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FACTORS AFFECTING THE SERUM IMMUNE GLOBULIN
CONCENTRATIONS OF NEWBORN CALVES

Summary of a thesis presented for the degree of
Doctor of Philosophy of the University of Glasgow
by Ian Edward Selman, B.V.M.S., M.R.C.V.S.

The work described in this thesis is concerned with studies on the factors affecting the absorption of immune lactoglobulin by newborn calves. The work is divided into four parts as follows:

Part I

Studies on Natural Suckling in Cattle
During the First Eight Hours Post Partum

In this part, the behaviour of thirty dams and their calves left together in a loose box for the first 48 hours post partum was examined. It was found that suckling did not necessarily occur within the eight-hour observation period. Certain factors which delayed the time to first suckling were defined. A significant negative correlation was found between the time to first suckling and the 48-hour serum immune globulin concentrations of the fifteen dairy calves in the series which suckled during the eight-hour observation period, thus suggesting that intestinal shutdown in calves is a progressive process initiated at birth. It was decided that the marked seasonal variation in mean serum immune globulin concentrations in dairy calves in the West of Scotland was due to the fact that calves do not, under the traditional form of calf management, get the opportunity to suckle their dams through the winter months. In short, this seasonal variation is managerial in origin.

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Part 2

Studies on Calves Allowed to Suckle Their Dams at Fixed Times Post Partum

Test weighing of thirty newborn calves allowed to suckle their own dams to satiation at fixed times post partum showed that relatively large amounts of colostrum were usually ingested. However, these high intakes did not necessarily result in high serum immune globulin concentration. Calves separated from their dams between sucklings were less efficient at absorbing immune lactoglobulin than those muzzled and left with their dams.

Part 3

Studies on the Absorption of Immune Lactoglobulin by Newborn Dairy Calves

An experimental approach was devised to rule out variations due to time of feeding, amount and immune lactoglobulin content of colostrum fed, method of feeding, environmental conditions and breed and birthweight of the experimental calves. Using this standardised approach on 120 newborn calves, it was found that within each group of ten calves, a constant uptake of immune lactoglobulin occurred, and no individual calf with a poor absorptive efficiency was found. It was found that: (a) Ayrshire calves were less efficient but more consistent absorbers of immune lactoglobulin than were Friesian-cross-Ayrshires; (b) low ambient temperatures did not depress the absorptive efficiency of calves; (c) corticosteroid therapy resulted in an increase in the absorptive efficiency of non-mothered calves; (d) a distinct and

significant linear relationship existed between the immune lactoglobulin content of colostrum fed to calves and the subsequent 48-hour serum immune globulin concentrations of those calves; and (e) dividing a colostrum meal into smaller feeds did not increase the amount of immune lactoglobulin absorbed from that colostrum. Moreover, the findings in Parts 1 and 2, that a significant negative relationship occurred between the time of the first colostrum meal and the subsequent serum immune globulin concentration, and that mothering increased the globulin absorptive efficiency of newborn calves, were also confirmed under these more critical conditions.

Part 4

Studies on Colostrum

Analysis of 100 samples of colostrum obtained from Ayrshire cows and helpers by a standard milking technique showed that the wide variations in immune lactoglobulin concentrations were individual and not seasonal. The marked variations in colostral carotene and vitamin A levels was, however, found to be both individual and seasonal in origin. No relationship was found to exist between colostral vitamin A and immune lactoglobulin concentrations.

FACTORS AFFECTING THE SERUM IMMUNE GLOBULIN CONCENTRATIONS
OF NEWBORN CALVES

by

Ian Edward Selman, B.V.M.S., M.R.C.V.S.

Thesis submitted for the Degree of
Doctor of Philosophy in the Faculty of
Veterinary Medicine, University of Glasgow

1969

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ACKNOWLEDGEMENTS

I should like to express my thanks to my supervisors, Professor W. I. M. McIntyre and Dr. E. W. Fisher, for their help and guidance during the course of these studies.

I am extremely grateful to Miss Christine Sharp and Miss Selina Gregory for carrying out so carefully the laboratory work and for assistance in the final preparation of this thesis.

My sincere thanks also go to Mrs. Julie Curtis and Mrs. Elma Sayer for typing this work and to Messrs. A. Finney and D. Irving for taking and preparing most of the photographs.

Finally, I should like to thank Mr. Edward White for the care and attention with which he tended the experimental animals and to Messrs. David French, Netherhouses, Strathaven; Ian McNicol, Kippendavie Mains, Dunblane and Alistair McNicol, Pendreich, Bridge of Allan, for supplying the adult cattle.

The work presented in this thesis was financed by the Agricultural Research Council.

Ian E. Selman
April 17th, 1969

GENERAL INTRODUCTION

GENERAL INTRODUCTION

Neonatal calf diseases and their associated mortality have long been recognised as one of the major problems encountered in rearing cattle, and high losses have been reported from many parts of the world (Lovell, 1955 and Sojka, 1965). Investigations into mortality rates have usually dealt with dairy calves, and Withers (1952-1953), Lovell (1955) and Sojka (1965), among others, have all stressed that a true assessment of the problem in dairy calves can only be made if the data for heifer calves is examined. This is due to the common practice in dairy herds of disposing of bull calves within the first few days of life (i.e. before deaths usually occur).

Several surveys have been carried out in Britain and the following table shows the mortality rates in predominantly home-reared dairy heifer calves that have been found by various workers.

TABLE I

The Mortality Rates in Newborn Dairy Heifer Calves
(from Lovell, 1955)

Date of Observations	Region	No. of Calves At Risk	Percentage Mortality	Authors
1931	Ayrshire	548	20.6	Jordan (1933)
1932 (Spring)	Ayrshire	236	23.7	Jordan (1933)
1931-1934	Ayrshire	-	20.0	Smith (1934)
1936-1937	England & Wales	12,544	5.5	Lovell & Hill (1940)
1936-1937	Scotland	1,383	11.4	Lovell & Hill (1940)
1946-1948	England & Wales	3,208	6.0	Withers (1952-1953)
1946-1948	Scotland	1,172	11.1	Withers (1952-1953)

Three investigations which have been carried out on a national scale (Lovell and Hill, 1940; Withers, 1952-1953 and Leech, Macrae and Menzies, 1968) have demonstrated quite clearly that the mortality rates in Scotland greatly exceed those of England and Wales. Moreover, extremely high losses of up to 75% on some individual farms in Scotland, are not unknown (Withers, 1952-1953). The survey carried out by Leech, et al. (1968) involved a total of 32,373 homebred calves and the results obtained suggested that regional differences in mortality rates was an over-simplification of the situation. They concluded that "regional problems appear to be special problems only in so far as the husbandry of the region is specialised." This view was also stated by Withers (1952-1953) although he considered that climatic factors also played an important role. Leech, et al. (1968) also found that the mortality rates of Ayrshire and Channel Island calves were significantly higher than those of other breeds. In view of this, it is interesting to note that almost 50% of the Ayrshire calves born on the farms they were studying were born in Scotland (38% of the total number were, in fact, born in the southwest of Scotland). It is difficult to see how these workers were able to divorce breed factors and husbandry factors in this area. It would therefore seem more likely that the high mortality rates in Scotland are due to a combination of poor calf husbandry and a breed susceptibility to neonatal disease.

All of the surveys already mentioned have found that there is a definite seasonal variation in calf mortality rates, with most calves dying in the first quarter of each year. This seasonal variation has been found to be more pronounced in Scotland than in England and Wales (Lovell and Hill, 1940, and Withers, 1952-1953).

The economic loss to the agricultural industry is very difficult to assess for a variety of reasons. Leech, et al. (1968) estimated that a total of approximately 89,000 calves die annually in Britain. However, no data is available regarding the total mortalities within different branches of the cattle-rearing industry. A high proportion of the total mortalities occur in calves (i.e. dairy heifer calves and, to a limited extent, single suckled beef calves) which would normally not be sold and which are therefore very difficult to value. An agricultural census carried out by the Department of Agriculture and Fisheries (Scotland) found that in 1968 there was a total of 329,561 dairy cows and heifers in Scotland. Using the factor, 0.80 calves per cow, calculated from an earlier survey carried out by Leech, Davis, Macrae and Withers (1960), it is possible to estimate that a total number of 263,649 parturitions occurred during 1968. Both surveys (Leech, et al., 1960, and Leech, et al., 1968) found that approximately 40% of parturitions produced heifer calves. Consequently, using the latest figures available (Withers, 1952-1953) it is possible to estimate that in Scotland alone slightly more than 14,000 dairy heifer calves die annually. Moreover, if an individual farmer sustains high losses he not only has to meet the cost of veterinary attention but also is frequently forced to either buy adult cattle or to retain every heifer calf that survives, even inferior calves, in order to keep his farm stocked to its economic capacity. These hidden losses must substantially increase the losses sustained by individual dairy farmers with a neonatal disease problem.

Various studies have shown that most calves which die do so during the first two weeks of life (Lovell and Hill, 1940, and Withers, 1952-1953). Leech, et al. (1968) found that in their series, losses in home-bred calves were highest in the first week of life and then decreased logarithmically thereafter.

It is now generally accepted that most of the neonatal deaths in calves are associated with Escherichia coli infection of one form or another (Lovell and Hill, 1940; Withers, 1952-1953; Lovell, 1955; Gay, 1965 and Sojka, 1965). In order to minimise the confusion surrounding colibacillosis of calves, Gay (1965) recommended that it should be divided into the following three forms "on clinical and bacteriological grounds and on the grounds of possible pathogenesis."

1. Enteric toxæmia
2. Colisepticaemia
3. Enteric colibacillosis (neonatal diarrhoea)

The enteric toxæmic form of colibacillosis has not yet been described in Britain although it has been reported in Canada. It is said to be a rapidly fatal condition, associated with the massive proliferation of certain specific strains of E. coli (mucoid strains) in the small intestine. There is no bacteraemia, and death is possibly due to the absorption of endotoxin (Gay, 1965).

Colisepticaemia has been found in various investigations to be a common and usually fatal condition of calves in this country (Withers, 1952-1953; Gay, Anderson, Fisher and McEwen, 1965a). It may or may not be accompanied by diarrhoea (Fisher, 1965). Moreover, colisepticaemia has been found to be the commonest cause of death in

calves which have been deprived of colostrum (Smith and Little, 1922a; Smith, 1962 and Gay, 1965). It is probably for this reason that most experimental investigations have been concerned with colisepticaemia.

All of the investigations so far mentioned have stressed the high incidence of enteric colibacillosis (diarrhoea) in the young calf. This diarrhoea may produce severe fluid, electrolyte and circulatory derangements (Dalton, Fisher and McIntyre, 1965 and Fisher, 1965), and it has been shown by Fisher (1965) that whether or not death supervenes depends on the degree of metabolic acidosis attained.

Since the work of Jensen (1893) it has been assumed by most workers that the aetiological agent involved in the diarrhoeic syndrome in calves was E. coli. Smith (1962), however, suggested that there was strong evidence that "neither E. coli nor any other bacteria were concerned in the causation of the scours." He listed the following reasons for this view: (1) the absence of incriminating serological findings; (2) the diversity of phage types of E. coli found in the small intestine of diarrhoeic calves; (3) the finding of the same types in healthy calves; (4) the observation that predominant phage type of E. coli in the faeces of a diarrhoeic calf may change several times during the diarrhoeic period and (5) the negative results in most of the transmission experiments. Sojka (1965) also stressed that E. coli was a normal inhabitant of the intestines. However, both Smith (1962) and Sojka (1965) suggested that under certain conditions, E. coli may assume a pathogenic role. It is obvious from these observations that the interpretation of post mortem findings in young calves that have apparently died from a non-septicaemic, diarrhoeic illness must be with care.

The mere fact that E. coli is found in the faeces cannot mean that the animal necessarily died of enteric colibacillosis.

Since Smith (1962) first questioned the belief that E. coli was the prime aetiological factor in neonatal diarrhoea, many suggestions have been made as to the necessary predisposing factors. It has been frequently stated that, in the colostrum-fed calf, such things as breed of calf, chilling, draughts, the stress of prolonged travel, infrequent, irregular and over-feeding of milk, badly formulated milk substitutes, cold milk, dam nutrition, over-crowding, the build up of infection in pens, vitamin deficiency and virus infections might well allow E. coli to assume pathogenic role (Payne, 1949; Withers, 1952-1953; Inglis, 1960; Fincher 1963; Edgson, 1964; Loosmore, 1964; Amstutz, 1965; Noll, 1965 and Reisinger, 1965). All, save the work of the first two authors, consisted of statements of opinion rather than scientific fact. When some of these factors were investigated, for example, with the overfeeding of milk (Walker, 1950 and Mylrea, 1966) and the feeding of cold milk (Walker, 1950), no adverse effects were noted. However, there would seem to be a breed susceptibility to neonatal diseases even in colostrum-deprived calves (Lundquist and Phillips, 1943) and a relationship was found between calf mortality and the length of time that a calf house had been occupied (Roy, Palmer, Shillam, Ingram and Wood, 1955). To further add to the confusion in the literature regarding colibacillosis, there is still the tendency of individual workers to use different names for colibacillosis (e.g. scours, white scours, calf scours, enteritis and neonatal diarrhoea). Finally, very few workers, either in the field or in the laboratory, have attempted to differentiate between colisepticaemia and enteric colibacillosis. Even in the relatively recent studies of Dalton (1965),

Radostits (1965) and Watt (1965), where attempts have been made to describe the clinical signs of colibacillosis in calves the distinction has not been made. This is especially surprising when it is realised that these workers were attempting to define colibacillosis prior to the institution of different forms of therapy.

Smith and Little (1922a) first demonstrated that under controlled conditions, the early feeding of colostrum protected calves against colisepticaemia. Other workers have since confirmed that colostrum-fed calves that have absorbed gamma globulins are resistant to septicæmic invasion of E. coli unless this invasion occurs prior to the feeding of colostrum (Gay, 1965). In fact, if it is intended to experimentally induce colisepticaemia in colostrum-fed calves, it is necessary to infect such calves by the intraperitoneal route (Dam, 1967). If, as it appears from the literature, colisepticaemia is preventable merely by the conscientious feeding of colostrum, then the need for the clinical and/or bacteriological differentiation of this syndrome from other forms of colibacillosis by field workers is obvious.

Colostrum-fed calves are still susceptible to enteric colibacillosis (Gay, 1965). This has been noted under experimental conditions by Aschaffenburg, Bartlett, Kon, Roy, Walker, Briggs, Lovell (1949a), Roy, et al. (1955) and Smith (1962). However, there was a suggestion that less diarrhoea occurred in those calves which obtained most colostrum in the experiments of Aschaffenburg, et al. (1949a).

Since the work of Smith and Little (1922a) much interest has been paid in the protective factor or factors in colostrum. Most of this work is reviewed later, and all that need be said at this stage is that it is now generally accepted that the protective property of colostrum lies in its immune lactoglobulin content (Gay, 1965).

Fey and Margadant (1961), Smith (1962) and Gay, et al. (1965a) have all recorded that some calves apparently fed colostrum early in life have subsequently been found to have absorbed little or no immune lactoglobulin. However, in those calves studied by Fey and Margadant (1961) and Smith (1962) the feeding of colostrum was presumed and not definitely known to have taken place. Gay, et al. (1965a) claimed that in an unspecified number of calves, the ability to absorb immune lactoglobulin from colostrum was absent at four to six hour post partum. Unfortunately, no other information regarding these calves was presented.

Smith (1962) and Gay, et al. (1965a) also noted that a wide variation existed in the serum concentrations of colostrum-derived immune globulins in young calves. The latter workers (Gay, et al., 1965a) also showed a definite relationship between these levels and the subsequent fate of these 178 calves. When calves with very low levels of serum immune globulin died, they usually did so from the effects of colisepticaemia. However, calves with slightly higher levels appeared to be protected against septicæmia invasion of E. coli but nevertheless frequently died from the effects of severe diarrhoea. It is especially interesting to note that although those calves with very high levels of immune globulin become diarrhoeic, they did not die.

These studies were later extended by Fisher, Selman, McEwan and de la Fuente (1968) to include a total of 415 Ayrshire bull calves bought from markets in the West of Scotland, and these earlier results were confirmed.

Gay, Fisher and McEwan (1965b) later found that a definite seasonal variation existed in the mean serum concentrations of immune globulin of Ayrshire bull calves aged less than one week. It is of especial interest to note that the lowest mean levels occurred during the first quarter of the year (the period of highest calf mortality).

These observations prompted a series of investigations into factors affecting the absorption of globulins from colostrum by the very young calf and the results of these studies are presented in this thesis. It was felt that if calves with universally high immune globulin levels could be produced, then a significant reduction would result in the mortality due to neonatal calf disease.

PART I

PART I

STUDIES ON NATURAL SUCKLING IN CATTLE DURING THE FIRST EIGHT HOURS POST PARTUM

Introduction and Review of the Literature

The fact that colostrum protects calves against death from colisepticaemia was conclusively demonstrated by Smith and Little (1922a). Following work by Stewart and McCallum (1938a, 1938b), the protective factor in colostrum was initially thought to be vitamin A. However, Blakemore, Davies, Eysenberg, Moore, Sellers and Worden (1948) and Aschaffenburg, *et al.* (1949a) finally showed that the protective factor was in the aqueous and not the fatty fraction of colostrum. Subsequent work confirmed that protection is afforded by the immune lactoglobulin fraction of colostrum (Pierce, 1962; Gay, 1965). The actual nature of this protection is still in doubt, and the theory of strain specific *E. coli* antibodies is by no means proven (Gay, 1965). A full discussion of the protective role of colostrum appears in Part 4 of this thesis.

Recently, Gay, *et al.* (1965a) demonstrated a definite relationship between the total serum immune globulin concentration as measured by the Zinc Sulphate Turbidity Test of four to seven day old Scottish market calves and the subsequent fate of these calves. When calves with little or no serum immune globulins died, they usually did so from colisepticaemia. Above a certain concentration, colisepticaemic deaths did not occur, but deaths from the results of severe diarrhoea were still experienced. Calves with very high levels

of serum immune globulin under similar conditions developed diarrhoea, but did not die. Gay, *et al.* (1965b) later showed that a marked seasonal variation existed in the mean serum immune globulin concentrations of such calves. High mean serum concentrations were encountered in the summer and very low mean serum concentrations occurred in the winter months when calf mortality is usually at its highest (Wither, 1952-1953). Smith, O'Neill and Simmonds (1967) claimed that a seasonal variation of this type did not occur in the 230 newborn calves they examined in England. However, close examination of their data reveals that very high levels of serum immune globulin occurred only during the summer months. Smith, *et al.* (1967) suggested that different forms of newborn calf management might account for the more marked seasonal variation seen in Scottish calves. In view of this last observation, it is interesting to note that Lovell and Hill (1940) stated that the seasonal variation in calf mortality rates was much more marked in Scotland than in England.

In the west of Scotland calves born in the winter are managed very differently from those born in the summer (Selman, de la Fuente and Fisher, 1968). In this area most cows are housed continuously throughout the winter in byres and calves are usually born in the byre where the dams are tied by the neck. Under these conditions, nursing cannot take place, and a calf born at night may spend several hours lying and struggling to rise in the dung channel behind its dam. If such a calf does eventually stand, the chances that it will either suckle its own mother or be able to suckle another cow are remote.

Soon after calves are born, or when night-born calves are found in the morning, they are removed to separate accommodation and later fed colostrum from a bucket. This first feeding is often delayed for up to 15 hours. During the summer calves are usually born out of doors and are removed from their dams when it is convenient to do so. Often summer calves remain with their dams for 24-48 hours. It might well be significant that the appearance of high serum immune globulin concentrations as demonstrated by Gay, *et al.* (1965b) coincides with the time (April) that cows start to be turned out in the west of Scotland, and that mean low levels commence in November when cows are housed for the winter.

Wither (1952-1953) carried out a survey of calf mortality in England, Wales and some parts of Scotland over a three-year period, and a total of 3744 dairy helper calves were studied. When colostrum was obtained by suckling, the mortality rate was 3.9%, and when it was fed by bucket, the rate was 9.1%. Seventeen out of 27 herds in England and Wales allowed calves to suckle colostrum, but only 2 of the 8 Scottish herds studied did so. In the same survey, Withers (1952-1953) also cites one instance where a change from bucket feeding of colostrum to suckling for the first four days of life was associated with a drop in mortality rate (all causes) from 30% (1945) to 3.6% (1946), nil (1947), and 1.6% (1948). In another herd, a change from suckling to bucket feeding of colostrum resulted in an increase in deaths (all causes) from nil (1946) to 5.9% (1947) and 18.75% (1948). These findings confirmed those of Lovell and Hill (1940). Recently, Leech, *et al.* (1968) in a study on calf mortality carried out during 1962-1963

Involving 32,373 home-bred calves, found that calves born in fields generally stood a higher chance of survival than calves born indoors at the same time of the year. It was again noted that calves fed colostrum by suckling were more likely to survive than calves fed colostrum by bucket.

Flinscher (1963) not only stated that calves should be left with their dams for an unspecified length of time, but suggested moreover that the calves should be fed colostrum within two hours of birth. Gay, et al. (1965a) suggested that premature cessation of immune lactoglobulin absorption occurred in some of their calves, rendering them incapable of absorption by four to six hours post partum. In view of this, Walker-Love (1966) stressed that calves should be left with their dams for a period (again unspecified) after birth. The assumption here, presumably, was that suckling naturally occurs before the calf is four hours old. Inglis (1960) questioned whether the claims that feeding colostrum by suckling reduced calf mortality. He stated that many farms he had investigated with diarrhoea problems fed colostrum in such a way. However, he presented no data to support this opinion.

The method of feeding colostrum has, in fact, been shown to affect the subsequent level of serum immune globulin (Smith, et al., 1967). It was shown in a survey of 190 home-bred calves that calves receiving colostrum by suckling or by suckling and bucket feeding together, in general, absorbed larger quantities of immune lactoglobulin than calves fed colostrum by bucket alone.

Even where colostrum is fed by suckling, the resulting serum immune globulin concentrations are very variable and from 10%-36% of these calves may be agamma- or hypogammaglobulinaemic (Fey and Margadant, 1961; Smith, 1962 and 1965; and McEwan, 1966). However, neither Fey and Margadant (1961), Smith (1962, 1965), nor McEwan (1966) knew for sure when, or whether, their agammaglobulinaemic calves had suckled. Smith (1962), in fact, concluded "that in practise it is obviously necessary to ensure that calves obtain colostrum in very early life and not to assume that they will do so if left with their mothers." Fey and Margadant (1961) assumed that their calves had obtained colostrum and felt that in the cases where agammaglobulinaemia existed, premature cessation of immune lactoglobulin absorption had occurred. Smith, *et al.* (1967) recorded very low levels of serum immune globulin in eight of eighty calves "seen to have suckled within a few hours of birth." These observations were, however, made by farmers and no detailed information regarding birth and suckling times were presented so that no great importance can really be placed on these statements.

Very little has been recorded specifically regarding the behaviour of cattle in the early post-parturient phase. Actual observations on newly calved cows with their calves are scarce, and only two publications, those of Walker (1950) and Hafez (1962), appear to be available. The latter account gives no details as to the number of animals observed and contains no references, although it basically agrees with the earlier work by Walker (1950). This latter work is on observations carried out on two dairy shorthorn cows and one heifer and

their calves only. Due to this lack of detailed information regarding cattle, reference will be made to work in other species whenever it is felt to be relevant.

The fact that a newborn ungulate, on rising, moves directly towards its dam has been noted by Fraser (1962) and Smith (1966). The gait of the newborn calf while making its initial teat-seeking advances has been described by Fraser (1962), who pointed out that the head is out-stretched and the muzzle elevated to make contact with the dam's ventral abdomen or legs. This posture is very similar to the teat-seeking postures of other herbivores such as lambs (Collias, 1956, and Smith, 1966), kids (Collias, 1956) and foals (Hafez, 1962). Adler, Linn and Moore (1958) and Fraser (1962) considered that the teat-seeking drive in newborn ungulates was an inherent instinctive pattern. The releaser for teat-seeking activities was considered by Fraser (1962) to be the ventral underline of the dam and by Kingscote (1962) to be the shade below the dam. Rylands (1967) claimed that orphaned lambs would frequently carry out teat-seeking advances towards a source of heat (e.g. a kitchen fire).

