, time spent eating, or in free stalls, and individual dry matter intake were described predominantly by production variables, not dominance values.

Adrenal responsiveness to 200 IU adrenocorticotrophic hormone (ACTH) was determined by quantifying plasma corticosteroids in two groups of lactating Holstein cows. One group of 18 cows received ACTH via jugular catheter 0, 2, or 9 days after introduction to an established group in restricted space (3.96 m^2 lot space and .67 free stalls per cow). Differences in total plasma corticoids (area under curve) in response to ACTH were not statistically different although corticoid response 2 and 9 days of stress tended to be greater than day 0. A second group of 16 cows received ACTH at 0, 1, 2, or 3 days after introduction to a new group and crowding (2.97 m^2 lot space and .5 free stalls per cow). Mean corticoid response to ACTH (area under the curve, ng/ml ± SD) were 161.6 ± 12.6, 158.2 ± 28.2, 227.7 ± 32.2 and 229.9 ± 40.3 for cows injected days 0, 1, 2, and 3 respectively, days 2 and 3 differed from day 0 ($P<.05$). Bacteriological status of quarter milk samples was not changed by stress. In non-infected quarters of 24 stressed cows, 8 quarters increased above 150,000 somatic cells/ml while 5 quarters decreased below 150,000 cells/ml of milk after 4 days of stress when compared to 2 and 6 days prior to stress. Stress did not affect milk production relative to controls.