Adler, et al. (1958) noted from motion pictures of newborn kids taken by Blauvelt, (1954) and of lambs taken by Moore (1958) that the initial teat-seeking advances were occasionally directed towards objects other than the dam. Collias (1956), Adler, et al. (1958), and Smith (1966) all record instances where kids and lambs followed humans soon after rising and also made advances to any object of a convenient height (e.g. a fence rail or a coat hanging over a trellis). Rylands (1967) claimed that the suckling drive could be strengthened in weak lambs by covering their heads with a warm cloth.

The teat-seeking activity of newborn lambs was investigated by Alexander and Williams (1966). They found that depression of this activity occurred with fasting or continual lack of reward (i.e. when the ewe's udder was covered with a cloth). The same workers also noted that when these lambs were finally allowed access to the ewe's udder (i.e. when the cover was removed), fewer suckled successfully than control lambs which had been maintained under conditions designed to minimise teat-seeking activity (i.e. a darkened box with out-sloping walls).

The "pushing syndrome" (head pressing) of herbivores suffering from certain forms of cerebral disease has been likened to the "pushing" or teat-seeking activity of the newborn (Adler, *et al.*, 1958). A parallel, these workers claimed, exists in humans suffering from some types of brain disease, wherein suckling motions of the mouth and hand grasping, which normally disappear at weaning, reappear in deep coma. Fraser (1962) convincingly argued against this contention on the grounds of "direction" (i.e. random in cerebral disease, but releaser oriented in teat seeking); "motivating force" (i.e. depressed cerebral function in disease states as opposed to feeding drive); and finally, "posture" (i.e. head lowered in disease states, head extended during teat seeking). According to many workers, the motivating force for teat-seeking advances is not solely a feeding drive, but this point will be discussed later.

The later, stronger and more individual-specific bond between dam and offspring was claimed by Smith, Van-Toller and Boyes (1966) to resemble "imprinting" as seen in some birds (Lorenz, 1935). Altmann

(1963), however, contended that this mother-offspring bond as seen in ungulates does not completely resemble classical imprinting in ducks and geese. She pointed out that in true imprinting the event is irreversible and takes place within narrow time limits, whereas in ungulates the bond is sometimes reversible, is more flexible and is less limited in respect of time.

Maternal orientation, aiding teat-seeking advances and suckling, has been recorded in cattle (Hafez, 1962), in sheep (Collins, 1956). This aspect of maternal or nursing behaviour would generally seem to consist of the dam standing still to allow suckling to take place, sometimes with one or both hindlegs abducted to trap the muzzle of her offspring. Occasionally, ewes have been observed to push lambs posteriorly when teat-seeking advances were being made around the dam's forelegs (Smith, 1966). Altmann (1963) observed the nursing and suckling behaviour of wild moose and elk. She found that in many cases the new-born moose-calf was too small to reach its dam's udder and in these cases, the dam frequently squatted and sometimes even lay down to allow suckling to take place. The general consensus of opinion would seem to be that although moving towards the dam and teat seeking are inherent, instinctive movements in newborn ungulates, the actual localisation of teat-seeking activity to the udder area is due to the dam's orientation. However, Alexander and Williams (1964) in a series of experiments designed to investigate the importance of maternal orientation by ewes, found that this was not important in helping a robust lamb to find a teat. It was pointed out that these

experiments were carried out in warm weather in small pens, so that under less ideal conditions, maternal orientation might be of more importance.

The only worker claiming that a "homing device" exists for teat detection was McBride (1962) working with newborn pigs. These animals were consistently seen to press their noses while teat seeking firmly onto the recumbent sow's belly and then follow the direction of the bristles until an unoccupied teat was located. When the bristle pattern of a sow's underbelly was plotted, it was found that all bristles led to the midline of the udder and thence outwards past the udder and back to the hindlegs and vagina. Adler, *et al.* (1958), Hafoz (1962), and Smith (1966) have all suggested that in newborn ungulates the actual location of teats is by trial and error.

Maternal grooming (licking) has been studied in sheep by Collias (1956) and Smith (1966) and in goats by Collias (1956). Barron (1954) suggested that licking may have a survival value in raising the general neural excitability and hence presumably increasing the chances of the newborn finding teats and suckling. If this theory is true, then with the complicating problem of colostrum shutdown to contend with and hence the importance of early suckling, licking may be a very important factor in lamb and calf survival. Blauvelt (1954) claimed that lambs were licked stood sooner than lambs left unlicked by their dams, and Herscher, Richmond and Moore (1963) cited instances where newborn lambs and kids which were not licked shortly after birth failed to stand and occasionally died. However, neither of the

latter two claims were substantiated by timed observations. Alexander and Williams (1964) investigated the importance of grooming by ewes and found that robust lambs left unlicked found teats and suckled as quickly as licked controls. However, when the effects of licking and maternal orientation together were studied, slightly earlier suckling appeared to take place. Once again, this work was carried out on lambs and ewes confined in small pens during good weather. Under more adverse conditions, licking might well help a weak or chilled lamb to rise and suckle.

Maternal recognition and rejection of offsprings have been studied in sheep and goats by Collias (1956) and in sheep by Smith, et al. (1966). Both studies agree that the critical time for the establishment of the attachment between dam and offspring is short. In sheep, it varied between 20 and 45 minutes, and in goats it was about 15 minutes. Special stress was placed by both workers on the importance of a ewe licking amniotic fluid off its lamb. Yet another factor in fixing the dam-offspring bond would appear to be the licking by the dam of the newborn animal's perineum during the initial teat-seeking advances (Walker, 1950, and Collias, 1956). Provided a ewe was kept in complete isolation from other lambs following parturition, the period of licking for 20-30 minutes followed by attachment could occur within a period of several hours following parturition (Smith, et al., 1966). Licking was more likely to be readily carried out if the lamb or lambs were still wet (Smith, et al., 1966). The recognition of an offspring by its dam is not, however, solely an olfactory or gustatory function since newborn lambs and kids cleaned

and washed with a strong detergent after the initial grooming phase was over were nevertheless recognised by their dams (Collins, 1956). Evidence is also presented from fostering experiments by Smith, et al. (1966) that an ewe will normally accept the first lamb or lambs presented to her within the first few hours of life. The dam-offspring bond may be "blurred" if shortly after birth a female goat fails to repel other inquisitive goats. It is claimed, again without convincing proof, that this may have lasting effects on the subsequent social and physical development of that offspring (Blauvelt, 1955).

An interesting situation occurs in the wild Elk (Altmann, 1963). In these animals, the female usually drives off her yearling calf a few weeks before the next calf is to be born. If, however, the new calf is stillborn or dies soon after birth, the dam once again allows the yearling calf to approach and suckle. In periods of severe weather, for example, when neonatal losses were high, this would appear to be a very useful mechanism allowing for the survival of at least the yearlings in the herd.

Hafex (1964) underlined the differences seen in the nursing behaviour of primiparous and multiparous dams and stated that nursing behaviour in older animals is facilitated by the reflexes conditioned during previous lactations, and in primiparous animals is inhibited by the pain and shock of parturition. Wallace (1949) noted that in sheep, poor mothers were commonly those that had experienced difficult or prolonged parturition.

It has already been mentioned that teat-seeking postures of newborn herbivores are very similar. Yet another interesting point

is that the actual positions taken up while suckling are also very similar in the different species, the offspring usually standing parallel to its dam, with the head and neck bent to allow easy suckling to take place (Hafez, 1962). Schloeth (1958) claimed that 5-10% of sucklings in the half wild Camargue cattle took place with the offspring standing behind its dam. However, the calves suckling in this manner were not newborn.

Walker (1950) observed that newborn dairy calves suckled from one or both sides of the udder and occasionally from behind the dam. The number of teats that a calf suckled from one side of the dam depended on the size of the dam's udder (Walker, 1950, and Hafez, 1962). Attempts by newborn dairy calves to suckle from recumbent dams were normally unsuccessful (Walker, 1950). Head butting and tail wagging while suckling or attempting to suckle has been noted in lambs (Smith, 1966) and in calves (Hafez, 1962). The latter author suggested that the butting movements were stimulated by a decrease in milk flow to the calf.

Considerable teat mouthing and nibbling has been observed before actual suckling takes place in lambs (Hafez, 1962, and Smith, 1966), lambs and kids (Collins, 1956) and calves (Walker, 1950). These workers all claimed that this early experience was very necessary and that subsequent sucklings were carried out much more skillfully. This increase in suckling efficiency is possibly due to a reward mechanism (Alexander and Williams, 1966).

Collins (1956) found that two lambs and one kid first suckled at 68 minutes, 108 minutes and 56 minutes post partum respectively. In observations on 248 newborn twin and single lambs, Wallace (1949) found that first suckling occurred at the following times:

TABLE 1
(from Wallace, 1949)

The Times to First Suckling of 248 Newborn Lambs

<u>Suckling</u> (hours post partum)	<u>0 - $\frac{1}{2}$</u>	<u>$\frac{1}{2}$ - 1</u>	<u>1 - 2</u>	<u>≥ 2</u>
% singles (165)	24	39	27	10
% twins (83)	18	40	26	16

From these figures it can be seen that although most lambs (i.e. 90% singles and 84% twins) had suckled by two hours post partum, many had not. Unfortunately, the possible reasons for delayed suckling were not individually recorded, although mention was made of the difficulties some lambs experienced in suckling from abnormal udders. Walker (1950) noted that although the three calves in his series of observations attempted to suckle within three hours of birth, the first real feed was not before five or six hours post partum. Hafez (1962) stated that calves suckled from two to five hours post partum without producing any evidence to support this claim.

Newborn pigs show a marked preference for the anterior teats of the sow (Wyeth and McBride, 1964, and Hafez, 1962). McBride (1963) also established that the birthweight of a pig and the size of its chosen teat were closely correlated. Once established, the teat order

In a litter of pigs appears to be irreversible. When pigs were held onto strange teats, or mixed together before even the last pig was born, the original teat order was quickly established. Young pigs can be induced to accept teats in the same position on another sow (Hafez, 1962).

The belief that twin lambs develop a definite teat preference was investigated by Ewbank (1964), who found that by ten weeks post partum, 67% of twin lambs had such a preference. No observations appear to have been made on teat preference in very young lambs. However, Walker (1950) observed that for the first two days of life, calves preferred to suckle one teat.

Ross, Fisher and King (1957) stated that the neurological basis of the suckling act had not been established at that time. It was pointed out that, in the human infant suckling activity consisted of a series of simple mouth responses. Orientation of head, grasping movements of the hands, mouth movements and swallowing appeared to be chain reflexes. Moreover, respirations were coordinated with suckling. Hooker (1944) found that suckling movements could be elicited in human foetuses by the 29th week of gestation. Kessen (1967) has claimed that apparently normal suckling behaviour occurred in a human hydran-encephalic infant and postulated that the suckling centre was situated in the brain stem.

In general, milk or colostrum that is ingested by a newborn lamb or calf passes straight to the abomasum via the oesophageal groove, irrespective of the position of the animal's head (Hafez, 1962). Occasionally, however, this does not always happen (Benzie and Phillipson, 1957, and Watson, 1944). Shalk and Amadon (1928) claimed

that when large amounts of milk were ingested in large swallows, some was forced between the lips of the oesophageal groove into the rumen. Watson (1944) found that whether or not liquids passed into the abomasum depended not on the temperature or consistency of the fluid, the posture of the lamb or even the act of suckling, but rather by the activation of the behaviour patterns surrounding the act of suckling.

Ploog (1967) fed two groups of babies by bottle, the only difference between the treatment of the two groups being the diameter of the hole in the teat. Those children fed with the teats with large holes finished their meals quickly, then proved restless and needed a further ten minutes suckling on the empty bottle before settling down. The other group took longer to finish their meals and settled down better afterwards. Frequently, these latter children were satisfied with less milk than those of the first group. Alexander and Williams (1966) did not inhibit the teat-seeking activity (sucking drive) of lambs fed by stomach tube although some degree of depression occurred. James (1955), working with stomach-tubed pups, completely failed to depress teat-seeking activity in these animals. Ross, *et al.* (1957) after reviewing the literature on suckling behaviour concluded that there was a need for sucking quite apart from the need to alleviate hunger.

Suckling deprivation in early childhood has been claimed to give rise to non-nutritive (overflow) sucking of the thumbs in young children, and in later life obesity, such oral traits as social drinking, and smoking, and even to characteristic personalities (Ross, *et al.*, 1957). The work supporting these last claims has mostly been non-experimental and is purely opinion unsupported by data. However, experimental work

carried out in human infants (Levy, 1928), newborn puppies (Levy, 1934), and newborn kittens (Kovach and Kling, 1967) has shown conclusively that suckling deprivation may give rise to non-nutritive sucking.

Non-nutritive sucking has been recorded in young goats (Collias, 1956), young lambs (Collias, 1956, and Smith, 1966), and in young calves left with their dams (Walker, 1950, and Hafez, 1962). In these cases it has always been a prelude to first suckling.

Non-nutritive sucking did not occur once in 250 babies fed on a self-demand schedule (Davis, 1940), and rarely occurs in suckled calves (Finscher, 1963). It is, however, a problem commonly met when groups of dairy calves are housed together (Finscher, 1963, and the British Veterinary Association Handbook, Section VI, "The Husbandry and Diseases of Calves") and may give rise to such conditions as haematomas and septic infections of ears and navels and possibly chronic tympany and "blind" tests (Finscher, 1963, and the British Veterinary Association Handbook, Section VI, "The Husbandry and Diseases of Calves"). It will occasionally persist into adult life, and a case has been recorded where the habit was so widespread in a herd of dairy cows that the owner was forced to dispose of it (Wood, Smith and Lisle, 1967). Unfortunately, the method of calf husbandry in this herd was not discussed by the authors. Thorpe (1961) stated that the young dairy calf was frequently a prime example of an animal suffering from suckling deprivation, and he believed this to be the cause of "overflow suckling." Generally where a problem of inter-sucking exists between bucket-fed calves, the advice that is offered

is either to pen separately or else to tether each calf for one hour following each meal (Finscher, 1963, and Wood, et al., 1967).

The fact that frightened kids and lambs will rush to their dams and immediately attempt to suckle has been recorded by Fulton (1949) and Brownlee (1950, 1954). Adler (1958) noted a similar phenomenon in African children frightened by the approach of a white man. From the observations it would seem likely that suckling has an anxiety-alleviating effect on frightened young animals.

To sum up, evidence has been presented from the literature indicating the mortality rates are considerably lower in those herds where calves are allowed to suckle colostrum naturally, compared with herds where colostrum is bucket fed. This may be due to the fact that natural suckling tends to produce higher serum immune globulin concentrations than does bucket feeding. A wide variation exists in the serum immune globulin concentrations of calves left with their dams, but there is no real evidence that those calves with little or no serum immune globulin had actually suckled their dams. The nursing and suckling behaviour of cattle in the period immediately following parturition has not been extensively studied and nothing is known regarding factors affecting the time to first suckling. Delayed suckling in lambs has been linked with udder abnormalities in the ewes.

From the few experiments so far carried out in cattle, sheep and goats, it would seem that teat-seeking, maternal grooming, and finally, suckling, combine to produce the dam-offspring bond which is very necessary for the immediate survival of all species, save perhaps

the dairy calf. The fact, however, that most calves survive separation from their dams and artificial feeding does not necessarily mean that this is an ideal method of management. "Overflow sucking" in bucket-fed calves is probably a manifestation of sucking deprivation.

In view of the dearth of information regarding the nursing and suckling behaviour of cattle in the early neonatal period, it was decided that a study of this over the first eight hours post partum would be of value. It was felt that both the dam's and the neonate's behaviour patterns should be recorded because these patterns are probably so interrelated that the consideration of, for example, the times to first suckling would not reveal all that was needed to be known. Nevertheless, the wide variations seen in the serum immune globulin concentrations of calves left to suckle their dams might purely be a reflection of variations in these times, with the occasional agammaglobulinaemic calf found following such management, simply a result of suckling delayed beyond the period of globulin absorption. A study of the factors responsible for delayed suckling, if such occurs, was therefore also indicated.

Finally, it was felt that the observation that suckling of colostrum generally produced higher serum immune globulin concentrations than did bucket feeding, and that a seasonal variation in such levels has been shown to occur with low mean levels in winter when suckling cannot usually occur, should be tested. Consequently, it was decided to carry out at least half of the suckling experiments during the winter months in an effort to rule out this seasonal variation by a purely managerial approach.

The results of these observations are recorded in Sections I, II and III of this part.

MATERIALS AND METHODS

1. Experimental Animals

(a) Cows and heifers

All of the dairy cows and heifers were obtained from one cattle dealer and the beef cows from two Stirlingshire farmers. All of the dairy pregnant cows were of the Ayrshire breed and all were admitted to the Veterinary Hospital within the last few days of gestation. The beef cows were of varying breeds and, due to the difficulties of knowing exactly when this type of cow is due to calve, they were all admitted at an earlier stage of gestation than the dairy animals. This had the advantage of allowing these cows, some of which had not been housed during the preceding four years, to become used to housing.

None of the Ayrshire helpers had more than two pairs of permanent incisor teeth and hence all were judged to be between two and two-and-a-half years of age. Exact aging of the beef and dairy cows was not possible, but all had four pairs of permanent incisor teeth, and from a careful examination of udder and teats, all appeared to have had at least one preceding lactation.

None of the cows or helpers used in this experiment showed any sign of ill health during its stay in the Veterinary Hospital, nor any clinically detectable abnormalities of udder, teats or milk. None of the dams in this series required assistance at parturition although in some cases, labour was prolonged.

(b) Calves

All of the calves born to the dairy cows and heifers were dairy calves. Most of these were pure Ayrshire, but some were Ayrshire cross Friesian calves. The beef calves were of varying breeds.

Every calf in the series was judged to be healthy at birth although there was a great variation in the demeanour and vigour of calves, and all were judged healthy at 48 hours post partum. Almost every calf in the series was found to be profusely diarrhoeic at about 20-24 hours post partum. This diarrhoea followed soon after the passage of meconium with the diarrhoeic period lasting about four hours. It then resolved spontaneously. Some calves were seen to be dull during this period and a few were found to be passing blood clots in the very fluid faeces. By 30-36 hours post partum, the faeces were light yellow in colour and formed a definite stool on being passed.

2. Care of Experimental Animals

(a) Accommodation

When an animal was found to be in the first stage of labour or, on vaginal examination was found to be approaching this stage, it was transferred from a holding box or byre into one of the observation boxes. The two observation boxes used measured 13 ft. x 11 ft., and both had high ceilings and good ventilation. Prior to admitting an animal to one of these boxes, the floor was deeply covered with wheat straw and approximately 20 lbs. of hay and approximately 8 lbs. of cattle nuts were placed in the manger. After the animal had been placed in the box, both doors of the box were closed, and these doors

were left closed until ten hours post partum at the earliest. Both cow and calf were left in the observation box for at least 24 hours, and they were then moved to another similar box only if the observation boxes were needed for another parturient animal. Dams and calves were left together for a minimum of 48 hours.

(b) Feeding

As already stated, sufficient food was placed in the observation boxes prior to parturition to last the cows for at least ten hours. After this, they were fed twice daily at 7.30 a.m. and 5.30 p.m.

All of the cows and heifers were offered about 15-20 lbs. of hay daily, but many of the beef cows took clean wheat straw from their bedding in preference to the hay.

The dairy cows and heifers were fed twice daily approximately 8 lbs. "summer nuts," (16% protein) as supplied by Shearer and Company, Primrose Hills, Glasgow. The beef cows were fed about 4 lbs. twice daily of a special low protein, high magnesium cake made by the above company to their owner's specifications.

The beef cows' calves obtained their sole subsistence from their dams' milk. Usually, the dairy calves were also left with their dams until returned to their owner and they were therefore not fed by bucket. Occasionally, dairy calves were removed from their dams after 48 hours of age and these were then fed fresh whole milk obtained from a bulk tank at approximately the rate of 5% of their bodyweight, twice daily.

(c) Milking

The beef cows were not milked, and the dairy cows and helpers were never milked before 24 hours post partum. When milking was carried out between 24 and 48 hours post partum, only small amounts of colostrum were removed in an attempt to reduce the tension in the udder.

3. Experimental Procedures

(a) Prediction of parturition

It was very soon realised that the traditional methods of predicting parturition (i.e. relaxation of pelvic ligaments, tension of colostrum in udder and teats and behavioural changes) were not sufficiently precise to predict accurately the time of parturition. As a result, in the early stages of the following experiments, many observations on calves were not made due to unsuspected calvings. Moreover, much time was wasted sitting up with cows that were erroneously thought to be about to calve.

It was finally decided to carry out vaginal examinations of all cows and helpers at 9.00 a.m., 5.00 p.m. and 11 p.m., or whenever it was felt necessary. The features noted during vaginal examination were as follows:

(1) The ease of entry of a soaped hand into the vagina.

Entry became easier as parturition approached, due first to the increase in size of the vagina and second to the increased amounts of mucus present. Some helpers had such small vulvas that it was impossible to introduce a hand into the vagina until shortly before parturition.

(2) The size and consistency of the cervix.

A wide variation was found on first examination between the sizes of cervixes. The heifers all had small, hard, conical cervixes, approximately one inch in diameter at the base and one inch high. The cervixes of the cows varied a great deal. A few were slightly larger but similar in shape to the heifers, but most were approximately two inches in diameter at the base and more oval than cone shaped. Many older cows had a posterior os to the cervix which was extremely hard, nobby and as large as a clenched fist. Frequently old cervical tears were palpable.

(3) The rate of softening and/or dilatation of the cervix.

As parturition became imminent the cervix enlarged and softened until it was frequently very difficult to find. At this stage, progressive dilatation of the posterior os of the cervix started. When the posterior cervical os became four-fingers dilated, it was usually possible to introduce one finger through the anterior os and palpate the calf's feet. Later still, only a rim of cervix was palpable, and this gradually and completely disappeared. Even at this late stage, cows frequently appeared comfortable and at ease.

As a result of these observations, it became possible to predict the approach of parturition on the time scale in Table 2.

TABLE 2

The Prediction of Parturition In the Bovine Animal on the
Grounds of Consistency and Degree of Dilatation of the Cervix

	<u>Consistency of Cervix</u>	<u>Diameter of Posterior Os of Cervix</u>	<u>Predicted Time of Parturition</u>
1.	Hard	3 fingers	> 15 hours
2.	Soft	3 fingers	8 - 15 hours
3.	Soft	4 fingers (anterior os, 1 finger)	4 - 10 hours
4.	Soft	1 in. rim remaining	2 - 6 hours
5.	Complete dilatation		$\frac{1}{2}$ - 5 hours

Some cows appeared to progress faster than others and generally, any cow with a soft, difficult to locate, cervix was viewed with suspicion irrespective of the degree of dilatation of its posterior os.

(b) Observation Techniques

Observations were carried out through an observation hatch, the door of which was kept slightly ajar. At all times, the boxes were kept well lit and the room where the observer sat was either in almost complete darkness or, at most, very gloomy.

As stated above, cows or helpers were introduced to one of the observation boxes during first or early second stage labour. During the first few minutes following introduction to the observation box, most animals moved around and appeared to be exploring the new accommodation. At this time, most of them found the observation hatch and appeared to register the fact that the author was present. No attempt was made by the author to conceal himself, but every attempt

was made to minimise noise and movements. After first noting the presence of the author during the phase of exploration, no further interest was shown in the observation hatch by any dam. As parturition proceeded, all of the cows and heifers became increasingly less interested in their surroundings and following parturition, all save the dams with poor maternal ability focused their attention almost entirely on their calves.

Observation was carried out continuously for eight hours following parturition by the author himself. As much data as possible was collected regarding the behaviour of both dam and offspring, and all of the individual cow and calf observations are presented in Appendix I. The data presented in Sections I and II of the following part of this thesis represent the overall picture of what was observed, although reference is made whenever necessary to individual dams or calves of interest.

(c) Definitions

(1) Throughout this work, the terminology suggested by Cowie, Folley, Cross, Harris, Jacobsohn, Richardson (1951) for use in lactational physiology has been adhered to. These workers suggested the following definitions:

"Nursing"--the behaviour of the lactating mammal in promoting access of the young to the nipples and teats.

"Suckling"--the activity of the young (or operation of the milking device) with the aim of obtaining milk from the mammary glands.

(2) Due to the incoordinated and frequently unsuccessful attempts at standing made by most newborn calves, some difficulty was experienced in deciding when a calf was actually standing properly for the first time. Consequently, it was decided that calves could only be considered standing if they did so for one continuous minute with the soles of their feet in contact with the ground.

(3) Some difficulty was experienced in deciding what constituted a "suckling spell" or period. It was therefore decided at the outset that any number of suckling spells separated by less than five minutes should be regarded as one suckling spell. However, the "total suckling time" in any suckling spell was the total time spent actually suckling and apparently obtaining milk (i.e. intermittent spells of teat seeking were not included in total suckling time).

(4) Timing of observations was started when the hips of a calf passed through its dam's vulva and all time factors were corrected to the nearest minute except in the case of suckling spells and total suckling time. In the latter two instances it was decided to time to the nearest half minute.

(5) Earlier observations had suggested that the shape of a dam's underbelly was a very important factor in deciding how quickly a vigorous calf suckled its dam. Consequently, the terms "good shape" and "poor shape" were applied to dams prior to parturition (see Figures 1 and 2).

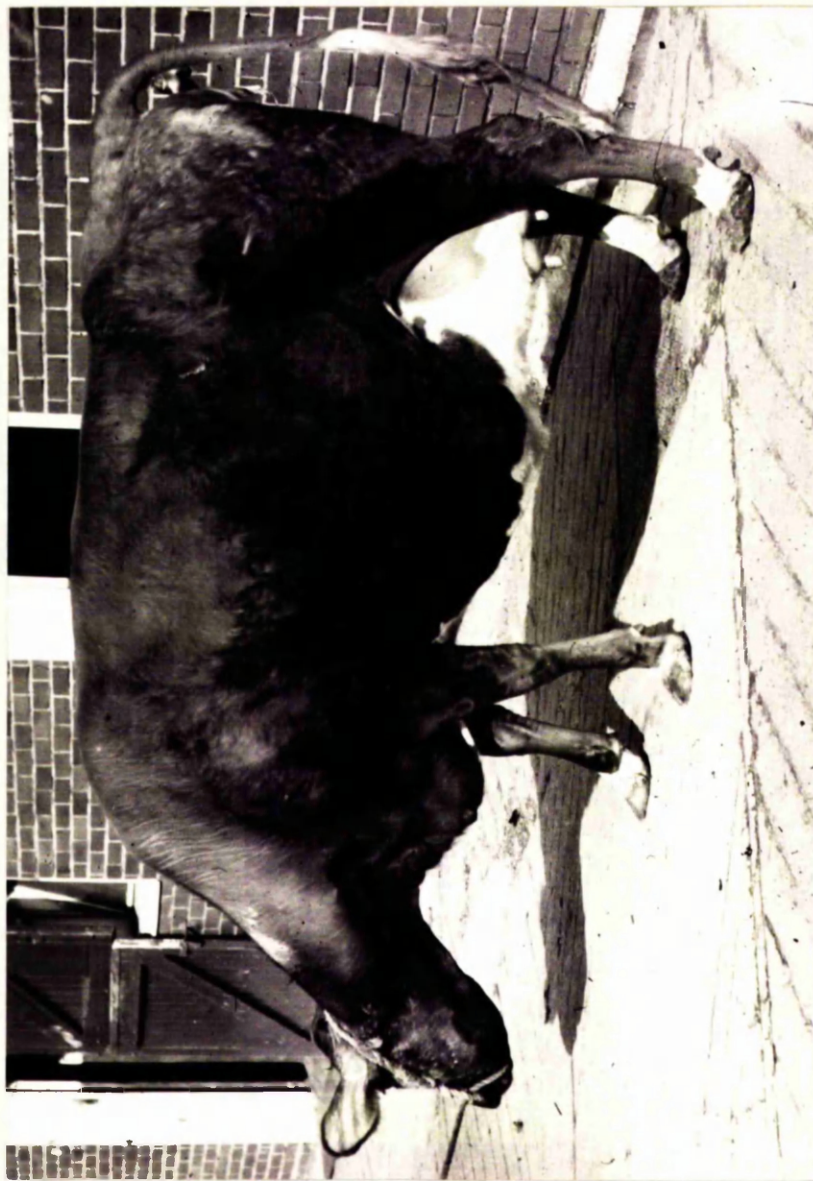


Fig. 1. An example of a dan (DC10) with a poor shape from the viewpoint of suckling. In this case, the calf did not suckle during the eight hour observation period and its 48-hour serum immune globulin concentration was found to be 5 Z.S.T. units. The total amount of time spent teat seeking during the observation period was 107 minutes.

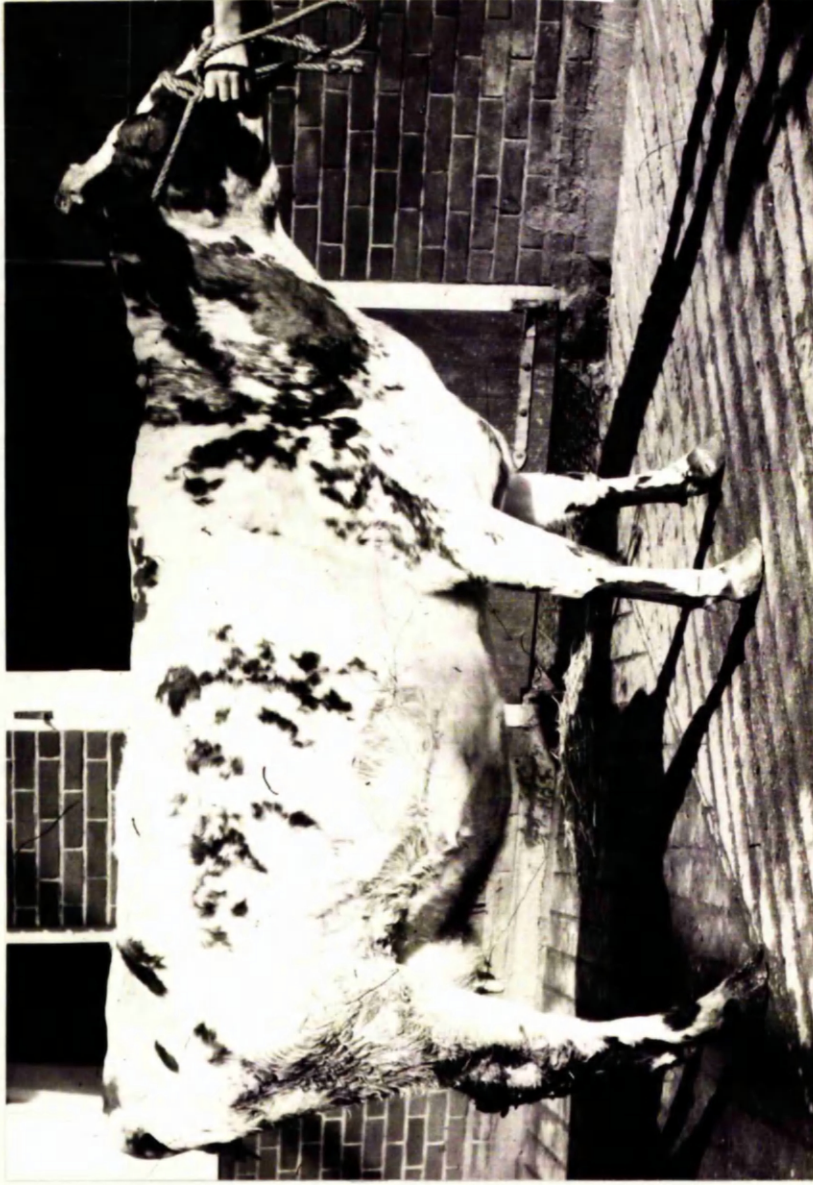


Fig. 2. An example of a dam (DC8) with a good shape from the viewpoint of suckling. In this case, the calf was observed to first suckle at 91 minutes post partum and its 48-hour serum immune globulin concentration was found to be 46 Z.S.T. units. The total amount of time spent teat seeking prior to first suckling was 16 minutes.

"Good shape" meant that the udder and teats were either on a level with or higher than the dam's xiphisternum. "Poor shape" meant that, due to the large size of abdomen and/or udder and teats, the xiphisternum was the highest part of the dam's underbelly. No degrees of "goodness" or "poorness" were allotted to cows and heifers.

(6) The term "cleansing" was reserved for the act of passing the placenta.

(7) Each dam and calf were allotted the same number. In order to make identification easier, the beef cows and calves were given the prefix "BC," the dairy heifers and their calves, "DH," and the dairy cows and their calves, "DC."

(d) Blood sampling and preparation of serum

A blood sample was removed at 48 hours post partum from the jugular vein of each experimental calf. This was allowed to stand and clot at room temperature. Serum was then removed and its immune globulin concentration was either immediately measured or, if this was not convenient, the serum sample was then stored at -4°C until the procedure could be carried out.

(e) Estimation of the serum immune globulin concentration

This was by the zinc sulphate turbidity test as described by McEwan (1968).

A solution of zinc sulphate (208 mgm. $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ /litre) was prepared in a volumetric flask using carbon dioxide-free distilled water. Two matched colorimeter tubes were then taken and 6 ml of distilled water were placed into the first (control) and 6 ml of the

zinc sulphate solution were placed into the second (test). A sample (0.1 ml) of the serum under test was then delivered into each tube. The tubes were then gently shaken and allowed to stand for a timed 30 minutes at room temperature. After this time had elapsed, the tubes were inserted in turn into an E.E.L. colorimeter (Evans Electroselenium, Halstead, England) after the instrument had been set at zero with a "blank" tube containing 10 ml distilled water only. An Ilford blue-green filter (No. 623) was used. The zinc sulphate turbidity value (Z.S.T. units) was then obtained by subtracting the control tube turbidity value from that of the test tube. The value, whenever necessary, was then corrected to the nearest whole number ($0.5 = 1.0$).

It was not possible to obtain a precolostral blood sample in the behavioural experiments and consequently the 48-hour serum immune globulin concentration was assumed to be due purely to absorbed immune lactoglobulin.

(f) Statistical methods

Throughout this work, the statistical methods employed are those suggested by Bishop (1966). Throughout this thesis, all deviations of the mean are expressed as both Standard Deviations (S.D.) and Standard Errors (S.E.) of the means.

Section I

Behaviour of the Dam

(a) Dam's position at calving

In 20 calvings (eight beef cows, seven dairy cows, and five dairy heifers) the calves were born to their hips while the dams were recumbent. At this stage, the dams rose to their feet and dropped their calves, with little further effort, on rising or shortly afterwards. Only one cow (a beef cow) remained standing throughout the act of parturition.

Nine dams (one beef cow, three dairy cows, and five dairy heifers) completed parturition while lying down.

Most dams that completed parturition from the standing position immediately turned and commenced to lick their calves. Two dams (BC7 and DH7) which remained recumbent for the whole of the calving act also immediately rose and started to lick their calves.

All other dams calving from the recumbent position remained so for a variable time following the completion of parturition. Dams DH3, DH4, DH9 and DC1 remained recumbent for eight, three, two and two minutes respectively after calving, but on rising all immediately started to lick their calves. Of these dams, DH3 and DH4 appeared very stunned and shocked as a result of calving and DC1, an old cow, seemed exhausted by the efforts of calving.

Dams DC4 and DC5 remained recumbent for four and seven minutes respectively. On rising, both cows initially ignored their calves and began licking and eating the bedding that was soaked in amniotic fluid. However, their attention was soon focused on the calves as these made initial attempts to rise into sternal recumbency, and the normal grooming processes followed.

The remaining helper, DH5, remained recumbent for 70 minutes following a difficult and protracted calving, completely ignoring her calf. On eventually rising, all teat-seeking advances by the calf were aggressively rejected until licking was initiated 258 minutes post partum.

(b) Licking and grooming behaviour

As already stated, most dams initiated licking of their calves immediately on calving. They whirled around, quickly smelled at their calves in a peculiar jerking position and then began licking. The licking process was almost always accompanied by initially, loud blaring, and later, quieter grunting on the part of the dam.

Every beef cow followed the pattern outlined above, but some other dams (five dairy helpers and five dairy cows) were slow or completely failed to initiate this licking of their offspring.

Delayed licking in four dams (DH3, DH4, DH9 and DC1) resulted from the fact that in each case, calving was completed in the recumbent position and these animals were subsequently slow to rise. On rising, however, licking was initiated immediately. Three dairy cows (DC2, DC4 and DC5) showed great interest in amniotic strands,

fluid and bedding contaminated with these immediately after calving and at first ignored their calves. However, their interest was immediately focused on the calves as these made tentative efforts to rise into sternal recumbency. Although DC4 then spent a considerable time licking her calf, the other two cows (DC2 and DC5) still paid most attention to the contaminated bedding.

Cow DC6 first licked her calf at two minutes post partum and thereafter only licked it occasionally although continual grunting occurred in the normal way. Later, as the calf struggled to rise and then made teat-seeking advances, the cow's licking became only slightly stronger.

Heifer DH5 finally calved in the recumbent position after a long and difficult labour. After this, she remained recumbent for 70 minutes. During this time, and after rising, she persistently ignored her calf and at first appeared very dull although she later brightened and started cuddling. All teat-seeking advances by the calf were aggressively rejected and licking was not initiated until 258 minutes post partum. The calf weakened itself considerably in attempting to suckle and the helper started licking it, not as teat seeking took place, but as it stood shivering and exhausted. The first few licks by this dam were hesitant, but almost immediately the licking became bold and continuous even though the calf was almost recumbent by this time. Once licking was established, teat-seeking advances were accepted in the normal way and suckling occurred after a further 43 minutes. The subsequent dam-offspring bond was strong.

The only dam (DH6) which failed to lick its calf at all during the observation period also aggressively rejected its teat-seeking advances so that suckling did not occur. In this case, the parturition was quick and easy, but nevertheless, the dam subsequently appeared to be extremely dull and completely ignored the calf until teat seeking was commenced. Interest was never shown in amniotic fluid or in bedding contaminated by this.

After spending some time continuously licking their calves, most dams then became interested in, and started eating, amnion, amniotic fluid and contaminated straw. The duration of this initial phase of continuous calf licking was investigated in the 25 dams which behaved in this way. Dams DH5, DH6, DC2, DC5 and DC6 have already been described as being abnormal in their licking behaviour and have been excluded from the following table.

TABLE 3

The Duration of Initial, Continuous Calf Licking

<u>Dams</u>	<u>No.</u>	<u>Mean</u> (mins.)	<u>S.D.</u>	<u>S.E.</u>
Beef cows	10	48.3	±37.1	±11.7
Dairy heifers	8	11.0	± 8.5	± 3.0
Dairy cows	7	32.9	±18.5	± 7.0

From these results it is found that the dairy heifers spent significantly less time licking their calves in the initial phase of grooming than the beef cows or dairy cows ($p = <0.01$). Moreover, heifers DH5 and DH6 were so abnormal in their behaviour (this has already been discussed) that they have been excluded from this table.

Similarly, although no significant difference exists between the initial licking times of the beef and dairy cows, it must be remembered that three dairy cows (DC2, DC5 and DC6) showed very little inclination to lick their calves; although later, as their calves struggled, DC2 and DC5 showed slightly more interest in them.

The areas licked by the dams were very constant in most cases. The early, often frenzied licking, was aimed mainly at the thorax, back and abdomen of the calf. Only four dams (DH2, DH4, DC1 and DC8) showed special interest in the umbilicus. As a calf rose into sternal recumbency, attempted to rise and made initial teat-seeking advances, licking was concentrated on its head and neck. Later still, as the calf pushed at its dam's underbelly or suckled, the dam frequently licked the perineal region. Occasionally while suckling, a calf passed meconium, and when this occurred, it was carefully licked up in every case by the dam.

Although interest in the calf gradually diminished, and dams became keen to lick and eat contaminated straw, this was not an irreversible change. Most dams became extremely excited whenever calves struggled to rise or made teat-seeking advances and calf licking was quickly resumed. Only two dams (DH6 and DC6) showed no interest in eating up amnion and contaminated bedding. Both cows have been discussed previously in relation to calf licking as DC6 only licked her calf on rare occasions, and DH6 never licked her calf one during observation.

All 30 dams in the series had cleansed by ten hours post partum. All dams save DH6, DC1 and DC6 ate their afterbirths. Of these, DH6 and DC6 have already been discussed in relation to calf licking. The process of eating the placenta, although obviously difficult, was carried out usually without pause. All of the dams that ate their afterbirths during the observation period, concluded by eating any straw that had been contaminated by this.

Four dams in the series (BC4, BC9, DC2 and DC5) exhibited "Fleham" during the observation, two while licking calves, one while eating contaminated bedding and one during the act of cleansing. "Fleham" is a movement usually associated with sex play in either the male or the female herbivore and involves an animal elevating its muzzle for several seconds while partially opening the mouth and retracting the upper lip (Hafez, 1962).

Three distinct sounds were usually discernible in the dams in this series. During the initial phase of calf licking, the dams frequently became very excited and loud drawn-out bellows were emitted and delivered with the mouth open and the head extended in the direction of the calf. Later, when the dams had quietened down, a series of soft, pharyngeal grunts would be made with the mouth closed. This series of sounds were frequently repeated during calf licking or while eating contaminated bedding. A similar sound to this nursing sound, but louder and only repeated once or twice, was made when dams were worried. This could be heard, for example, when a calf wandered too far from its recumbent dam.

(c) Maternal orientation

On rising, a calf almost immediately directed its movements towards the dam and in all save two cases (DH5 and DH6), the result was that the dam turned to face the advancing calf and usually resumed licking its head and neck. At this stage many dams backed away from their calves in order to continue this licking. Later, however, the dams ceased backing away and allowed the calves to push at their bodies. This routine usually resulted in the calf starting to teat seek at the dam's shoulder, and it then advanced along her underbelly.

The times between a calf's rising to its feet and its dam actually standing well to teat-seeking advances varied considerably. It, of course, depended to a certain extent on how quickly calves initiated teat-seeking advances, but this was soon after standing. In only three cases (BC7, DH1 and DH8) did dams stand still immediately their calves arose.

TABLE 4The Time Taken by Dams to Stand for Their Calves' Teat-Seeking Advances

<u>Dams</u>	<u>No.</u>	<u>Mean</u> (mins.)	<u>S.D.</u>	<u>S.E.</u>
Beef cows	10	16.5	±12.2	±3.9
Dairy heifer	8	21.3	±25.0	±8.8
Dairy cows	8	23.8	± 9.1	±3.2

No significant difference exists between these groups of dams. However, for these figures to be seen in their true light, it must be remembered that although all beef cows quickly stood to the teat seeking

of their calves, two dairy heifers (DH5 and DH6) did not. For this reason, these two heifers have been excluded from the table. Two dairy cows (DC1 and DC6) have also been excluded because although their calves stood quite easily for the required minute, they subsequently fell and stayed recumbent for 23 and 31 minutes respectively. Their dams would presumably have stood to teat-seeking advances earlier had they been made. In fact, when their calves eventually started teat seeking, these cows immediately stood still to receive their advances.

Actual postural changes on the part of the dams which made teat seeking and/or suckling easier for the calf were seen only on a few occasions. Two dams (BC2 and DH1) presented their udder areas to their teat-seeking calves by circling their hindquarters towards the calves as the latter advanced. Moreover, DH1 was clearly seen to abduct a hindleg with the result that her calf's nose was trapped momentarily in her inguinal region.

Two dairy cows (DC2 and DC3) appeared to make attempts to remove their calves' teat-seeking advances from the axillary area to the udder. Several times, DC2 walked forward a few steps so that the calf, which had been pushing behind her elbow, was forced to make advances in the area of the udder. The calf of DC3 occasionally pushed at the anterior aspect of the elbow and brisket of its dam. Whenever this happened, the dam raised her foreleg with the result that the calf's advances carried it along the dam's underbelly to the udder.

The only other occasion when a dam showed an obvious intention to aid her calf's teat seeking was in the case of BC5. After this calf had apparently emptied both left quarters of the udder but still continually tried to suckle from them, its dam several times presented the right side of her body to the calf.

(d) Maternal rejection

Teat-seeking advances were rejected at some time by 15 of the 30 dams and consisted of simply moving away from or kicking at their calves. In 13 of the 15 dams, however, rejections were only occasionally seen, and usually occurred only when calves pushed extra vigorously at udders, sucked udder or belly skin or nibbled the sides of teats. This mild form of rejection inhibited only the weakest calves (e.g. DH1 and DH10). As soon as suckling commenced, all 13 dams became very quiet.

Heifers DH5 and DH6 rejected their calves' advances for 301 minutes and for the whole of the 480 minute observation respectively. Both heifers kicked very hard but only as their calves made contact with their belly or udder. DH6 was frequently observed to flinch whenever her calf made contact. In the case of the 13 good mothers mentioned above, teat-seeking avoidance consisted purely of side stepping, but DH5 and DH6 often ran away from their calves.

Six dams were seen to feint at their calves with their heads, but in four cases (BC3, DH3, DC2 and DC4) it seemed that no harm was intended and all four were otherwise good mothers. If these dams did connect (and occasionally calves were knocked over), it seemed to

be accidental. Initially, DH5 feinted at her calf, but later, as she became more excited, she started butting it. Finally, this heifer repeatedly charged her calf and frequently knocked it several feet away. Although DH6 frequently feinted at her calf, she never butted it.

On several occasions, while rejecting their calves' advances, both DH5 and DH6 rapidly switched the proximal part of their tails from side to side. This movement was never observed in any other dam.

(e) Resting behaviour

During the observation period, all dams and calves spent some time lying down. While it was common for a calf to be down while its dam was standing, it was far less common for a dam to be down while its calf was standing or walking around the box. In a total of approximately 4,600 minutes lying time shown by 28 of the 30 dams (i.e. excluding heifers DH5 and DH6), only about 500 minutes (11% of this time) were spent lying while their calves were standing or walking about.

Usually, if a dam was lying while its calf was standing, the dam appeared uneasy and usually watched the calf continuously. If the calf ventured too far away or in some cases approached the door of the calving box, the dam usually grunted once or twice and rose, approached the calf and then licked it. The distance that a calf moved away from its dam often appeared critical. If it rose and stood about less than about four feet from its recumbent dam, then she appeared relatively at ease, but once the calf moved more than eight feet away, the dam usually rose and approached it. Dams and calves usually lay down close together and only rarely more than four feet apart.

Twenty-one dams in the series (70%) did not lie down until their calves had either suckled or at least carried out prolonged teat seeking. Of the remaining nine dams, six (DH3, DH5, DH8, DC1, DC5 and DC10) did so before their calves stood. In all six cases, note had been taken that labour had been long and difficult, and, in fact, five calvings (all save that of DC10) had been completed from the recumbent position.

Discussion

Every beef cow in the series was judged to be at least four years old and was hence assumed to be an experienced mother. All helpers were found to be less than two years, three months of age (i.e. they had two permanent incisor teeth only) and were therefore assumed to be primiparous. The maternal experience of the dairy cows was unknown. In spite of these differences, with a very few exceptions, a strikingly similar behaviour pattern emerged.

It seemed that those dams calving from the recumbent position had generally longer and often more difficult parturitions.

The beef cows in the series appeared to be the best mothers in that their every function was carried out more vigorously and efficiently than most of the other dams. Every beef cow licked its calf immediately after birth, and carried on doing so for longer than the other two groups of dams. As a group, they accepted their calves' teat-seeking advances sooner than the other dams although these times were clouded by other variables. All beef cows ate contaminated bedding and cleansings and only one (9C6) lay down before its calf had carried out prolonged teat seeking.

It is probably significant that the two poor mothers were both dairy heifers. Nevertheless, one of the best mothers was another heifer, DH1 (one of the few dams in the series to exhibit definite maternal orientation in an effort to facilitate teat-seeking advances by her calf). As a group, the heifers spent significantly less time licking their calves immediately after birth than the other dams.

The abnormal behaviour of DH5 would seem to reinforce the suggestion by Collias (1956) and Smith (1966) that licking of the offspring has an important bearing on the fixing of the dam-offspring bond. Why this heifer should start licking its calf at 258 minutes post partum is an interesting question. It is perhaps significant that the calf was not teat seeking at the time that licking was timourously initiated, but was standing exhausted and motionless a few feet in front of its dam. Once licking started, it continued for six minutes and then suckling occurred after a further 37 minutes.

The early struggling by a calf seemed to be important in fixing the dam's attention upon her offspring.

In view of the fact that the dealer supplying the dairy cows and heifers had a problem of retained placentas under his conditions, it seemed surprising that all dams in the series had cleansed by ten hours post partum. However, Roberts (1956) reported that placental retention occurred in 22.7% of cows in one herd where suckling was not allowed. When suckling was allowed, the incidence dropped to 4.9%. It has been claimed that this phenomenon is due to the release of oxytoxin from the post-pituitary gland due to the stimulus of suckling (Hafez, 1962).

In this context, it is interesting to note (see Appendix 1, Table 2) that nine heifers cleansed at a relatively fixed time (232 ± 33 minutes (S.D.) post partum), while the tenth (DH6) did not cleanse until 480-600 minutes post partum. This helper never instituted nursing activities and never allowed suckling to take place.

Two of the three cows (DH6, DC1 and DC6) which did not eat their afterbirths proved to be abnormal dams in that DH6 almost continuously rejected its calf, while DC6 almost continually ignored its calf and rarely licked it. It may well be that grooming of the calf and eating of the placenta, followed as they both are by the eating of large amounts of bedding contaminated by membranes and fluid, have a common basic motivation.

The findings would seem to suggest that much of maternal behaviour is instinctive, but that the patterns are reinforced by experience. Probably, too, the suggestion by Hafez (1964) that in the primiparous female the instinctive aspects of early maternal behaviour are blocked by the pain and shock of parturition, is true.

Section II

Behaviour of the Calf

(a) Behaviour prior to standing

The first obvious movements exhibited by the majority of calves consisted of head shaking, snuffling and sneezing. This was often apparent as soon as a calf's shoulders were passed. Five calves, however (BC7, BC8, DH7, DH10 and DC10), in spite of being well licked by their dams, remained almost motionless for between seven and 32 minutes after birth.

Eventually, all calves rose into sternal recumbency, but the time that calves took to do this was extremely variable as can be seen by the following table.

TABLE 5

Time after Birth at which Calves Rose into Sternal Recumbency

<u>Time</u> (minutes post partum)	<u>≤5</u>	<u>≤10</u>	<u>≤20</u>
Beef cows' calves (10)	5	8	8
Dairy heifers' calves (10)	3	6	9
Dairy cows' calves (10)	<u>5</u>	<u>8</u>	<u>10</u>
Overall percentage	43.3%	73.3%	90%

The above table also shows that no real differences were demonstrable between the three groups of calves. Of the three late risers, calves BC7 and BC8 both attained sternal recumbency at 22 minutes, and DH7 at 50 minutes post partum. Note had been made that

in all three cases, labour had been prolonged and, in the case of dams BC7 and DH7, difficult.

Again, after a variable time, all calves started making definite attempts to rise. In every case, these attempts resembled the act of rising in older cattle (i.e. the hindquarters were raised first). In many cases, calves remained in a kneeling position for long periods. The time taken by calves to start making attempts to rise were as follows.

TABLE 6

Time after Birth when Calves Made Definite Attempts to Rise

<u>Time</u> (minutes post partum)	<u>≤10</u>	<u>≤30</u>	<u>≤60</u>
Beef cows' calves (10)	4	8	9
Dairy helpers' calves (10)	2	7	10
Dairy cows' calves (10)	<u>3</u>	<u>10</u>	<u>10</u>
Overall percentage	30%	83.3%	96.7%

Once again, a marked similarity exists between the three groups of calves. The one calf (BC8) which had not attempted to rise by 60 minutes started attempts at 65 minutes post partum.

Eventually all calves stood up, some with apparent ease. However, due to the many falls experienced early on by the majority of calves it was decided that to be counted as standing, calves had to remain on their feet for one minute (see Materials and Methods). As a result, some calves were on their feet, but only momentarily so before the stated standing time. Also, one calf (BC10) was up, but kneeling,

from 17 minutes post partum, although it did not stand until 75 minutes post partum. Another calf (DC2) was occasionally kneeling and often standing on the anterior aspects of its fore fetlocks long before 43 minutes post partum. As mentioned earlier, some calves were repeatedly knocked over by their dams during the grooming phase.

The times taken by individual calves are recorded, and the mean times for each group are compared in Table 7.

TABLE 7

The Time Taken for Calves to Stand

<u>Beef Cows' Calves</u>		<u>Dairy Heifers' Calves</u>		<u>Dairy Cows' Calves</u>	
<u>Calf</u>	<u>Time</u> (minutes)	<u>Calf</u>	<u>Time</u> (minutes)	<u>Calf</u>	<u>Time</u> (minutes)
BC1	29	DH1	21	DC1	52
BC2	45	DH2	30	DC2	43
BC3	16	DH3	99	DC3	55
BC4	26	DH4	22	DC4	54
BC5	26	DH5	69	DC5	77
BC6	20	DH6	31	DC6	76
BC7	55	DH7	79	DC7	61
BC8	65	DH8	261	DC8	65
BC9	37	DH9	45	DC9	23
BC10	35	DH10	70	DC10	75
Mean	35.4		72.7		58.1
S.D.	±14.8		±71.6		±20.6
S.E.	± 4.7		±22.7		± 6.5

A significant difference ($p = <0.02$) exists between the beef cows' and the dairy cows' calves.

In general, calves born from recumbent calvings were slower to stand than calves born from the standing position. This appeared to be largely due to the fact that when parturitions were completed from the recumbent position, the calves were delivered onto a large pool of amniotic fluid, and hence the area in which they attempted to rise was very slippery. Two other calves, BC8 (standing time 65 minutes) and DH8 (standing time 261 minutes) had difficulty standing for similar reasons. This was due in the first case to the fact that dam BC8 was a very quiet cow and hardly moved at all during labour; hence the amniotic fluid was concentrated in one area into which the calf was born. Secondly, dam DH8, a very nervous heifer, developed a habit of scraping at the bedding during labour and thereby cleaned the floor of straw, again making it difficult for calf DH8 to gain a footing.

(b) Teat-seeking behaviour

Once standing, most calves soon initiated teat-seeking advances at their dams (see Figures 3, 4 and 5). As mentioned in Section 1 (Maternal Behaviour) at this point the dams were generally standing in front of their calves, licking their heads. Six calves (BC5, BC6, DH4, DH6, DC1 and DC8), however, made initial teat-seeking advances not at their dams, but at the calving box walls and mangers. It is interesting to note that the dams of five of the six calves were at that time revealing slightly abnormal maternal behaviour. Dam DH4 was not licking the calf's head but its abdomen and was therefore not standing in front of it, and dam DH6 had persistently ignored her calf from birth and was standing nowhere near it as it commenced teat seeking. The dams of the remaining calves, BC6, DC1 and DC8 were all lying down as their calves commenced teat seeking.



Fig. 3. Teat seeking advances in an Aberdeen-Angus-cross-Beef Shorthorn calf aged approximately one-and-a-half hours. The posture is the typical one for teat seeking calves with the muzzle extended, the ears held back and the tail held slightly erect.



Fig. 4. Teat seeking advances by the same calf as in Fig. 3 some minutes later. It can be seen that the calf has reached the udder area but, due to the poor shape of its dam, its advances are directed above the level of the teats.



Fig. 5. Teat seeking advances by the same calf as in Figures 3 and 4. At this stage, the calf's advances have become focused on the area round the dam's forelegs (i.e. the highest part of the underbelly).

As a dam finally stood to accept its calf's advances, the calf would push along her body, exploring in turn shoulder, brisket, axilla, body wall, udder or groin and thigh. Frequently, a calf at this stage, pushed on past its dam's thighs and continued until it reached a wall, then continued along the wall until it ended pushing at the corner. Later, pushing became increasingly orientated towards the dam although an occasional calf (e.g. DC1) repeatedly ceased pushing its dam in order to push at the box walls.

Pushing was largely directed at the dam's fore or hind quarter and there was an obvious preference on the calf's part to thrust its nose high and, if possible, into a recess (for example, axilla or groin). The area of a dam's body where a calf concentrated its teat-seeking activities quite definitely depended on the shape of its dam's underbelly. When the udder and teats were situated at the highest part of the underbelly (in five beef cows, nine dairy heifers, and only one dairy cow) pushing was mainly, sometimes almost exclusively, concentrated on the udder area. In the remaining dams the abdomen and udder were large, and hence the highest part of the underbelly was the xiphoid-axillary region, and consequently teat seeking was frequently carried out around the forelegs. If in these cases pushing occurred around the udder, the calf's nose was usually thrust high above the teats into the dam's groin or up the dam's flank. Dams were classed as either "good shaped" (i.e. optimal shape for suckling) or "poor shaped" prior to parturition (see Materials and Methods). The effect of dam shape on the total teat-seeking times of calves prior to first suckling is recorded in Table 8.

TABLE 8

Total Teat-Seeking Time of Calves Prior to First Suckling
Effect of Dams' Shape

<u>Dam Shape</u>	<u>No. of Calves</u>	<u>Mean Teat- Seeking Time (minutes)</u>	<u>S.D.</u>	<u>S.E.</u>
* Good	12	17.1	±10.7	±3.09
				$p = <0.02$
Poor	10	39.6	±25.6	±8.1

- * The calf from heifer DH5 was excluded from the "good-shaped" dams because, in spite of the fact that suckling finally took place, the calf's early teat-seeking advances were consistently and aggressively rejected by the dam.

As can be seen from Table 8, the effect of dam shape had a significant effect ($p = <0.02$) upon the total teat-seeking time prior to first suckling in the above calves.

Once calves had suckled, they spent considerably less time teat seeking prior to the next spell of suckling. In fact, out of 15 calves which suckled twice during the observation period, eight calves did not carry out teat-seeking advances at all prior to the second suckling but moved directly to their dams' udders and commenced suckling. The mean teat-seeking times prior to the first and second suckling spells are compared in Table 9.

TABLE 9

Teat-Seeking Times Prior to First and Second Suckling Spells

	<u>No. of Calves</u>	<u>Mean Teat- Seeking Time</u> (minutes)	<u>S.D.</u>	<u>S.E.</u>
First suckling spell	15	19.3	±9.6	±2.5
p = <0.001				
* Second suckling spell	7	5.0	±2.5	±0.9
* Eight calves did not teat seek at all prior to the second suckling spell.				

It is clear from Table 9 that significantly less time ($p = <0.001$) was spent teat seeking after calves had previously experienced one suckling spell.

Two calves (DH1 and DH10) were observed to be weak teat seekers from birth. Neither made strong advances to its dam and both were easily put off by the dam moving or by a gentle kick. Calf DH1 spent long periods standing, not pushing, with its nose among its dam's teats and moreover did not suckle during the observation period. Calf DH10 was very similar but did finally suckle for $4\frac{1}{2}$ minutes at 363 minutes post partum.

(c) Suckling behaviour

Actual suckling occurred during the observation period in 23 of the 30 calves (76.7%). The times taken for individual calves to first suckle are presented in Table 10.

TABLE 10

The Times Taken by Calves to First Suckle Their Dam

<u>Beef Cows' Calves</u>		<u>Dairy Heifers' Calves</u>		<u>Dairy Cows' Calves</u>	
<u>Calf</u>	<u>Time</u> (minutes)	<u>Calf</u>	<u>Time</u> (minutes)	<u>Calf</u>	<u>Time</u> (minutes)
BC1	45	DH1	*	DC1	*
BC2	58	DH2	60	DC2	170
BC3	35	DH3	299	DC3	421
BC4	*	DH4	104	DC4	258
BC5	39	DH5	301	DC5	358
BC6	180	DH6	*	DC6	414
BC7	70	DH7	110	DC7	116
BC8	158	DH8	346	DC8	91
BC9	66	DH9	163	DC9	*
BC10	*	DH10	363	DC10	*
Mean	81.4		218.3		261.1
S.D.	±52.2		±113.8		±129.1
S.E.	±18.5		±40.2		±48.7

Significance: Beef cows' and dairy heifers' calves $p = < 0.01$
 Beef cows' and dairy cows' calves $p = < 0.01$
 Dairy cows' and dairy heifers' calves n/s

* These calves did not suckle during the eight hour observation period.

It can be seen that the beef cows' calves were significantly faster ($p = < 0.01$) to suckle than either the dairy heifers' or dairy cows' calves. However, a few calves in each group did not suckle during observation, and in some others the time to first suckling appeared to be delayed. However, delayed suckling was far less apparent in the beef cows' calves. In fact, all of the eight calves which suckled during the observation period did so before they were three hours of age. Calf BC3 suckled earlier than any other, at 35 minutes post partum.

The position taken up by suckling calves was very constant. Although teat seeking was frequently carried out with an angle between the long axes of dam and calf, directly suckling commenced the calf tended to move its body closer to the dam, until their bodies were actually touching. The tendency for dams to lick the hindquarters of suckling calves also served to keep them closer together. Only two calves (DC2 and DC7) were seen to suckle their dams from behind and suckling from this position was not continued for long.

Twenty-one of the 23 calves which suckled during the observation period (91.3%) started by suckling one of the anterior teats. Moreover, there was a marked tendency in those calves suckling more than once to suckle from the same side of the dam as previously. Of the 16 calves that suckled more than once during observation, eight (50%) suckled from only one side of the dam and the remainder spent most of their suckling time on the side of the dam from whence they first obtained colostrum.

The number of suckling spells during the first eight hours of post partum varied from none to four. Most calves which suckled early, suckled more often than the late suckling calves. The mean total time spent suckling and apparently obtaining milk in the 23 calves was 16.7 ± 9.2 minutes (S.D.).

While the flow of colostrum during suckling appeared to be satisfying a calf, it fed quietly with the tail held slightly erect and waving gently from side to side. Rapid tail wagging and bunting did not occur except during teat seeking or when it seemed the milk flow was reduced or had stopped.

(d) Factors responsible for delayed suckling

It has already been shown that calves born to poor-shaped dams spent significantly longer at teat seeking than those calves born to good-shaped dams. This difference was consequently reflected in the times to first suckling as can be seen by the following table.

TABLE 11The Overall Effect of Dam Shape on Calves' Times to First Suckling

<u>Calves from Poor-Shaped Dams</u> (15)		<u>Calves from Good-Shaped Dams</u> (15)	
<u>Calf</u>	<u>Suckling Time</u> (minutes)	<u>Calf</u>	<u>Suckling Time</u> (minutes)
BC4	*	DC1	45
BC6	180	DC2	58
BC8	158	DC3	35
BC9	66	DC5	39
BC10	*	DC7	70
DH2	60	DH1	*
DC1	*	DH3	299
DC2	170	DH4	104
DC3	421	DH5	301
DC4	258	DH6	*
DC5	358	DH7	110
DC6	414	DH8	346
DC7	116	DH9	163
DC8	*	DH10	363
DC10	*	DC8	91
Mean	220.1		155.7
S.D.	±129.1		±120.0
S.E.	± 40.9		± 33.2

* These calves did not suckle during the observation period.

Dam shape, per se, was found not to exert a significant effect on the times to first suckling in those calves which actually suckled during the observation period. However, seven calves failed to suckle during observation, and the reason for this in five of these (BC4, BC10, DC1, DC9 and DC10) was considered by the author to be poor dam shape. Moreover, several other factors delayed or prevented suckling in some of the calves from good shaped dams. These other factors, significantly perhaps, occurred only in calves born to dairy heifers.

Two calves (DH1 and DH10) have already been discussed in relation to their weak teat-seeking drive. Calf DH10 suckled for only $4\frac{1}{2}$ minutes at 363 minutes post partum, and DH1 did not suckle at all. A further two calves (DH3 and DH8) were born onto very slippery straw and consequently their standing times were delayed until 99 minutes and 261 minutes respectively. On finally rising, DH8 made immediate teat-seeking advances to its dam, but DH3 seemed to have weakened itself in its efforts to rise and teat seeking was also delayed. Eventually, both calves were seen to suckle, DH3 at 299 minutes, and DH8 at 346 minutes post partum respectively. Finally, two dams (DH5 and DH6) rejected their calves' teat-seeking advances. Dam DH5 did finally allow suckling to occur at 301 minutes post partum, but calf DH6 never suckled during the observation period.

The above six dairy heifers' calves (DH1, DH3, DH5, DH6, DH8 and DH10) were excluded from the group of calves born to good-shaped dams in order to demonstrate more clearly the effect of dam shape on the time to first suckling. The results appear in Table 12.

TABLE 12

The Effect of Dam Shape on Calves' Times to First Suckling When Other Factors Delaying or Preventing Suckling in Six Dairy Heifers' Calves Are Excluded

<u>Calves from Poor-Shaped Dams</u> (10)		<u>Calves from Good-Shaped Dams</u> (9)	
<u>Calf</u>	<u>Suckling Time</u> (minutes)	<u>Calf</u>	<u>Suckling Time</u> (minutes)
BC6	180	BC1	45
BC8	158	BC2	58
BC9	66	BC3	35
DH2	60	BC5	39
DC2	170	BC7	70
DC3	421	DH4	91
DC4	258	DH7	104
DC5	358	DH9	110
DC6	414	DC8	163
DC7	116		
Mean	220.1		79.4
S.D.	±129.1		±41.8
S.E.	± 40.9		±13.9

Significance: $p \geq 0.01$

A highly significant difference ($p = <0.01$) was therefore demonstrated between the times to first suckling in calves from poor- and good-shaped dams, provided the other variables outlined above were excluded.

Apart from the shape and size of the dams' underbelly and udder, the size and position of teats seemed to be of some minor degree of importance with regard to early suckling. If they were too long or too large, calves had difficulty getting the ends into their mouths. Several calves spent a long time mouthing and nibbling teats that were too large.

Discussion

There seemed to be no appreciable difference between the three groups of calves regarding the time taken to rise into sternal recumbency or to start making attempts to rise onto their feet. However, the beef calves attained the standing position significantly earlier than the others and here, it must be remembered from Section I (Maternal Behaviour) that these calves were initially licked by their dams for a significantly longer period following birth than at least the dairy helpers' calves, and that every beef calf was well licked. In view of the observations on lambs already cited by Herscher, *et al.* (1963) and Blauvelt (1954), it may well be that early vigorous and prolonged licking by the dams does have a marked stimulating effect on a newborn calf. In a study such as this, it was, of course, impossible to measure the birthweights of the calves. It did appear, however, that the beef calves were, in general, smaller than the others and this, too, may have made it easier for them to rise.

The teat-seeking postures adopted by the calves were just as described by Fraser (1962) and very similar to those shown in photographs of other herbivores (Hafez, 1962). On rising, most calves moved towards their dams as stated by Adler, *et al.* (1958) and Smith (1966), but usually the dams had already positioned themselves in front of their offspring at this stage. In five of the six instances when calves moved away from their dams, the dams were not directly in front of their calves as the latter rose.

All of the calves in the series showed a preference for thrusting their noses high under their dams and, if possible, into a recess. Two calves exhibited weak teat-seeking drives, but nevertheless this instinct was still obvious.

The shape of the dam's underbelly was all-important in regard to where teat-seeking advances were concentrated. Where the abdomen and udder were large or pendulous, teat seeking was frequently carried out around the forelegs or high above the teats. Where the udder was the highest point of the dam's underbelly, the calves quickly "homed" on to the udder and suckling quickly occurred in most cases. A significant difference was demonstrated between the calves' total teat-seeking time prior to first suckling when dams were divided into good shapes or poor shapes from the viewpoint of suckling.

Reference to photographs of many wild ungulates and horses (Sanderson, 1959, and the Larousse Encyclopaedia of Animal Life, 1967) together with observations on captive ungulates (Selman, unpublished observations) indicates that in all cases the udder is at the highest part of the female's underbelly. Dairy helpers, and most beef cattle, usually resemble wild ungulates in this respect. The dairy cow, however, has been evolved for purposes other than suckling and consequently, the udder, and to a lesser extent, the abdomen, have been abnormally developed. It would seem that this fairly recent development has rendered the classic dairy cow unsuitable for suckling. Calves born to these dams still instinctively thrust their muzzles high and as a result teat seeking is prolonged and suckling may be delayed or may not occur at all.

Calves quickly learned to suckle with the result that after one successful suckling, teat seeking was either markedly reduced or absent prior to the second feed. This, and the fact that calves tended to favour one side of their dam for suckling, and that the side where colostrum was first obtained, would support the suggestion by Alexander and Williams (1966) that increased suckling efficiency was due to a reward mechanism.

Suckling occurred during the first eight hours post partum in 23 out of 30 calves (76.7%), but the time to first suckling was extremely variable. The beef cows' calves suckled significantly earlier than the other two groups of calves, but the former were standing significantly earlier too, and every beef cow stood very early to its calf's advances.

The factors responsible for delayed suckling were as follows:

- (i) Poor dam shape (15 cases)
- (ii) Calves with weak teat-seeking drives (2 cases)
- (iii) Abnormal maternal behaviour (2 cases)
- (iv) Delayed standing (2 cases)

The six calves where delay in suckling was due to either weak calves, abnormal mothers or delayed standing, were all dairy helpers' calves. Of the seven calves which did not suckle at all during observation, five were inhibited by extremely poor dam shape, one had a very weak teat-seeking drive, and the advances of the seventh were continually rejected by its very abnormal dam.

These observations have shown that calves left in a loose box with their dams for eight hours post partum will not necessarily suckle during that time.

Section III

The Serum Immune Globulin Concentrations of 30 Calves Left with Dams for the First 48 Hours of Life

The results of the investigations into the serum immune globulin concentrations of the dairy calves and of the beef calves in this series are presented separately in the following section. The decision to do this was for two reasons. First, as all of the previous work in this unit had concerned dairy calves, it was felt that from a comparative viewpoint, the dairy calf values were of more value grouped alone. Second, the vastly different selection, feeding and management practices that are applied to dairy and beef cows makes them two probably very different populations. In the absence of any previously reported work on the immune globulins of suckled beef calves, or on the immune lactoglobulin content of their dams' colostrum, it was felt that a separate presentation might be more informative and possibly might demonstrate differences between the two types of calf.

(a) Dairy calves

The 48-hour serum immune globulin concentrations of the 20 calves born to the dairy heifers and dairy cows in the series were as follows.

TABLE 13

Serum Immune Globulin Levels of Dairy Calves Left
with Their Dams for 48 Hours Post Partum

<u>Dairy Heifers' Calves</u>		<u>Dairy Cows' Calves</u>	
<u>Calf</u>	<u>48-Hour Serum Immune Globulin Concentration (2.S.T. Units)</u>	<u>Calf</u>	<u>48-Hour Serum Immune Globulin Concentration (2.S.T. Units)</u>
DH1	1	DC1	3
DH2	23	DC2	30
DH3	20	DC3	30
DH4	50	DC4	35
DH5	35	DC5	39
DH6	1	DC6	11
DH7	29	DC7	49
DH8	22	DC8	46
DH9	25	DC9	6
DH10	4	DC10	5
Mean	22.1		25.5
S.D.	±11.3		±16.8
S.E.	± 3.6		± 5.3

Significance: n.s.

It can be readily seen that a wide individual variation was found to exist in the immune globulin concentrations of the above calves and that seven (DH1, DH6, DH10, DC1, DC6, DC9 and DC10) had negligible or very low serum levels of immune globulin. However, only two of these calves (DH10 and DC6) actually suckled during the observation period.

The 48-hour serum immune globulin concentrations of the 15 calves suckling during the first eight hours of life are compared in Table 14 with those values in five calves which were observed not to suckle during this time.

TABLE 14

A Comparison of the Serum Immune Globulin Levels of 15 Dairy Calves Observed to Suckle before Eight Hours of Age and Five Calves Observed Not to Suckle

<u>Calves Suckling before 8 Hours</u> <u>Post Partum</u>				<u>Calves Not Suckling Before</u> <u>8 Hours Post Partum</u>	
<u>Calf</u>	<u>Serum Immune</u> <u>Globulin</u> <u>Concentration</u> <u>(Z.S.T. Units)</u>	<u>Calf</u>	<u>Serum Immune</u> <u>Globulin</u> <u>Concentration</u> <u>(Z.S.T. Units)</u>	<u>Calf</u>	<u>Serum Immune</u> <u>Globulin</u> <u>Concentration</u> <u>(Z.S.T. Units)</u>
DH2	23	DC2	30	DH1	1
DH3	20	DC3	30	DH6	1
DH4	50	DC4	36	DC1	3
DH5	35	DC5	39	DC9	6
DH7	29	DC6	11	DC10	5
DH8	22	DC7	49		
DH9	25	DC8	46		
DH10	4				
Mean	29.9				3.2
S.D.	±12.7				±2.3
S.E.	±3.3				±1.0

Significance: $p = < 0.001$

A highly significant difference was found to exist between the two groups of calves depending on whether they had suckled before eight hours of age or not. Nevertheless, two calves (DH10 and DC6) were found to have low values in spite of the fact that they were observed to suckle at 363 minutes and 414 minutes post partum respectively.

Calf DH10 was quite obviously a calf with a very weak teat-seeking drive. Its dam was of a good shape for suckling and good mother, but nevertheless suckling occurred late in the observation period due solely to the fact that the calf never made determined

efforts to suckle. When suckling did finally occur, it was only continued for four-and-a-half minutes. Calf DC6 appeared to be a strong calf and the only abnormality associated with either it or its dam was that the dam was never keen to lick it. This calf's teat-seeking advances were never rejected and frequently, although the dam did not show great interest in her calf, she grunted at it in the normal mothering voice. Suckling finally occurred at 414 minutes post partum, was continued for five minutes, and appeared to satiate the calf.

To determine the effect of time of first suckling on the 48-hour serum immune globulin a graph was constructed (see Figure 6).

A significant negative correlation ($r = -0.52$, $p = <0.05$) was found to exist between the time of first suckling and the 48-hour serum concentrations of immune globulin in these dairy calves.

This series of observations was carried out between the months of January and July (inclusive). However, in this admittedly small series of 20 dairy calves, no suggestion of a seasonal (upward) trend in serum immune globulin concentrations was noted when the mean zinc sulphate values of calves born during the winter were compared with those of the spring and summer born calves (see Table 15).

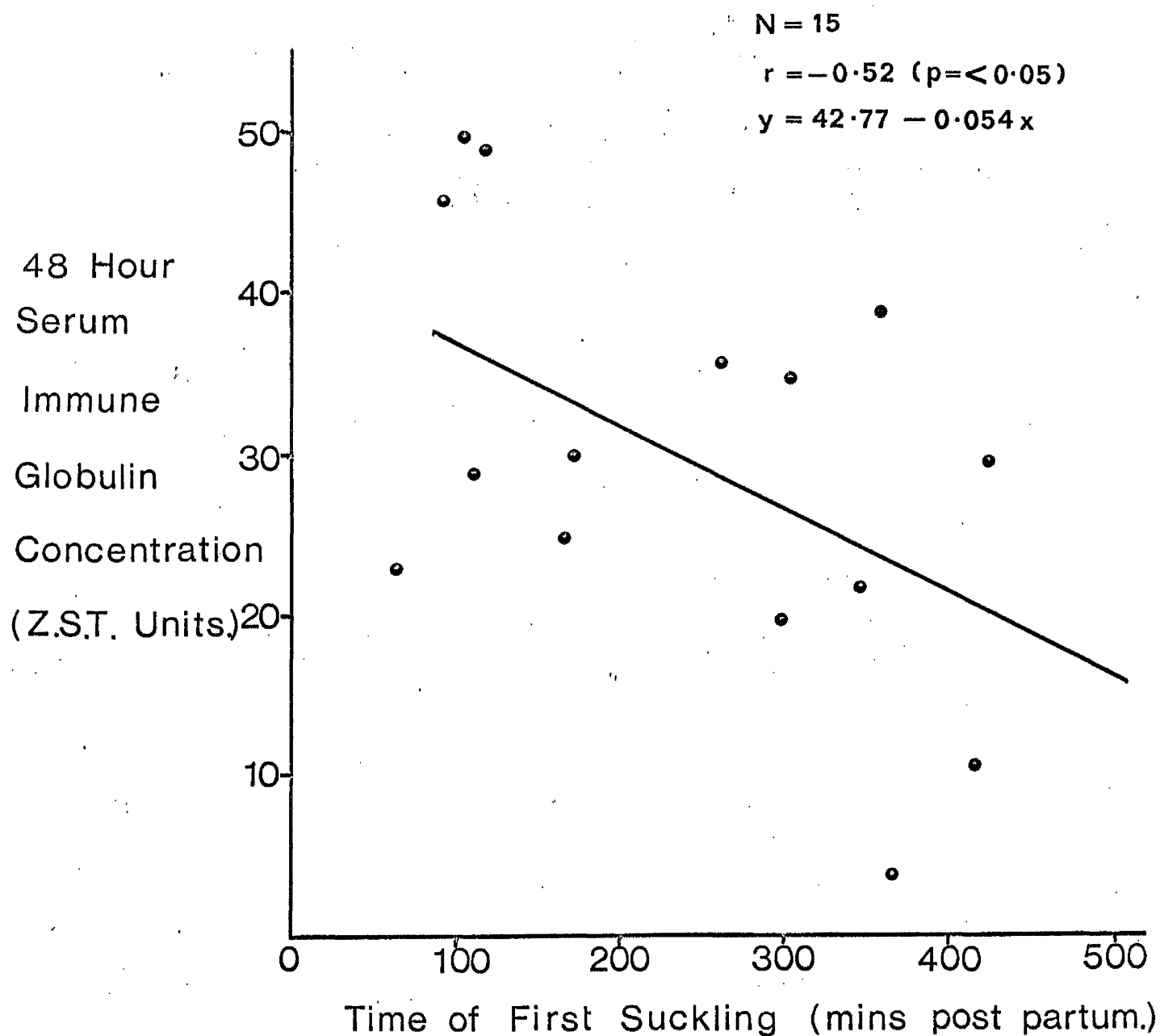


Fig. 6. The effect of time of first suckling on the 48-hour serum immune globulin concentration of fifteen newborn dairy calves.

TABLE 15

The Effect of Season on the Mean Serum Immune Globulin Concentrations of Twenty Calves Born in a Box and Left with Their Dams for 48 Hours

	<u>Mean</u>	<u>S.D.</u>	<u>S.E.</u>	<u>Significance</u>
Serum Immune Globulin Concentration (Z.S.T. Units) in Eleven Calves Born Between January and April (Inclusive)	25.3	± 14.7	± 4.4	
				n.s.
Serum Immune Globulin Concentration (Z.S.T. Units) in Nine Calves Born Between May and July (Inclusive)	20.8	± 18.2	± 6.1	

(b) Beef calves

The 48-hour serum immune globulin concentrations of ten beef calves in the series were as follows.

TABLE 16

Serum Immune Globulin Concentrations of Beef Calves
with Their Dams for 48 Hours Post Partum

<u>Calf</u>	<u>48-Hour Serum Immune</u> <u>Globulin Concentration</u> (Z.S.T. Units)
BC1	24
BC2	14
BC3	38
BC4	19
BC5	7
BC6	29
BC7	32
BC8	24
BC9	33
BC10	1
Mean	22.1
S.D.	± 11.3
S.E.	± 3.6

If the above concentrations of serum immune globulin are compared with the levels (see Table 13) obtained from the dairy calves, it can be shown that no significant difference exists between the three groups of calves. Once again, a very wide individual variation in serum immune globulin concentration is apparent.

It was found (see Table 10) that the beef calves were significantly faster to first suckling than either of the two groups of calves. Moreover, all of the eight beef calves which suckled during the observation period, did so before they were three hours of age. In view of the fact that in the dairy calves a significant negative relationship was found between the time of first suckling and

the 48-hour serum immune globulin concentration (see Figure 6), the eight beef calves, due to their significantly earlier suckling, might therefore have been expected to attain very high concentrations. Consequently, the 48-hour serum immune globulin concentrations of these beef calves were compared with the concentrations found in the seven dairy calves which suckled before three hours post partum. The result is presented below in Table 17.

TABLE 17

A Comparison of the 48-Hour Serum Immune Globulin Levels of Eight Beef Calves and Seven Dairy Calves All of Which Suckled before Three Hours Post Partum

<u>Calves</u>	<u>No.</u>	<u>Mean Serum Immune Globulin Concentration (Z.S.T. Units)</u>	<u>S.D.</u>	<u>S.E.</u>
Beef	8	25.1	± 9.7	±3.4
Dairy	7	36.0	±11.0	±4.2

n.s.

The marked difference in the mean 48-hour serum immune globulin concentrations of the two groups of calves was found to be not significant. Note must also be made, however, of the low 48-hour serum immune globulin concentrations of two of the beef calves, BC2 and BC5, in spite of the fact that they suckled at 58 minutes and 39 minutes post partum, respectively.

Discussion

A wide variation was found to exist in the serum immune globulin levels of 20 dairy calves left with their dams in a loose box for the first 48 hours of life, and seven of these calves were subsequently found to have low or negligible amounts of circulating immune globulin. These findings agree with those of Smith (1962 and 1965) and McEwan (1966). Five of the seven calves with low immune globulin levels had not suckled during the observation period, but two (DH10 and DC6) had done so.

Calf DH10 was a calf with a weak suckling drive, and although it finally suckled at 363 minutes post partum, it only did so weakly, for four-and-a-half minutes. Calf DC6 was poorly mothered and rarely licked by its dam and, again, suckling occurred late during the observation spell, at 414 minutes post partum, and lasted for five minutes. It is interesting to note (see Appendix 1, Tables 5 and 6) that in both of these calves, the total suckling time was considerably shorter than the mean total suckling time of 16.7 ± 9.2 minutes as seen in the other 21 calves that suckled. The wide individual variation in 48-hour serum immune globulin concentrations was also found to exist in the beef calves, even though the eight calves which were observed to suckle, did so before three hours post partum. Two calves (BC2 and BC5) which suckled within one hour of birth and at least once more during the observation were finally found to have low levels of serum immune globulin.

In spite of the fact that the 20 dairy calves in the series must have been ingesting varying amounts of colostrum of varying immune lactoglobulin content, a significant correlation ($p = < 0.05$) was found to exist between the time of first suckling and the 48-hour serum immune globulin concentration. It must, however, be stressed here that those calves suckling early generally suckled at least once more during the observation so that this correlation might in part be due to frequency of suckling. Improved absorptions of E. coli somatic antibody has been found by Kaeckenbeeck, Colinet and Shoenaers (1961) to result from repeated feeding of small quantities (i.e. 200 ml) of colostrum. However, this question will be discussed during later parts of this thesis.

In view of the above finding in the dairy calves, it is surprising that the eight beef calves which suckled during the first three hours of life did not attain extremely high serum immune globulin concentrations. In fact, their mean serum concentration was very much lower than that of the seven dairy calves which suckled within three hours of birth. Although this difference was found to be not statistically significant, the relatively small number of calves involved and the very wide individual variations in serum concentrations of immune globulin must be considered. Therefore, in spite of the lack of statistical corroboration, the possibility that relatively poorer absorptions of immune lactoglobulin may occur in early suckling beef calves must be borne in mind.

The fact that a very pronounced seasonal variation in mean serum immune globulin concentration occurred in the Ayrshire bull calves in the West of Scotland has already been mentioned (Gay, et al., 1965b). These workers found very low mean levels during the winter months. In the present study, high, often very high, immune globulin concentrations were found during January to April. Admittedly, the number of animals in this present study was small, but generally suckling produced high immune globulin levels, and there was no difference between the levels in calves born from January to April and in those born from May to July.

It would seem, from the results presented in Section III, that the early management of a calf plays a great part in determining its 48-hour serum immune globulin concentration. Allowing dairy calves to suckle colostrum from their dams during the winter months, when this usually is not possible under the traditional form of dairy calf management in this area, results in serum immune globulin concentrations that are higher than those quoted for the same time of the year in the same area (Gay, et al., 1965b). In the present study no significant difference was found between the mean serum immune globulin concentrations of winter-born dairy calves and those of spring- and summer-born dairy calves. This suggests that the seasonal variation noted by the above workers is purely a function of the difference early management of winter- and summer-born calves.

Smith, et al. (1967) showed, in a survey of 190 home-bred calves, that only a slight variation was found to occur when samples were grouped on a quarterly basis, in comparison with that shown to occur in

a monthly survey of calves in the West of Scotland (Gay, et al., 1965b). However, in at least one of their areas (i.e. East Sussex) winter calving is usually carried out in a box or yard (Salmon, unpublished observations) and rarely do cows calve while neck tied in a byre. The fact that many of these calves in East Sussex, at least, suckle their dams before they are separated will almost certainly serve to minimise the seasonal variation.

The recent survey on calf wastage and husbandry in Britain, 1962-63 (Leach, et al., 1966) found that calves obtaining colostrum by suckling their dams stood a far higher chance of survival than those that had been bucket fed colostrum. It also established that calves born in a field were more likely to survive than those born indoors at the same time of year. When the findings of the present study are viewed together with the various other observations already discussed, it would seem that the important common factor of suckling colostrum and calving in a field (and hence probably suckling colostrum) is the resulting higher serum immune globulin levels.

Unfortunately, the Weybridge survey did not differentiate between calves born indoors in a box and calves born indoors in a byre with the dam tied by the neck. If it had, it is tempting, in view of the present findings, to postulate that a higher percentage of calves with low serum immune globulin levels (and hence higher mortality rates) occurred in the byre-born calves.

On the other hand, allowing a calf the opportunity of suckling does not necessarily guarantee higher serum immune globulin concentrations and the findings of this study support the view by Smith (1962) that it is necessary to ensure that calves obtain colostrum in very early life and not to assume that they will do so if left with their mothers. In fact, five dairy calves out of twenty (25%) failed to suckle within the first eight hours of life, and these calves had negligible immune globulin levels at 48 hours of age. It therefore appears that if a calf has not suckled within eight hours of birth, it is unlikely to learn to do so before 'colostral shutdown' occurs.

PART 2

PART 2

STUDIES ON CALVES ALLOWED TO SUCKLE THEIR DAMS AT FIXED TIMES POST PARTUM

Introduction and Review of the Literature

In Part 1, attention was drawn to the high mean levels of the serum immune globulins in market calves in the summer months in Scotland as observed by Gay, et al. (1965b). The observations of Smith, et al. (1967) that suckling colostrum generally produced higher absorptions of immune lactoglobulin than did bucket feeding were also noted. The higher levels of immune globulin resulting from suckling might be due to either earlier feeding or higher intakes of colostrum. Little has previously been published regarding the time of first suckling in calves although it is generally assumed that most calves will have suckled by five to six hours post partum (Walker, 1950, and Hafez, 1962). This is earlier than most calves are bucket fed colostrum in the West of Scotland (Selman, de la Fuente and Fisher, 1968).

Although general advice on calf rearing always includes a reference to the feeding of colostrum, little information regarding the amounts to be fed is available. Hector and Rowat (1948) in a survey of 84 closed herds in Dumfriesshire found that the amounts of colostrum bucket fed to calves varied considerably. Some farmers apparently fed to satiation, but more usually one to four pints per day were fed, with the majority of farmers favouring a feed of one pint twice daily.

Aschaffenberg, et al. (1949a) fed newborn calves on varying amounts of the aqueous fraction of colostrum and found that the best performances occurred in those calves which obtained 7200 ml of this fraction. The improved performance in these calves was almost entirely due to a lower incidence of diarrhoea. Roy, Shillam, Hawkins and Lang (1958) fed their experimental calves on "not more than six pints of colostrum." Flinscher (1963) stated that because of the fact that colostrum itself caused severe diarrhoea, calves should be kept slightly hungry over the first few days of life. He went on to recommend feeding 4 lbs.-7 lbs. per day depending on the breed of the calf.

It is generally recommended that slightly older calves should be fed at the rate of 1 lb. of milk/10 lbs. bodyweight/24 hours (Walker, 1950, B.V.A. Handbook, "The Husbandry and Diseases of Calves," and Mylrea, 1966). However, both Walker (1950) and Mylrea (1966) showed that when milk was offered ad libitum from a bucket, intakes were generally much higher than this. Mylrea (1966) found that the daily milk intake of young calves increased from 10%, while restricted, to as high as 26% of their bodyweight when offered ad libitum. However, calves soon settled to ad libitum feeding and daily milk intake became steady at between 17%-19% of their bodyweight.

Very little information is available regarding the amounts of colostrum ingested by suckling by newborn calves. Kuttner and Ratner (1923) stated that "the cow's udder at the time of parturition is filled with approximately 20 lbs. of colostrum which is immediately and reflexly ingested by the calf shortly after birth to the extent of

about 2 lbs. at the first feeding." No data is presented to support this assertion. Walker (1950) test weighed two newborn dairy shorthorn calves before and after suckling their dams and found that very large amounts of colostrum were ingested over the first four days of life. The total amounts of colostrum consumed by each calf daily are presented in Table 18, together with some other information.

TABLE 18

(From Walker, 1950)

	<u>Total Colostrum Consumed by Calf/Day (lbs.)</u>	<u>No. of Feeds</u>	<u>Average per Feed (lbs.)</u>	<u>Total Production by Cow (lbs.)</u>
<u>Calf I: 90 lbs. birthweight</u>				
1st Day	18.5	4	4.6	18.5
2nd Day	13.75	6	2.3	28.0
3rd Day	13.00	7	1.9	21.0
4th Day	21.75	4	5.4	34.0
<u>Calf II: 90 lbs. birthweight</u>				
1st Day	14.75	8	1.8	23.4
2nd Day	16.0	6	2.7	25.7
3rd Day	18.75	5	3.7	27.1
4th Day	26.25	5	5.2	33.7

No work has been published to date comparing the amounts of colostrum ingested by suckling with the amount of immune lactoglobulin subsequently absorbed. However, McEwan (1968) has found that under conditions of early bucket feeding of colostrum, a linear relationship existed between the serum immune globulin concentration at 72 hours post partum and the weight of immune lactoglobulin presented to the

calf when due allowance has been made for the variations in the birthweights of calves. This might explain the generally higher serum immune globulin concentrations in calves which obtain colostrum by suckling if the very large amounts ingested by the two calves studied by Walker (1950) are representative of the intakes of suckling calves in general.

It was consequently decided that a study of the amounts consumed by calves suckling their own dams at fixed times post partum was necessary. An attempt to correlate, if possible, the amounts ingested with the resulting serum immune globulin concentration was also undertaken.

It was readily appreciated that removing calves from their dams soon after birth and then reintroducing them only for suckling at six hours and 12 hours post partum was in no way comparable to leaving calves together with their dams for the whole of the first 12 or more hours of life. It was felt that the lack of continual grooming and cossetting by the dam might, for example, affect the amounts of colostrum ingested by the calf. A further group of ten "mothered calves" was therefore planned. These stayed with their dams for the whole of the first 24 hours of life, but were muzzled except for their suckling spells at six hours and 12 hours post partum.

The results of these investigations are presented in Sections I and II of Part 2 of this thesis.

MATERIALS AND METHODS

1. Experimental Animals

(a) Cows and helpers

All of the cows and helpers used in the following series of experiments were of the Ayrshire breed, and all were obtained from the same source as those used in Part I. All remained healthy while in the Veterinary Hospital, and none showed clinically detectable abnormalities of udder, teats or milk. In a few cases, assistance was required at parturition; but where more than gentle traction was required to deliver a calf, it was not used in the experiment (gentle traction was the term applied to traction performed by one man only on ropes tied to a calf's forelegs).

(b) Calves

The calves used in the following experiments were as described in Materials and Methods, Part I.

2. Care of Experimental Animals

(a) Accommodation

The calving accommodation was as described in Materials and Methods, Part I. The dams stayed in these boxes for a variable period depending on the times that their respective calves suckled. When the calves were separated from their dams between suckling spells, the dams were often removed from the observation boxes and returned to a holding box or byre immediately after the twelve hour suckling spell.

When the calves were muzzled and stayed with their dams between suckling spells, they both remained in the loose box for at least 12 hours following the last period of suckling. When calves were maintained away from their dams between suckling, they were housed in individual pens in a thermostatically heated room where the temperature was constantly kept between 60°F and 70°F (see Materials and Methods, Part 3).

(b) Feeding

The feeding of adults was as described in Materials and Methods, Part 1.

Calves were first offered milk at approximately 12 hours after their last suckling spell. This milk was obtained from the same source, and fed at the same rate, as described in Materials and Methods, Part 1. Frequently, the first attempt to feed milk coincided with the diarrhoeic phase described in Materials and Methods, Part 1, and this made several calves somewhat disinclined to take milk. However, by 30-36 hours post partum, all calves were taking milk readily from a bucket.

(c) Milking

The milking procedures were as described in Materials and Methods, Part 1.

3. Experimental Procedures

(a) Prediction of parturition

The method of cervical examination described in Materials and Methods, Part 1, was used throughout this experiment.

(b) Weighing calves

Calves were weighed immediately before and after each suckling spell on an "Avery" scale weighing up to 135 lbs. in 1/4 lb. divisions (Avery, Birmingham, England, Type 3203 CLE), and their bodyweights to the nearest 1/4 lb. were recorded.

No calf urinated during suckling but the occasional calf did pass meconium. When this happened, the cow was prevented from licking it up, and it was collected on a sheet of paper and weighed with the calf after suckling was completed.

(c) Management of calves

All calves were licked for 15 minutes by their dams and then, depending on the experiment, were either removed to individual pens in a separate room as gently as possible, or were muzzled (see below) and left with their dams.

At 6 hours and, when necessary, 12 hours post partum, the calves were weighed and then encouraged to suckle their respective dams which at this stage were haltered. Those calves which had been separated from their dams between sucklings appeared to be recognised by their dams, but it was quickly found that the cows settled to their calves' teat-seeking advances and suckling more quickly if allowed to lick their calves for a moment.

Calves were initially held onto a teat and when they were slow to suckle, some colostrum was squirted into their mouths and the teat was then placed between the upper and lower lips. Occasionally some time elapsed before calves themselves initiated strong suckling movements. Calves were constantly attended during each suckling spell,

and sometimes they had to be guided back to a teat if suckling was interrupted. If a calf had apparently emptied one quarter, and yet still appeared to want to suckle, it was guided to another teat. The point of satiation was judged to be reached when a calf no longer showed an inclination to suckle (i.e. when teat-seeking advances were no longer made).

Considerable difficulty was experienced in getting those calves that were maintained away from their dams to stand and to suckle, whereas mothered calves, at six hours post partum, were generally very strong and vigorous. Originally, it was intended to initially allow calves to suckle at three hours post partum, but this was found in practise to be impossible with non-mothered calves. After unsuccessful attempts on three different calves, two of which had not even stood by three hours post partum, it was decided to allow calves to first suckle at six hours post partum. One calf was excluded from the experiment, because, due to extremely "poor" dam shape (see Materials and Methods, Part I), it was not possible to get it to suckle. One other calf exhibited such a weak teat-seeking drive that it did not ingest measureable quantities of colostrum and it, too, was excluded.

(d) Muzzling of calves

This was carried out using leather dog muzzles. In order to completely rule out the possibility of a calf suckling its dam through the end of the muzzle, a piece of rubber was sewn over the end (see Figure 7).



Fig. 7. A newborn Ayrshire calf with the modified dog muzzle in place.

(e) Other procedures

Blood sampling, preparation of serum, and the estimation of serum immune globulin concentrations were all carried out as described in Materials and Methods, Part I.

Section I

The Amounts of Colostrum Ingested by Suckling by Newborn Calves

It was found that generally large amounts of colostrum were consumed by calves put to their dams to suckle at six hours and again at 12 hours post partum, irrespective of whether they were mothered or not. The colostrum intakes of these calves were as follows:

TABLE 19

Colostrum Intakes in Ten, Non-mothered Calves Suckled at Six Hours and Twelve Hours Post Partum

<u>Calf</u>	<u>Birthweight</u> (lbs.)	<u>Intake</u> <u>at 6 hrs.</u> (lbs.)	<u>Intake</u> <u>at 12 hrs.</u> (lbs.)	<u>Intake</u> <u>(Total)</u> (lbs.)	<u>% of</u> <u>Birthweight</u>
D1	63.5	5.5	0.5	6.0	9.5
D2	81.25	4.75	1.0	5.75	7.1
D3	57.5	4.0	4.0	8.0	13.9
D4	73.0	4.75	4.5	9.25	12.7
D5	70.75	5.75	1.0	6.75	9.5
D6	100.25	4.5	2.25	6.75	6.7
D7	90.75	3.25	4.0	7.25	8.0
D8	89.75	6.75	1.75	8.5	9.5
D9	62.5	3.0	6.25	9.25	14.8
D10	100.0	5.5	1.25	6.75	6.8
Mean	75.8	4.8	2.7	7.4	9.9
S.D.	±14.4	±1.1	±1.8	±1.2	±2.8
S.E.	±4.6	±0.4	±0.6	±0.4	±0.9

It can be seen from the results expressed in Table 19 that although the total intakes of colostrum were high, there was nevertheless a wide variation in the amounts ingested by individual calves. When total

amounts of colostrum consumed by calves were expressed as a percentage of birthweight, a wide variation was still found. This variation is further discussed below (see Figure 8).

The amounts of colostrum consumed by a further ten calves left with their dams for 18 hours post partum and muzzled to prevent suckling except at six hours and twelve hours post partum when suckling was allowed are presented in Table 20.

TABLE 20

Colostrum Intakes in Ten Mothered Calves Suckled
at Six Hours and Twelve Hours Post Partum

<u>Calf</u>	<u>Birthweight</u> (lbs.)	<u>Intake</u> <u>at 6 hrs.</u> (lbs.)	<u>Intake</u> <u>at 12 hrs.</u> (lbs.)	<u>Intake</u> <u>(Total)</u> (lbs.)	<u>% of</u> <u>Birthweight</u>
A1	80.5	7.0	3.0	10.0	12.4
A2	78.0	7.5	3.0	10.5	13.5
A3	68.25	6.5	1.75	8.25	12.1
A4	84.0	7.0	3.5	10.5	12.5
A5	75.0	6.25	3.0	9.25	12.3
A6	79.0	3.25	3.5	6.75	8.5
A7	90.75	3.25	2.25	5.5	6.1
A8	58.25	5.5	3.0	8.5	14.6
A10	70.0	4.75	2.0	6.75	9.6
A11	42.5	4.0	3.0	7.0	16.5
Mean	72.6	5.5	2.8	7.6	11.8
S.D.	±13.6	±1.5	±0.6	±1.7	± 2.9
S.E.	± 4.3	±0.5	±0.2	±0.5	± 0.9

It can be seen from the above values that, in general, calves in both groups consumed approximately twice as much colostrum in their six hour meal than they did in their twelve hour meal. No significant

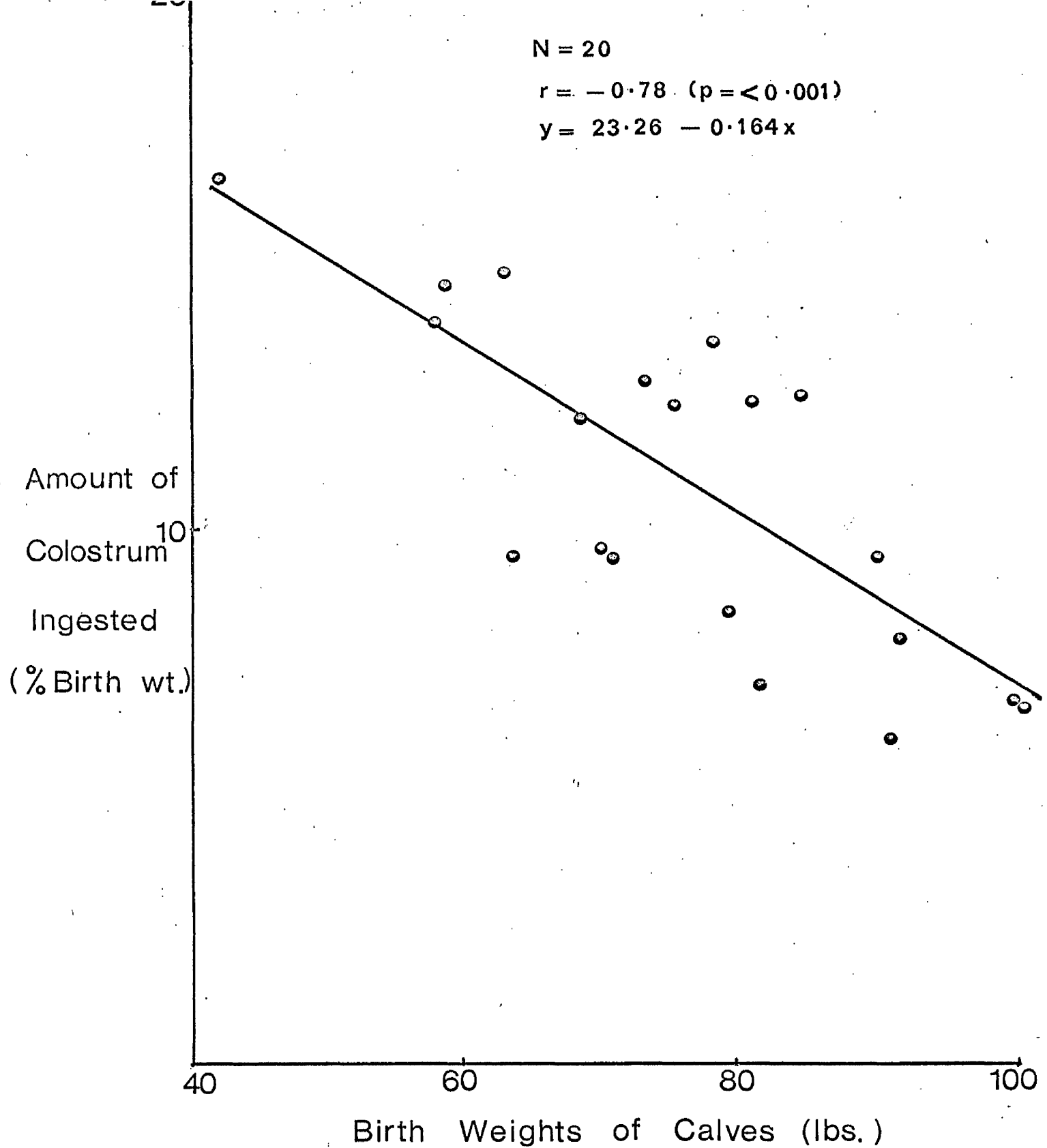


Fig. 8. The effect of birthweight on the total amounts of colostrum ingested (% birthweight) during suckling to satiation at six and twelve hours post partum by twenty newborn dairy calves.

differences were found between the two groups of calves in respect of birthweights, six-hour, twelve-hour or total intakes of colostrum. Neither was a significant difference demonstrated between the two groups when colostrum intake was expressed on a bodyweight basis. The mean total intake of colostrum for all 20 calves was $10.8 \pm 3.01\%$ of birthweight.

Although mothering did not significantly affect the quantities of colostrum ingested by calves, it was quite obvious when inducing the mothered calves to suckle, that they were much brighter, much better at standing and more eager to suckle than those in the non-mothered group. These latter calves were extremely dull by six hours post partum and often much difficulty was experienced in getting them in a suitable suckling position. Frequently, even when a teat was placed in such a calf's mouth, it only made a few suckling attempts and then stopped. Later, these calves brightened and showed more interest and stronger suckling drives.

A very wide variation did exist in the birthweight of the twenty calves in this series, although there was no significant difference between the birthweights of the two groups. The effect of birthweight on the total amount of colostrum consumed by calves was investigated (see Figure 8). It was found that the total amounts of colostrum consumed by each of the twenty calves in the series when expressed as a percentage of their birthweights bore a very close negative relationship ($p = <0.001$) with their birthweights).

Discussion

Initially, using calves that had been separated from their dams from 15 minutes post partum, attempts were made to encourage suckling at three hours post partum. In three attempts on three different calves, this was found not to be possible due to the extreme weakness and dullness of these calves (two of which had not stood by this time). Consequently, a regime was instituted whereby suckling was induced at six hours and again at twelve hours post partum, and calves were allowed to suckle their dams until they were apparently satiated. Considerably less trouble was experienced getting calves to suckle on the second occasion. The ten calves that were separated from their dams after 15 minutes licking were apparently recognised at six hours and twelve hours post partum by their dams and no trouble was experienced in getting the dams to accept their calves' teat-seeking advances. However, the dams were quicker to settle to these advances if allowed to lick their calf for a few moments prior to suckling.

The observation by Walker (1950) that two newborn calves ingested very large amounts of colostrum was borne out by the present study.

The mean total intakes of colostrum, the six-hour intakes and the twelve-hour intakes were not significantly higher in the mothered calves. In general, calves consumed twice as much colostrum at the six-hour feed than they did at twelve hours post partum. The mean total intake of colostrum (by suckling) for the first twelve hours

of life was 7.9 ± 1.3 lbs. (S.D.). This is considerably more than the one or two pints of colostrum usually offered to calves in the West of Scotland (Hector and Rowat, 1948; Selman, *et al.*, 1968).

The total intake of colostrum by all twenty calves during the first twelve hours of life was found to be $10.8 \pm 3.1\%$ of their birthweight. This is approximately twice the recommended daily rate for feeding whole milk to calves (British Veterinary Association Handbook, "The Diseases and Husbandry of Calves," and Morrison and Whitehair, 1962). A highly significant ($p = <0.001$) negative relationship was found between the total amount of colostrum ingested (% birthweight) and the birthweights of calves. The reason for this may be two-fold. First, large calves were generally found to have weaker suckling drives than small calves, and hence they may have just stopped suckling earlier than the smaller calves. Second, the limit on uptake might be purely an anatomical one due, for example, to abomasal size. In any event, it would seem from this finding that larger calves might be at an initial disadvantage due to their relatively lower intakes if the total amount of colostrum consumed has any bearing at all on subsequent serum immune globulin concentrations.

Section IIThe Serum Immune Globulin Concentrations of Dairy Calves
Suckling Their Dams at Fixed Times Post Partum

The 48-hour serum immune globulin concentrations of the ten non-mothered calves are presented in Table 21, together with their respective total colostrum intakes.

TABLE 21.

The Serum Immune Globulin Levels at 48 Hours Post Partum of Ten Non-Mothered Calves Which Suckled Their Dams at Six Hours and Twelve Hours Post Partum

<u>Calf</u>	<u>Total Colostrum Intake</u>		<u>48-Hour Serum Immune Globulin Concentration (Z.S.T. Units)</u>
	<u>Pounds</u>	<u>% Birthweight</u>	
D1	6.0	9.5	6
D2	5.75	7.1	8
D3	8.0	13.9	15
D4	9.25	12.7	20
D5	6.75	9.5	24
D6	6.75	6.7	13
D7	7.25	8.0	19
D8	8.5	9.5	19
D9	9.25	14.8	29
D10	6.75	6.8	30
Mean	7.4	9.9	18.3
S.D.	±1.2	±2.8	± 7.6
S.E.	±0.4	±0.9	± 2.4

Reference to the above table shows that a marked individual variation occurred in the 48 hours serum immune globulin concentrations of the ten calves in this group. Very low levels were found in two calves ingesting large amounts of colostrum by suckling. All calves,

save one, were found to have serum immune globulin concentrations of less than 30 Z.S.T. units. This is the level above which deaths were very rare in the survey of 415 calves carried out by McEwan (1968).

The 48-hour serum immune globulin concentrations of the ten mothered calves are presented in Table 22, together with their respective total colostrum intakes.

TABLE 22

The Serum Immune Globulin Levels at 48 Hours Post Partum of Ten Mothered Calves Which Suckled Their Dams at Six Hours and Twelve Hours Post Partum

<u>Calf</u>	<u>Total Colostrum Intake</u>		<u>Serum Immune Globulin Concentration (Z.S.T. Units)</u>
	<u>Pounds</u>	<u>% Birthweight</u>	
A1	10.0	12.4	39
A2	10.5	13.5	40
A3	8.25	12.1	19
A4	10.5	12.5	23
A5	9.25	12.3	36
A6	6.75	8.5	27
A7	5.5	6.1	36
A8	8.5	14.6	28
A10	6.75	9.6	31
A11	7.0	16.5	33
Mean	7.6	11.3	31.2
S.D.	±1.7	± 2.9	± 6.6
S.E.	±0.5	± 0.9	± 2.1

It can be seen from the above table that although the 48-hour serum immune globulin levels are considerably higher, there is still a wide individual variation. Nevertheless, the lowest value obtained

(19 Z.S.T. Units) is higher than the mean value of the previous group of non-mothered calves, and six calves have values in excess of 30 Z.S.T. units. The effect of total amount of colostrum consumed on the resulting serum immune globulin concentration was investigated in both groups of calves (see Figures 8, 9, 10 and 11). However, it was found that the amounts of colostrum that a calf ingested by suckling had no bearing on the final serum immune globulin concentration that it attained.

The mean 48-hour serum immune globulin concentrations of the mothered and the non-mothered calves were compared, and the results appear below in Table 23.

TABLE 23

A Comparison of the 48-Hour Serum Immune Globulin Levels
In Ten Non-Mothered and Ten Mothered Calves

<u>Calves</u>	<u>Mean Serum Immune Globulin Concentration (Z.S.T. Units)</u>	<u>S.D.</u>	<u>S.E.</u>	<u>Significance</u>
Non-mothered (10)	18.3	±7.6	±2.4	p = <0.001
Mothered (10)	31.2	±6.6	±2.1	

Hence, a highly significant difference ($p = <0.001$) was shown to exist between the 48-hour serum immune globulin levels of the two groups of calves, solely depending on whether they had been mothered or not.

In view of the finding made in Part I, Section III, that in the dairy calves the time of first suckling was apparently closely correlated with the subsequent 48-hour serum immune globulin

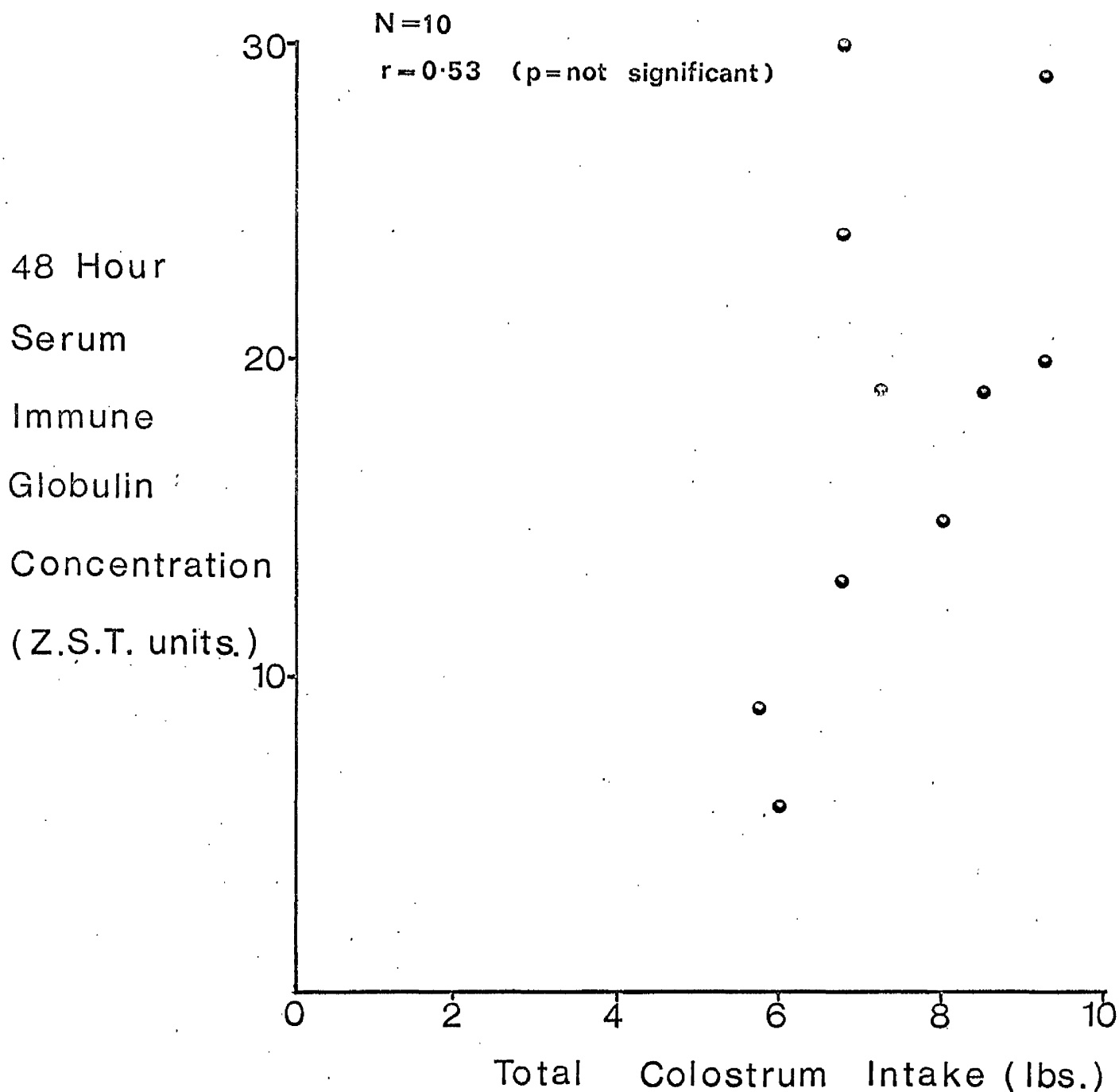


Fig. 9. The lack of correlation between the total colostrum ingested (lbs.) during suckling to satiation at six and twelve hours post partum by ten non-mothered dairy calves and their 48-hour serum immune globulin concentrations.

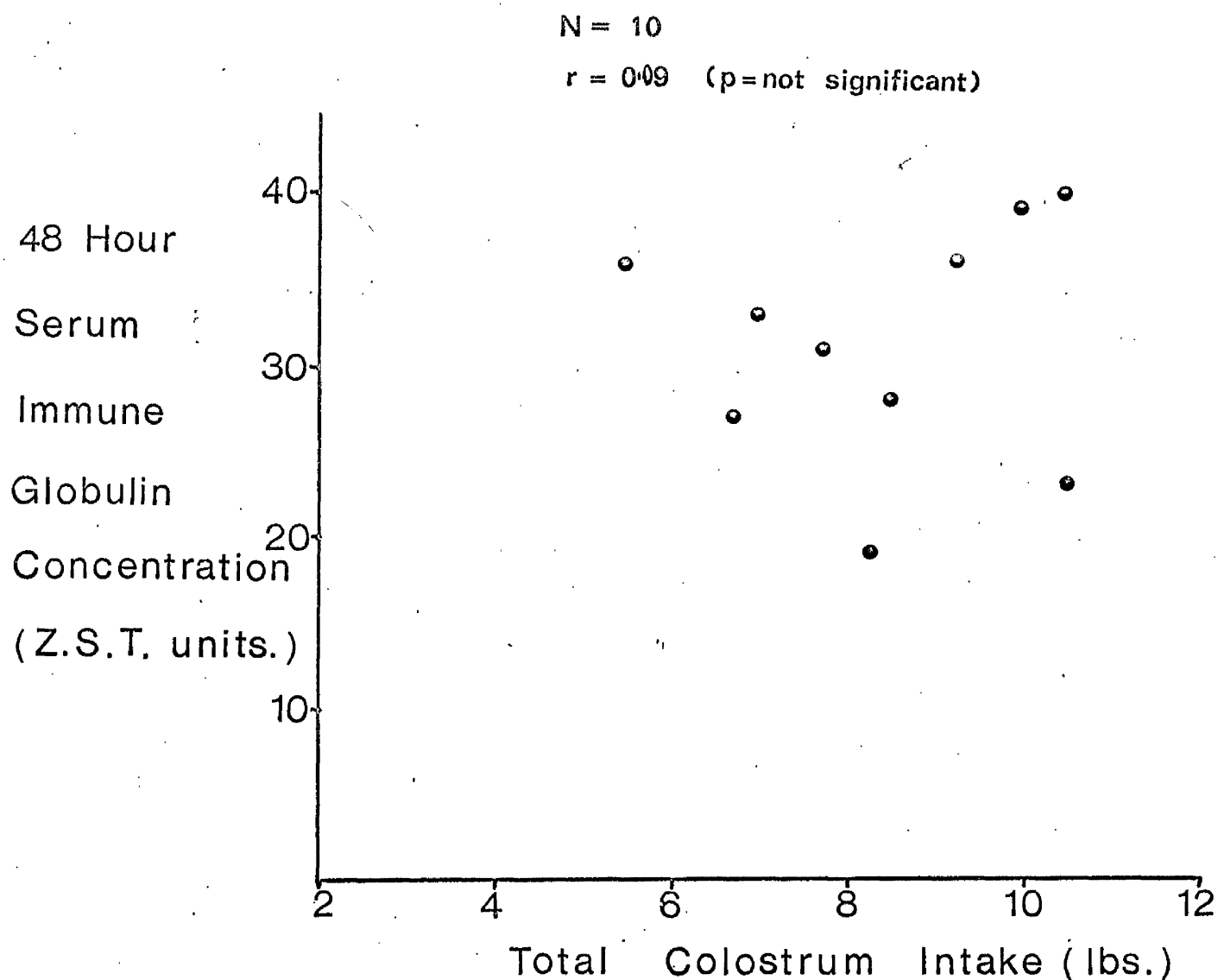


Fig. 10. The lack of correlation between the total colostrum ingested (lbs. during suckling to satiation at six and twelve hours post partum by ten mothered dairy calves and their 48-hour serum immune globulin concentrations.

N=10

$r = 0.25$ (p=not significant)

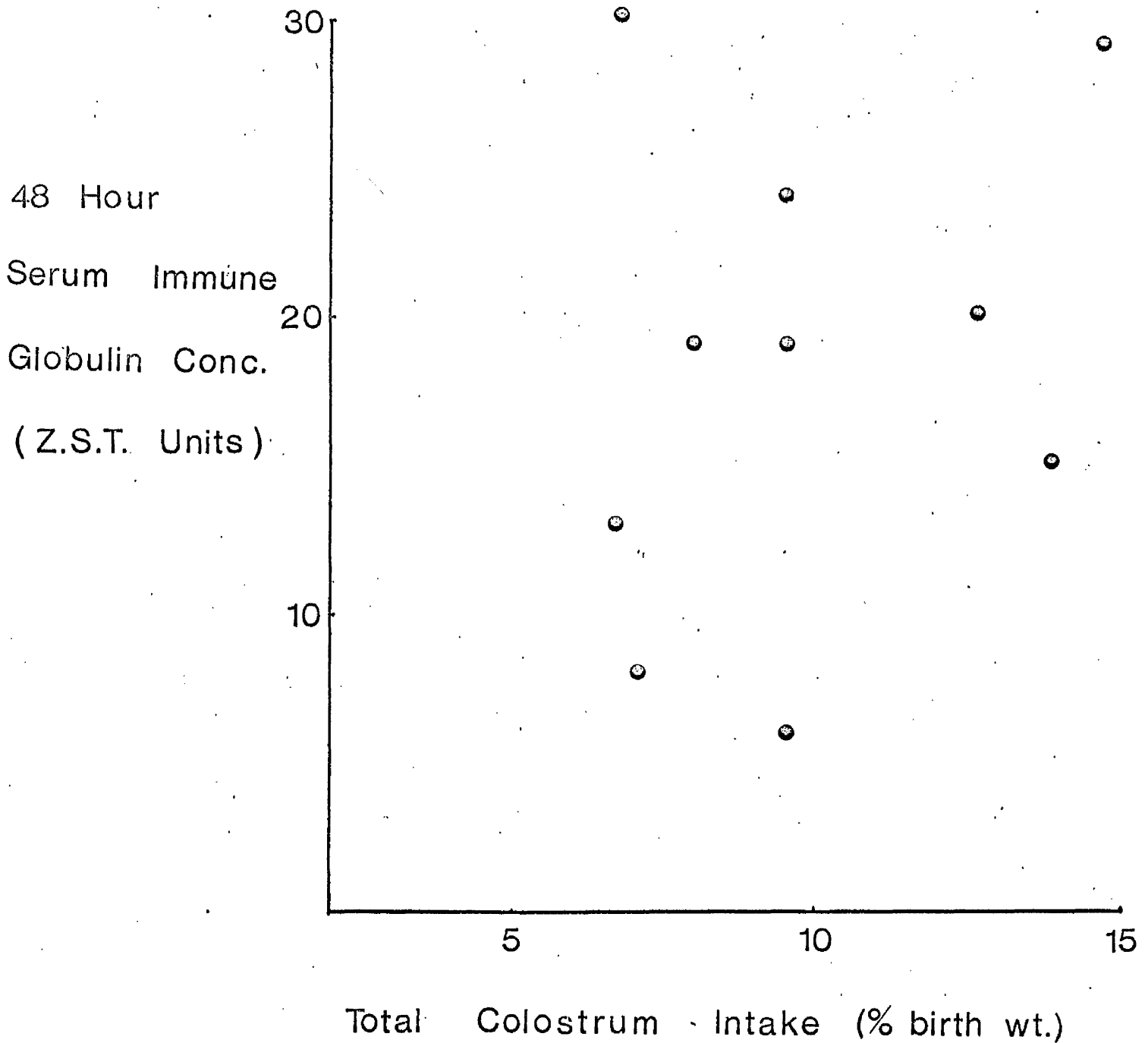


Fig. 11. The lack of correlation between the total colostrum ingested (% birthweight) during suckling to satiation at six and twelve hours post partum by ten non-mothered dairy calves and their 48-hour serum immune globulin concentrations.

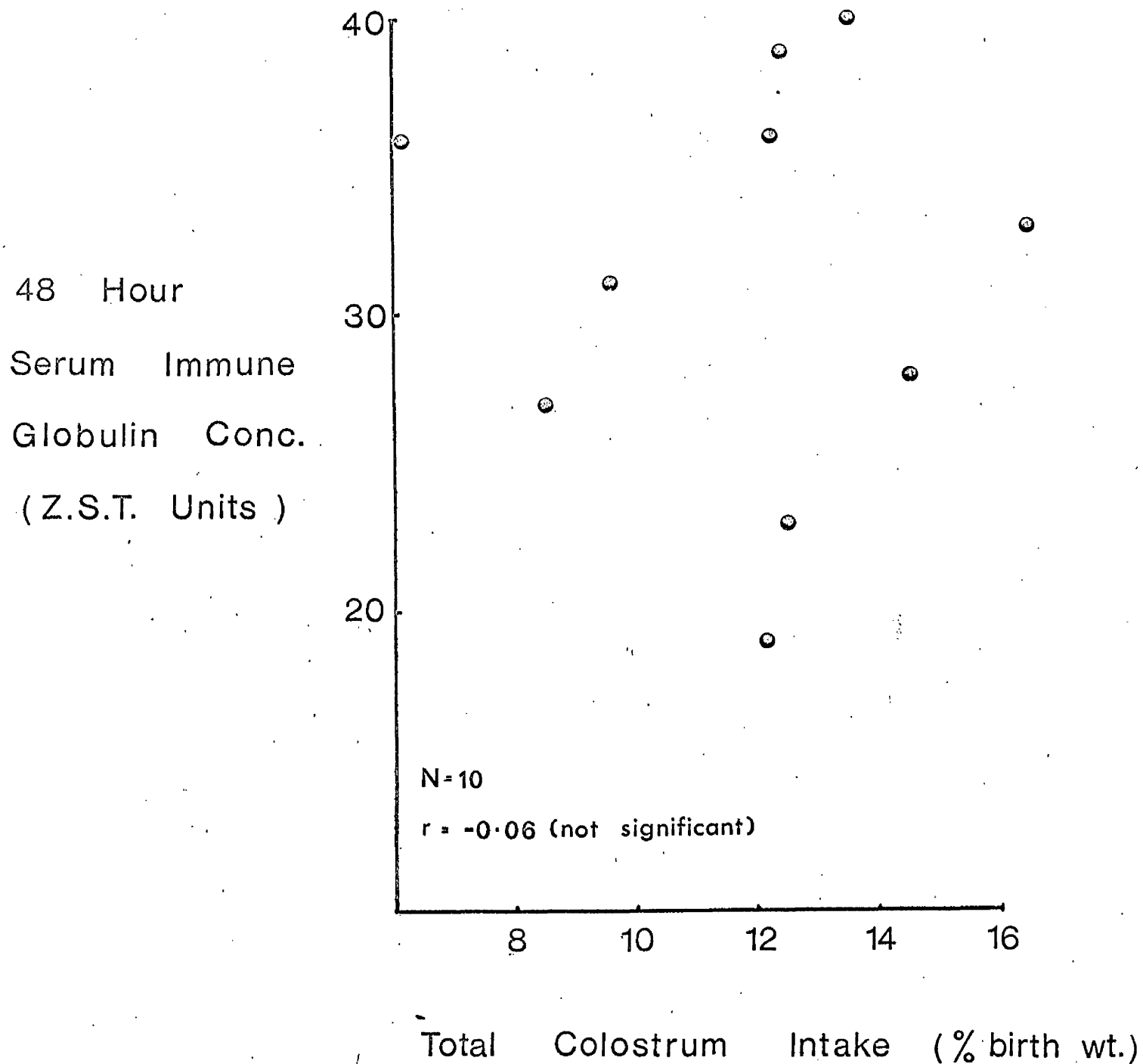


Fig. 12. The lack of correlation between the total colostrum ingested (% birthweight) during suckling to satiation at six and twelve hours post partum by ten mothered dairy calves and their 48-hour serum immune globulin concentrations.

concentration, a further group of ten mothered calves were allowed to suckle their dams to apparent satiation at six hours post partum only. In this series, note was also made of the time taken by calves to reach apparent satiation. The results are presented in Table 24.

TABLE 24

The 48-Hour Serum Immune Globulin Levels of Ten Mothered Calves Allowed to Suckle Their Dams at Six Hours Post Partum

<u>Calf</u>	<u>Birthweight</u>	<u>Total Colostrum Intake (lbs.)</u>	<u>Total Colostrum Intake (% Birth- weight)</u>	<u>Time Taken to Suckle to Satiation (minutes)</u>	<u>Serum Immune Globulin Concentration (Z.S.T. Units)</u>
A9	83.0	5.5	6.6	20	23
A12	81.25	6.75	8.3	18	24
A13	75.75	5	6.6	17.5	26
A14	66.25	3.25	7.9	29	32
A15	65.5	2.5	3.8	22	15
A16	66.25	5.0	7.6	13.5	26
A17	62.25	5.75	9.2	13	16
A19	59.25	4.0	12.7	26	30
A20	57.25	7.5	13.1	27	40
A21	64.0	6.75	10.6	11	17
Mean	68.1	5.4	8.6	19.7	24.9
S.D.	± 8.45	± 1.54	± 2.72	± 5.93	± 7.4
S.E.	± 2.67	± 0.49	± 0.86	± 1.88	± 2.3

Table 24 shows that it is possible to obtain reasonably high levels of serum immune globulin in calves suckled only once, at six hours post partum. The intakes of these calves were high, averaging $8.6 \pm 2.72\%$ of their birthweights. The length of time taken by these calves to reach apparent satiation was surprisingly long at 19.7 ± 5.93 minutes.

The mean serum immune globulin levels at 48 hours post partum in the above group of ten calves was then compared with the mean levels obtained by the ten mothered calves which were suckled twice, at six and again at twelve hours post partum. The results are presented below in Table 25.

TABLE 25

A Comparison of the 48-Hour Serum Immune Globulin Levels of Mothered Calves Suckled at Six Hours or at Six Hours and Twelve Hours Post Partum

<u>Calves</u>	<u>Mean</u>	<u>S.D.</u>	<u>S.E.</u>
Suckled at six hours post partum (10)	24.9	±7.4	±2.3
			n.s.
Suckled at six hours and twelve hours post partum (10)	31.2	±6.6	±2.1

No significant difference was found to exist between 48-hour serum immune globulin concentrations of the two groups of mothered calves suckled once, at six hours, or twice, at six hours and twelve hours, post partum. However, although in the ten calves which suckled only once, at six hours of age, the 48-hour serum immune globulin levels were reasonably high, only three were above 30 %S.T. Units compared with six calves above this level in the other group. The result, however, indicated that the effect of the extra suckling spell at twelve hours post partum was negligible. This supports the finding made in Part I, Section III, that the critical factor regarding the amount of immune lactoglobulin absorbed was the time of the first suckling.

Discussion

As seen in Part 1, a marked individual variation was obvious in the 48-hour serum immune globulin levels of the calves in this series irrespective of whether or not they were mothered. No correlation was found to exist between these levels and the total amount of colostrum ingested. In view of the work by McEwan (1968) showing that under certain conditions a linear correlation existed between the amounts of immune lactoglobulin presented to calves and the amounts subsequently absorbed, it seems highly likely that in presence of adequate total intakes of colostrum, the individual variation noted in the present study was largely due to variations in colostral immune lactoglobulin concentration. Data presented in Figure 6 suggested that where early suckling takes place, the concentration of immune lactoglobulin in colostrum is less important than the time of first suckling with regard to the final level of serum immune globulin.

It was previously shown (Part 2, Section 1) that although mothered calves were much brighter and easier to feed, their slightly higher intakes of colostrum were not significantly different from those of the non-mothered group of calves. In the present study, however, the mothered calves were found to have absorbed significantly more immune lactoglobulin ($p = <0.001$) than the non-mothered calves in spite of the fact that both groups of calves were suckled to satiation at fixed times (i.e. six hours and 12 hours) post partum, and no significant difference was demonstrable in their intakes.

Although inferior absorption due to maternal deprivation has not been recorded in ungulates, Halliday (1959), working with young rats, showed that premature shutdown could be brought about by separating these animals from their dams for 48 hours and maintaining them in an incubator. Poor absorption of Salmonella pullorum antibody was also noted in non-mothered artificially reared, pups compared with the absorptions in pups left with their dams (Filkin and Gillette, 1966). Halliday (1959) was able to reproduce the phenomenon of premature shutdown in young rats by oral or parenteral dosing with corticosteroids as was Moog (1953) working with young rats. Halliday (1959) suggested that the stress of maternal separation initiated premature shutdown. Payne and Marsh (1962) have also claimed that a similar phenomenon exists in newborn pigs treated with corticosteroids and Gillette and Filkin (1966) found that corticosteroid treatment of the bitch in late pregnancy rendered the pups less able to absorb antibody.

Filkin and Gillette (1966) suggested that pups separated from their dams lacked the sensory gratification of nursing and suckling and were hence less efficient at absorbing antibody. Herscher, et al. (1963) and Blauvelt (1954) have claimed that failure of the ewe to institute maternal grooming of the newborn lamb may have adverse effects, and Barron (1954) stated that grooming by the dam probably raised the general neural excitability of the newborn. These latter workers, however, were simply quoting opinions unsupported by any definite evidence of adverse effects due to maternal deprivation.

The results in the present study, together with similar observations on species as dissimilar as rats, mice, pups and pigs, show quite conclusively that nursing has very important physical as well as psychological functions.

Reyniers and Ervin (1946) found that certain reflexes of the newborn rat had to be triggered off by the dam and that initial attempts to rear rats artificially from birth were generally unsuccessful due to urine and faecal retention by the neonates. Licking of the newborn rat's perineum by the dam stimulated urination and defaecation, and when this stimulation was applied digitally to separated young rats, these functions took place and the animals thrived. A parallel may exist here with the oesophageal groove mechanism in newborn ungulates and maternal grooming might render this reflex more efficient at an earlier stage. In work carried out on the oesophageal groove reflex in newborn lambs, Watson (1944) underlined the importance of natural suckling and behaviour patterns surrounding this act in regard to the early and efficient functions of this reflex. It may be that the higher absorptions seen in mothered dairy calves are a result of colostrum passing straight to the abomasum by way of an efficient oesophageal groove and that non-mothering renders calves more likely to retain some or all of their colostrum in the rumen. In this context, it is interesting to note that one of the two calves in a previous experiment (Part I, Section III) with a disappointingly low serum immune globulin levels (DC6, 11 Z.S.T. units) was a calf which, although it finally suckled, was rarely licked by its dam during the observation period.

It has already been pointed out that the dairy calf is unusual in that the early removal from its dam and subsequent bucket feeding often results in manifestations of suckling deprivation (intersuckling). The regime of winter management of most dairy calves in the West of Scotland generally makes it impossible for grooming or suckling to take place, and this apparently renders the calf less able to absorb immune lactoglobulin than its mothered and suckled counterpart. The present study shows that, in addition, suckled dairy calves usually ingest far more colostrum than they are usually given the opportunity to ingest from a bucket. Moreover, over 50% of bucket-fed calves in Scotland are not offered colostrum until after six hours post partum (Selman, et al., 1968).

The results of the work presented in both Part 1 and Part 2 of this thesis indicated that where newborn calves were allowed to suckle colostrum, a seasonal variation in mean serum immune globulin levels did not occur. To test this observation still further, the suckling of the ten non-mothered calves was carried out during the summer of 1967 and that of the ten mothered calves, during the winter, 1967-1968. The effect of this was to reverse the seasonal variation recorded by Gay, et al. (1965b), although the variation was not as marked as that observed by these workers due to the relatively higher serum immune globulin concentration of the winter-born calves.

Mothered calves which were allowed to suckle their dams to satiation once only, at six hours post partum, attained only slightly lower 48-hour serum immune globulin levels than mothered calves suckling at six hours and twelve hours post partum. This difference

was found not to be significant. These levels were, however, reasonably high and far higher than the mean levels normally encountered in winter (Gay, et. al., 1965b). Therefore, dairy cows and their calves probably require only to be left together for approximately six hours, and by this time (see Part I, Section III) about 70% of the calves will have suckled. Any calves that have apparently not suckled by this time may be helped to do so, and they will generally absorb adequate amounts of immune globulin. However, the long time taken (approximately 20 minutes) for calves to suckle to satiation should be noted.

A traditional bias against allowing dairy calves to initially suckle their dams exists in the West of Scotland (Selman, et al., 1968). The belief is that this renders the dam less willing to allow "let down" of milk to take place with machine milking, tends to produce uneven udders and also makes the calf more difficult to subsequently bucket feed. However, the traditional form of management during the winter in this area would appear to be an attempt to rear what are, in effect, colostrum-deprived calves. If a farmer has a problem of colibacillosis in calves, then the difficulties arising from a change to calving in a box and allowing calves to initially suckle their dams would seem to be a small price to pay for fewer deaths.

PART 3

PART 2STUDIES ON THE ABSORPTION OF IMMUNE LACTOGLOBULIN BY NEWBORN DAIRY CALVESIntroduction and Review of the Literature

Increasing interest has been paid over the last few years to the transmission of immunity from dam to offspring and the nature of this protection. Much of the significant work concerning the absorption of gamma globulin by young animals has been carried out in the small laboratory animals. Although the following section will deal primarily with globulin absorption in calves, mention will be made of the work in other species whenever it is felt to be relevant.

McGirr (1947) reviewed the literature concerning colostrum transmission of antibodies from dam to offspring in the common domestic and laboratory animals. He discussed the then accepted belief that "In utero" acquisition of immunity could only take place in those animals having haemochorial or endotheliochorial placentas (i.e. with few layers separating maternal and foetal blood). In animals where thicker placentas are found (i.e. syndesmochorial or epitheliochorial placentas), reliance had to be placed upon absorbing antibodies from colostrum.

Table 26 demonstrates the relationship between placental type and antibody transmission. However, the rat and the mouse are notable exceptions to the general rule. These animals both have haemochorial placentas, but nevertheless acquire antibodies almost exclusively from colostrum and milk.

TABLE 26 (from McBirr, 1947)

<u>Animal Species</u>	<u>Placental Type</u>	<u>Antibody Transmission</u>		<u>Period from Birth During Which Intestinal Absorption of Antibodies Occurs</u>
		<u>Placental</u>	<u>Colostrum</u>	
Man	Haemochorial	+++	±	First few days
Mouse	Haemochorial	±	+++	Probably at least 20 days
Rat	Haemochorial	±	+++	Probably at least 20 days
Guinea-pig	Haemochorial	+++	-	Probably at least 20 days
Rabbit	Haemochorial Haemoendothelial	+++	-	Probably at least 10 days
Dog	Endotheliochorial	±	+++	Probably 10-12 days
Sheep	Syndesmochorial	-	+++	Less than 4 days
Goat	Syndesmochorial	-	+++	Less than 4 days
Cow	Syndesmochorial	-	+++	24 hours
Pig	Epitheliochorial	-	+++	No data available
Horse	Epitheliochorial	-	+++	No data available

It was, however, finally shown by Brambell (1958) that in rabbits and guinea pigs "in utero" absorption of antibodies occurs across the yolk sac endoderm and not the placenta. Brambell (1958) later suggested that in those species where the yolk sac persists until term (e.g. rabbits and guinea pigs), antibody absorption is "in utero," whereas where the yolk sac is present for only a short time in foetal life (e.g. ruminants, pigs, horse, rats and mice), colostrum transmission of antibodies is the rule. In either case, it may be seen that antibody absorption occurs across endoderm, be it yolk sac or gut. In man and the primates, however, where maternal globulins are obtained entirely "in utero" this is by way of the placenta (Brambell, 1958).

When a newborn calf ingests colostrum, it is diverted from the oesophagus to the abomasum via the oesophageal groove. It is said that the oesophageal groove functions irrespective of whether the calf is suckled or bucket fed and irrespective of the position of the calf's head while feeding (Hafez, 1962), but experiments with neonatal lambs suggest that this may not be so in the first few hours of life (Wallace, 1949). Once the colostrum is in the abomasum, the action of renin forms a casein clot even though the pH of abomasal contents at this early stage is only slight acid (Pierce, 1962). The proteolytic enzymes of the abomasum are not active until the pH drops to about 3 and in lambs, this does not occur until 36 hours post partum (Mill, 1956). Casein is retained in the abomasum, and the colostrum whey proteins pass into the small intestine. It has been demonstrated in

the anaesthetised calf that antibody absorption occurs in the small intestine (Comline, Roberts and Titchen, 1951a). Clarke (1959) found that young rats and mice absorbed gamma globulin and colloid materials (e.g. polyvinylpyrrolidone) from the jejunum and ileum but not from the duodenum. Filkin and Gillette (1966) confirmed this while studying antibody absorption in newborn puppies with the aid of fluorescent labelled antibodies.

Using colostrum whey with a high titre to Brucella abortus, Comline, Roberts and Titchen (1951a) showed that insignificant amounts were taken up by the hepatic portal system. Absorption was into the lacteals and hence to the bloodstream via the thoracic duct. These antibodies reached the thoracic duct 60-120 minutes following the introduction into the duodenum (Comline, et al., 1951a). Graves (1963) and Koeckenboeck and Schoenaers (1964) found that antibodies first reached the bloodstream of calves about 60-120 minutes following a meal of colostrum. Halliday (1957b) showed that 30 minutes after oral administration, antibodies may be detected in the circulation of the twelve-day-old rat.

Histological studies have been carried out on the intestinal epithelial cells of newborn calves (Comline, Roberts and Titchen, 1951b; Comline, Pomeroy and Titchen, 1953; and El Nagah, 1966), pigs and kittens (Comline, et al., 1953), kids (Hill and Hardy, 1956), rats and mice (Clarke, 1959) and pigs (Payne and Marsh, 1962 and Karlsson, 1966). The whole subject of the cellular phase absorption of antibodies by intestinal epithelium was reviewed by Brambell, (1966).

From these various studies it would appear that ingested proteins and other foreign substances enter the intestinal cells by pinocytosis which occurs between the microvilli of the brush border. The proteins then pass through the cells in pinocytotic vacuoles (phagosomes or phagolysosomes). Shortly after suckling, vacuoles with contents of the same staining properties as the colostrum whey in the intestinal lumen may be seen massed at the apical part of the cell. Later most of these vacuoles occupy the basilar part of the cell and this seems to displace the nucleus towards the brush border. These cells apparently absorb all of the globulin they can contain before releasing it into the lymphatics. Empty pinocytotic vacuoles may be seen in the intestinal epithelial cells of foetal pigs during the last three weeks of gestation and in newborn animals that have not suckled. In fact, Payne and Marsh (1962) claimed to have recorded absorptions in premature pigs removed by caesarian section at 100 days of gestation. After "colostrum shutdown" had occurred, intra-cytoplasmic vacuoles are rarely seen. Karlsson (1966) claimed that pinocytotic activity was present at birth but was greatly increased after the ingestion of colostrum. Clarke (1959) pointed out that fat, too, is ingested by pinocytosis so that this function is not lost by the intestinal cells when colostrum shutdown occurs. It is more likely that the cells become more selective as to what they ingest. Pinocytosis has also been seen to take place in the yolk sac endoderm of the foetal rabbit (Brambell, 1966).

Once globulin molecules are passed into the bloodstream by the thoracic duct, distribution occurs. Holborow (1967) stated that in the human, the IgG fraction of gamma globulin is equally distributed between intra- and extravascular compartments of the body, the IgM fraction, however, being exclusively intravascular. No figures for IgM distribution in newborn calves are as yet obtainable, but Nielsen and Nansen (1967) found a mean extravascular:intravascular partition ratio of 0.67 for IgG in healthy adult cattle.

Anker (1960) showed that a state of stabilisation between the intra- and extravascular globulin had almost completely occurred by 48 hours after radio-labeled (131) globulin was administered to a rabbit. Most workers have ignored the extravascular pool when studying the absorption of globulins. Clearly, any future quantitative approach on this subject must take this distribution into account.

The actual process of globulin absorption differs markedly between species. In the newborn rat or mouse, the cells of the gut are highly selective in what they transmit to the circulation. These cells transmit homologous gamma globulin far more readily than other serum proteins (Hemings, 1961). They can also distinguish between homologous and heterologous antibodies, transmitting the former more readily (Halliday, 1957b). Halliday (1957b) also showed that human, ox, guinea pig and rabbit sera interfered with the absorption of homologous antibodies. It was later shown by Brambell, Halliday and Morris (1958) that the gamma globulin fractions of human and bovine sera, but not the respective albumins, were responsible for this interference.

The Fc portion of the IgG molecule (Pierce, 1956) is responsible for both transmission and interference, and it has been postulated by Brambell (1966) that specific receptor sites for this portion exist on the walls of the pinocytotic vacuoles. Brambell (1958) pointed out that, clearly, selectivity was not based on molecular size because in young rats albumin (molecular weight 70,000) was less readily absorbed than gamma globulin (molecular weight 180,000). It has been shown by Halliday and Kekwick (1960) that the intestinal cells of the young rat select between antibodies produced in the same animal to different antigens. This difference is presumably due to the location of the antibodies in the serum proteins. Brambell, Halliday and Hemmings (1961) showed that when bovine gamma globulin containing agglutinins to Br. abortus is fed to young rats, the gamma globulin is transmitted, whereas the agglutinins are not. If this absorbed gamma globulin is then fed to other young rats, the absorption rate is increased. This shows that the gamma globulin must be somehow modified by the rat endothelium during its first transmission. Brambell (1966) makes the point that the cells of the intestine in the young rat and the yolk sac of the foetal rabbit show non-selective absorption, but very marked selectivity of what is transmitted to the young animal's circulation from the intestinal cells. Proteins which are not transmitted to the young rat's circulation undergo degradation in the intestinal epithelium.

The absorptive processes of the newborn pig do not, however, appear to be as selective as young rats, and all serum proteins and other foreign substances, such as polyvinylpyrrolidone and insulin,

may be absorbed readily (Lecce, Matrone and Morgan, 1961; Anon, 1962; and Asplund, Grummer and Phillips, 1962). Nordbring and Olson (1957) have presented some evidence suggesting that gamma globulin is absorbed from homologous serum more readily than beta-globulin or albumin.

The newborn calf's intestinal cells also show non-selectivity of absorption. Bangham, Ingram, Roy, Shillam and Terry (1958) using radioactively labelled serum and colostrum proteins showed that absorption of these substances occurred with equal facility. Pierce (1962) also showed that horse serum, gelatin and insulin could be absorbed by the gut of the newborn calf. It would appear that immune lactoglobulin is unchanged during absorption by the calf (Pierce, 1962).

Deutch and Smith (1957), Pierce (1959, 1960, 1961 and 1962) and Pierce and Johnson (1960) studied the proteinuria which occurs during the phase of colostrum absorption in newborn calves. This proteinuria was finally shown to be linked with the non-specificity of colostrum protein absorption in this species. Initially, Howe (1924) and Smith and Little (1924) found what they termed a transient albuminuria in newborn calves fed colostrum. The protein was later, however, shown by ultracentrifugation to be beta-lactoglobulin (Deutch and Smith, 1957; Pierce, 1960). Bangham, et al. (1958) noticed that labelled beta-lactoglobulin left the circulation far quicker than labelled gamma globulin. In fact, beta-lactoglobulin, and probably other low molecular weight whey proteins, which are absorbed non-selectively following a meal of colostrum, are cleared so rapidly by the kidneys that only traces can be found in the serum (Pierce, 1962).

Immuno-diffusion, but not electrophoresis, detects these low concentrations (Pierce, 1962). Other proteins (e.g. gelatin) with a similar molecular weight to beta-lactoglobulin (ca. 40,000) are also below the renal threshold and are cleared rapidly if fed to an absorbing calf or injected into an older animal. A transient proteinuria has been shown to occur in newborn lambs by McCarthy and McDougall (1949, 1953) and, more recently, Halliday (1965b) has produced evidence suggesting that lambs are, in fact, non-selective absorbers.

Famulener (1912) showed that anti-sheep erythrolysin were not absorbed from colostrum once kids were a "few days old." Later, Howe (1921) and Smith and Little (1922a) indicated that calves, too, lost their ability to absorb colostral globulins once they were two or three days old. Since that time, it has been shown by many workers, using many different techniques, that negligible absorption of whole protein molecules occurs in calves by 24-48 hours of age (Kerr and Robertson, 1944; Comline, Roberts and Titcher, 1951; Deutch and Smith, 1957; Smith and Erwin, 1959; and Pierce, 1961). However, Gay (1965) pointed out that although the period after birth during which absorption can occur is well known, little evidence was available regarding absorption rates during that time. One such study (Kaeckenbeeck, et al., 1961) indicated that maximal absorption occurred only for the first 12 hours post partum. The same workers claimed that by 16 hours post partum, the intestine had lost 50% of its absorbing function and that little or no absorption occurred after 36 hours. Cessation of antibody absorption occurred at approximately the same times in lambs (McCarthy and McDougall, 1949, 1953) and naturally reared pigs (Asplund, et al., 1962).

As yet, the basic reason for the cessation of immune lactoglobulin absorption is not known. Hill (1956) found that in the newborn lamb, parietal cells of the abomasal glands were sparse and the contents were neutral in reaction until about 48 hours post partum, and in the young rat the gastric glands were not fully developed until three weeks of age. They were, however, present at birth in the young guinea pig. This, he claimed, showed that conditions for proteolytic activity in the stomach were present at birth in the guinea pig which acquires its globulins in utero, and that the development of this gastric activity in the lamb and the rat coincided with the known times of cessation of globulin absorption in these animals. Laskowski and Laskowski (1951) had already shown that high levels of an anti-tryptic factor were present in cows' colostrum. Laskowski, Kassel and Hagerty (1957) later showed the same high levels of anti-tryptic factor in sows' colostrum. At this stage it appeared that cessation of globulin absorption by the intestinal cells occurred not because of changes in the intestinal cells themselves, but because of the initiation of proteolytic activity. In those animals where globulins are acquired post partum from colostrum, the globulins were protected against enzymic action in the stomach or abomasum by the neutral reaction of the contents, since pepsin is only formed from pepsinogen at pH3. Once in the intestine, protection against pancreatic proteolytic enzymes was guaranteed by the anti-tryptic factor in colostrum.

Deutch and Smith (1957) showed that no absorption of human gamma globulin occurred at 40 hours post partum in an unspecified number of colostrum-deprived calves fed a mixture of milk and

aluminium hydroxide gel and dosed orally with proanthine (anti-tryptic factor) every four hours from birth. Smith and Erwin (1959) failed to prolong absorption time in a calf by introducing colostrum into the cannulated duodenum surgically isolated from the abomasum. Chamberlain, Perry and Jones (1965) failed to demonstrate absorption of homologous gamma globulin by three-day-old pigs fed simultaneously with a trypsin inhibitor. Barrick, Matrone and Osborne (1954) found no evidence for increased absorption of porcine gamma globulin or bovine serum solids by newborn pigs using the same factor. However, Nordbring and Olsson (1957) did claim to have improved absorption of antibody by using an anti-tryptic factor prepared from bovine colostrum.

The general weight of opinion would seem to indicate from the above work that digestive degradation of colostral immunoglobulins does not play a major role in the cessation of intestinal absorption. Clark (1959) and El Nageh (1967) working with young rats and calves respectively, both showed that absorption of fluorescent labelled gamma globulin or polyvinylpyrrolidone was a function of the intestinal epithelial cells that the young of these species were born with. These cells were gradually replaced by new cells which were incapable of absorption.

Many workers have noted wide variations in the serum immune globulin concentrations of calves known or assumed to have ingested colostrum soon after birth. Fey and Margadant (1961) examined the sera of 46 calves known to have been fed colostrum and found that five (11%) were agammaglobulinaemic. Smith (1962) found that six calves from a total of 52 (12%) left with their dams for the first two days

of life, were also agammaglobulinaemic. Gay, et al. (1965a) found marked differences between serum globulin levels of 178, four to seven day old market calves in Scotland. In this survey, 53 calves (29.8%) were markedly hypo- or agammaglobulinaemic. However, as these calves were from local markets, no feeding history was available though it is probable that most received colostrum at least once during their first 24 hours of life. Smith, et al. (1967) also noted wide variations in the serum immune globulin levels of 190 home-bred and 40 market calves in England. Eight out of 80 (10%) calves said to have suckled within a few hours of birth by the farmers were found to have low serum globulin levels. Perry and Watson (1967a) noted wide variations in antibody levels in pigs born to sows vaccinated against Salmonella pullorum. This variation existed between litters and to a lesser extent between pigs of the same litter.

The marked seasonal variation in serum immune globulin levels that has been shown to occur in four to seven day old market calves in Scotland by Gay, et al. (1965b) has already been discussed. High mean values were found to occur in the summer months and low mean values in the winter, when calf mortality is at its highest in this region. In a similar survey in several parts of England, Smith, et al. (1967) claimed that there was no similar seasonal variation, and suggested that different calf management routines between the two countries might explain this difference. However, close examination of their data reveals that most of their high values occurred between June and November. A seasonal variation, therefore, does seem to exist in their calves although it is by no means as marked as that demonstrated by Gay, et al. (1965b).

Fey (1962) considered that agammaglobulinaemia was probably due to the early failure of the intestine to absorb globulins. Gay (1965) recorded the occurrence in an unspecified number of calves of an inability to absorb colostral globulins by 6-8 hours post partum. Unfortunately, he did not define the conditions of feeding or management of these calves. Neither Fey (1962) nor Gay (1965) suggested any reasons as to why early shutdown should occur in calves. No work appears to have been carried out in calves specifically to investigate this phenomenon. Where premature shutdown has been studied, it has been with laboratory animals and several interesting facts have emerged.

Moog (1953) working with mice, and Halliday (1959) working with rats, both showed that antibody absorption from the gut continued for 13-18 days in the former and 18-21 days in the latter animals. Apart from this minor difference, the young of both species showed startling similarities. Moog (1953) and Halliday (1959) showed in both species that premature cessation of antibody absorption could be initiated by removing the young animals from their dams for 48 hours and maintaining them in an incubator. Moreover, early shutdown could be initiated by oral or parenteral corticosteroid treatment. Both workers found that cortisone acetate was more efficient than deoxycorticosterone acetate (D.C.A.) and, in the experience of Halliday (1959), the former drug was less likely to kill the young rats. In an electron microscope study of absorbing rats and mice, Clarke (1959) found that corticosteroids caused the columnar absorptive cells of the intestine to lose their pinocytotic vacuoles and dense

Inclusions in response to gamma globulin administration (i.e. these cells came to resemble those of older animals). He also showed that the effect of corticosteroids was most marked on the younger columnar absorptive cells of the intestine. Moog (1953), Clarke (1959) and Halliday (1959) all noted that it took approximately three days before the effect of parenteral corticosteroids on absorption was noticeable. Halliday (1959) pointed out that in his rats, although oral and parenteral corticosteroid treatment induced premature shutdown, nevertheless, the rate of absorption during that shortened absorption time was higher than in untreated rats. The possibility that premature loss of absorption might also be induced by stress on the mother rat during pregnancy was suggested by Halliday (1965). Gillette and Filkin (1966) showed that corticosteroid treatment of pregnant bitches in the last day or two of gestation, initiated premature colostrum shutdown in their pups, although corticosteroids administered directly to other pups seemed to have no effect on absorption. Morris and Steel (1964) found that corticosteroid administration had little or no effect on the absorption of antibody by unweaned hedgehogs--an animal which normally absorbs for at least 40 days.

It has been noted in young mice (Moog, 1953, and Clarke, 1959) and young rats (Clarke, 1959, and Halliday, 1959) that rising levels of the enzyme, alkaline phosphatase, in the duodenal epithelium coincide with the onset of colostrum shutdown. This happens when this shutdown occurs at the normal time in these animals or when premature cessation of absorption is initiated by maternal deprivation or corticosteroid therapy and suggests that a similar mechanism causes shutdown irrespective of whether it is premature or not. Gillette and Filkin

(1966) found lower antibody absorptions in newborn pups removed from their dams, than in naturally reared pups.

Treatment of newborn pigs starved in the first 48-72 hours of life with cortisone acetate resulted in premature colostral shutdown (Payne and Marsh, 1962). Deutch and Smith (1957) treated an unspecified number of calves with A.C.T.H. and cortisone and found that the period of globulin absorption could not be prolonged. However, the nature of their experimental procedure meant that early shutdown would not have been detected.

Prolonged malnutrition and bad weather during pregnancy and in the early neonatal period have been suggested as causes of high neonatal mortality in hill lambs (Halliday, 1965a) and beef calves (Reisinger, 1965). In view of the findings in rats and mice, Halliday (1965a) suggested that the stress on ewes resulting from such adverse conditions might impair a newborn lamb's ability to absorb globulins. This stress, he added, must have to be initiated "in utero," presumably because premature shutdown following cortisone therapy in rats takes about three days to appear. In a study of 472 dead hill lambs, Halliday (1968) found that 236 (50%) were agammaglobulinaemic. Most lambs dying aged less than one day had not apparently walked, but close examination of those 99 lambs dying at one or two days of age (i.e. during the time that absorption might occur) shows that 51 (19.4%) were agammaglobulinaemic. Of this latter number, 51 lambs, 34 (66.7%) had no colostrum in their alimentary tracts on post-mortem examination. On this evidence, Halliday (1968) suggested that early colostral shutdown due to stress

on the dam might account for the lack of serum globulin in the lambs. However, it is not known at what time these lambs first suckled or even if the ewes had appreciable amounts of colostrum in their udders following such adverse circumstances. It would seem probable that the cold, wet weather at lambing time noted by Halliday (1968) depressed the newborn lambs to such an extent that it delayed standing times and teat-seeking advances and hence the time taken to first suckling.

When colostrum is bucket fed to calves, the resulting serum immune globulin concentrations are generally lower than when calves are allowed to obtain colostrum by suckling or by suckling and bucket feeding together (Smith, et al., 1967). This observation is of especial interest when the work of Withers (1952c) is taken into account. In a large scale survey of farms in the United Kingdom involving 3774 heifer calves, he found that mortality rates were on average three times higher in herds where colostrum was bucket fed than in herds where the calves were allowed to suckle colostrum from their dams.

Kaeckenbeeck and Schoenaers (1964), working on the false assumption (see Part 2) that newborn calves while suckling obtain colostrum frequently and in small amounts, investigated the effect of repeated feeding on colostral globulin absorption. They found that three, 200 ml, doses of colostrum produced a final titre which was almost three times as high as that from a single 600 ml dose. In short, early frequent feeds of colostrum had an additive effect and the first meal did not appear to effect a block on further absorption from subsequent meals.

Several workers have investigated the effect of previous feeding on absorption of colostral globulins from subsequent meals and in pigs, colostrum feeding would seem to trigger off colostral shutdown. Newborn pigs, maintained from birth on water, or fasted, are able to absorb gamma globulin as late as 106 hours post partum and cessation of absorption normally occurs at approximately 12 hours following a meal on sows' colostrum or cows' milk (Payne and Marsh, 1962). These same authors also showed that the effect of a previous ingestion of colostrum was limited to those intestinal epithelial cells which had experienced contact with colostrum.

Experimental work suggests that this phenomenon does not occur in calves. Deutch and Smith (1957) and Pierce (1961) did not prolong the phase of globulin absorption in calves maintained from birth on a lactose/dextrose solution and glucose-saline respectively. Schoenaers and Kaeckenbeeck (1964) failed to demonstrate decreased antibody absorption following the administration of glucose water or proteins such as cows' milk or egg white. However, Graves (1963) claimed to have blocked the absorption of Foot-and-Mouth Disease neutralising antibodies in calves by prefeeding 1.4 litres of skimmed milk or 80 ml immune serum in 500 ml saline 30 minutes after birth. Three hours after feeding, both calves were allowed to suckle their dams and their titres were compared with a third calf which was allowed to suckle naturally at five hours post partum. Although it is quite clear that no significant absorption occurred in the pre-fed calves, their respective intakes of colostrum are not recorded, and it is possible that neither calf suckled as vigorously as the single control calf

after receiving fluid at 30 minutes post partum. Baglioni and Locatelli (1966) claimed that although the prefeeding of polyvinylpyrrolidone of molecular weight 50,000 or 200,000 did not markedly diminish absorption of colostral globulins, the prefeeding of polyvinylpyrrolidone of molecular weight 700,000 resulted in negligible globulin absorption.

That the uptake by naturally reared newborn pigs of standard doses of homologous Salmonella pullorum antibodies varied considerably was noted by Perry and Watson (1967a). The difference between litters was greater than the difference between individuals and this, they concluded, indicated a strong maternal influence on antibody absorption. The nature of this influence has not yet been elucidated.

Smith, et al. (1967) stated that there was no correlation between the immune lactoglobulin level of a dam's colostrum and its calf's subsequent serum immune globulin concentration. However, he was dealing with 190 calves from 48 different farms, and presumably the times of first feeding and the amounts of colostrum offered to these calves varied considerably. Under more stringent conditions of management, McEwan (1968) found a linear correlation between the amount of immune lactoglobulin presented to a calf and its subsequent serum immune globulin concentration when allowance was made for the variations in birthweight of individual calves.

Perry and Watson (1967b) found a wide variation in the colostral whey antibody titres between sows and between the different mammary glands of the same sow. This latter finding, they claimed, was the cause of the marked intra-litter variations in titres to Salmonella

pullorum. Graves (1963) showed that the colostral whey titres to Foot-and-Mouth Disease virus varied from quarter to quarter in the same heifer. However, this is less likely to be a problem in cattle than in pigs due to the different suckling behaviour seen in the two species (see Part I). Moreover, this variation was probably a function of the fact that uneven milk formation frequently occurs in parturient heifers. McDiarmid (1946) suggested that absorption of Br. abortus agglutinins by calves was greatest from high titre colostrum. However, Kerr and Robertson (1944) claimed that the absorption by calves of skin sensitising antibodies to Trichomonas foetus was less efficient from high titre colostrum. Perry and Watson (1967b) working with newborn pigs showed that whereas absorption from high titre colostrum was only 2%, it was 13% for low titre colostrum. Although the latter workers ignored the intravascular pool of the IgG component of absorbed immune lactoglobulin, their findings are interesting from a comparative standpoint. They postulated that the cause of the inferior absorption from high titre colostrum was "pinocytosis fatigue" and pointed out that this phenomenon has been shown to occur in artificially fed Anoeba proteus.

The absorption of radio-ionated bovine gamma globulin by the small intestine of the anaesthetised calf is extremely slow when it is administered in aqueous solvents of similar ionic constitution to colostrum (Balfour and Comline, 1962; Hardy, 1968). However, the rate of absorption could be markedly increased by the addition of colostrum or certain of its constituents. Balfour and Comline (1962) showed that colostral and milk whey contained a small molecular weight protein

which accelerated absorption if mixed with inorganic phosphate and glucose-6-phosphate. Hardy (1968) using similar techniques to Balfour and Comline (1962) showed that sodium and potassium lactate slightly accelerated globulin absorption whereas citrate did not. He also showed that slight acceleration of absorption occurred on addition of the sodium and potassium salts of formic, acetic, propionic and isovaleric acids and that potassium butyrate had a very marked effect on absorption rates. As yet, no explanation for these findings has been made although these factors may be important in the increasing pinocytotic activity seen after colostrum ingestion by Karrlsen (1966). It is not known whether these factors are of any importance under more natural conditions of colostrum ingestion.

The presence in porcine and bovine colostrum of a trypsin inhibitor has already been discussed. However, in all save one experiment (Nordbring and Olsen, 1957) attempts to increase absorption rates or absorption times using such a factor have not met with success. Barrick, *et al.* (1954) suggested some factor or factors in sows' colostrum apart from anti-tryptic factor, favour immune lactoglobulin absorption. Attempts at blocking the effect of the pancreatic enzyme, deoxyribosenucleatase, with an extract of pigeon crops, were unsuccessful in prolonging the absorption time in calves (Deutch and Smith, 1957).

The possibility that hormonal constituents in colostrum bring about colostral shutdown has been investigated by some workers. The female rat comes into oestrous at 21 days post partum and this coincides with shutdown in the suckling rats. Halliday (1956) in cross-fostering experiments, showed that this oestrous had no bearing

on the cessation of absorption in the young. The same worker (Halliday, 1959) later showed that progesterone, testosterone, stilboestral and aldosterone were without effect on colostrum absorption in suckling rats. Deutch and Smith (1957) found that neither stilboestral nor progesterone, alone or in combination, increased the absorption time in calves.

Yet another aspect of the maternal influence on colostrum globulin absorption was investigated by Deutch and Smith (1957). These workers gave three 250 ml transfusions of maternal blood to newborn calves and found that gamma globulins subsequently fed at 40 hours post partum were not absorbed. This is not surprising as the more recent literature (see above) and the work presented herein, show that very little occurs in the calf after twelve hours post partum. Payne and Marsh (1962) gave porcine gamma globulin to newborn pigs by injection prior to colostrum feeding and failed to demonstrate any effect on the absorption of immune lactoglobulin. Working on the assumption that a permeability factor existed in amniotic liquor, Deutch and Smith (1957) investigated the effect of dosing this fluid in milk to calves, but again found that subsequent administration of gamma globulin at 40 hours post partum did not result in absorption. Once again, in the light of more recent knowledge, it is doubtful, even if a factor did exist in amniotic liquor, whether test feeding at 40 hours post partum would result in any significant globulin absorption.

Finally, Halliday (1966) showed that a breed variation in absorption efficiency seemed to exist in sheep under similar conditions of management. Finnish Landrace lambs, despite their relative large

litter sizes, had significantly higher total serum protein values than either Marino-cross-Cheviot or Blackface lambs.

In summing up, it would seem fair to say that first of all nothing is as yet known about the absorptive mechanism of newborn calves. It is evident that wide differences exist between the mechanisms of the different species and therefore care must be exercised in translating experimental findings from one species to another. The reasons why colostrum shutdown occurs are also not yet known, but it would seem from most of the work carried out, that this shutdown is a function of the neonate, not its dam's colostrum. The colostrum factors outlined above which definitely accelerate globulin absorption must, of course, be considered, but there is not yet any indication that under natural conditions they are of anything other than minor importance.

In some species corticosteroids and the stress of separation from the dam bring about early colostrum shutdown. However, it is not definitely known whether the phenomenon of early shutdown occurs in ruminants. Colostrum feeding methods influence absorption in calves. When bucket feeding is carried out, the generally lower serum immune globulin levels obtained may, in part, be a manifestation of the stress of maternal deprivation. Results obtained from experiments carried out in Part 2 of this thesis show that even when non-mothered dairy calves suckle their dams, maternal deprivation appears to cut down immune lactoglobulin absorption.

The very wide variations in serum immune globulin levels of colostrum-fed calves even under experimental conditions do not necessarily imply fundamental differences in the absorptive rate of individual calves. In many experiments too few calves have been used, too little attention has been paid to the time and manner of colostrum feeding, or to the amount and immune lactoglobulin content of the colostrum used. Also, the differing sizes of the calves under test and their management have largely been ignored. Until experiments are designed to obviate these and other variables, little information regarding the quantitative aspects of globulin absorption in the calf will be forthcoming. The following experiments were an attempt to study some factors affecting immune lactoglobulin absorption in newborn calves maintained under uniform conditions.

Materials and Methods

1. Experimental animals

The cows and helpers used in the following experiments were all of the Ayrshire breed and were all obtained in late gestation from the same source as those dairy cows and helpers used in Parts 1 and 2 of this thesis.

The calves that were used were all pure-bred Ayrshires with exception of the ten Ayrshire-cross-Friesians used in Section I (a). No attempt was made to use only one sex of calf for these experiments. Once again, calves which were born with assistance were only used if that assistance did not exceed one man pulling on ropes attached to the calf's legs.

2. Care and maintenance of experimental animals

The dams, save those used in Section II (a) were returned to holding accommodation from the heated calving box between 15 and 30 minutes after parturition. They were later machine milked out as completely as was possible using a standard method of milking (Materials and Methods, Part 4). Milking was carried out when convenient, but was always after calving and was never delayed for more than five hours after calvings even in the case of those dams used in Section II (a). On some occasions, colostrum was deep frozen for use in subsequent colostrum pools, and a 500 ml sample was always taken for further examination (see Part 4). The actual amounts of colostrum obtainable by standard milking techniques was recorded in the 20 dams used in Section III.

All calves, save those used in Section I (b), were born in a heated calving pen and were left with their dams for a timed 15 minutes. They were then removed, weighed and bled and, all save those calves in Section II (a), placed in the individual calf pens. The calves used in Section II (a) were then muzzled (see Materials and Methods, Part 2) and returned to their dams in the calving pens. The calves used in Section I (b) were born in unheated boxes during cold weather. They were immediately removed from their dams and placed in unheated, unlittered pens with concrete floors.

In no experiment was a calf fed colostrum after nine hours post partum. After colostrum feeding, they were all later fed raw milk from the same source and fed at the same rate and times as formerly (see Materials and Methods, Part 1). Once again, a profuse diarrhoea lasting four to six hours occurred at about 20 hours post partum, and this frequently coincided with the first bucket feed of milk. However, as with the calves used in the suckling experiments (Parts 1 and 2), all were clinically healthy and non-diarrhoeic long before the blood sampling at 48 hours post partum.

While colostrum was being fed, all calves, save those in Section II (a) were housed separately in individual metal pens in a thermostatically heated room. These pens were bedded with straw and were raised approximately six inches from the floor to allow for good drainage through the wire mesh pen bottoms (see Figure 13). These pens were cleaned out, washed and disinfected whenever the chance arose (i.e. whenever both pens were simultaneously empty at a convenient time). After the calves had received colostrum, they were then removed



Fig. 13. The individual metal pens used to house newborn calves during the time that they were receiving colostrum.

to individual pens in an adjoining room, sharing the same atmosphere and heated in the same manner and to the same temperature as the first room.

The calves which were used in Section II (a) were not kept in individual pens during the phase of colostrum feeding, but were housed in specially constructed calving pens with their dams for the first 18 hours of life. These pens measured approximately 10 feet x 10 feet and were constructed in the room usually occupied by experimental calves after their colostrum feeding. The calving pens were heated in the same manner as the calf pens.

3. Experimental procedures

(a) The preparation of a colostrum pool

Three different colostrum pools were used for the following experiments. For the first two pools, samples of colostrum obtained from the first milking of non-premilked cows and helpers were obtained from the cattle dealer who supplied the experimental animals. In the third case, colostrum was obtained in large quantities from the animals which calved at the Veterinary Hospital throughout the experiment. In all cases, the colostrum was stored at -4°C until enough had been accumulated to make up a pool. After the colostrum had been thawed out, it was mixed up in milk churns and then, with frequent stirring, aliquots of two litres were put into thick, polythene bags. These bags were then sealed and placed into one of several deep freezers maintained at -4°C .

As already stated, three different pools were used during the following experiments, and at the start of each section of results, mention is made of the pool used in that section.

Random samples of colostrum were taken from each pool when samples were rethawed for calf feeding. The levels of total protein and immune lactoglobulin in each of these random samples appear in the following table (individual total protein and immune lactoglobulin concentrations are recorded in Appendix 3, Tables 1 to 3):

TABLE 27

The Total Protein and Immune Lactoglobulin Concentrations of Wheys Prepared from Random Samples of the Three Different Colostrum Pools

(a) Total Protein Concentration (gm/100 ml whey)

	<u>Pool 1</u> (10 Samples)	<u>Pool 2</u> (16 Samples)	<u>Pool 3</u> (12 Samples)
Mean	10.58	9.69	11.47
S.D.	± 0.34	± 0.44	± 0.60
S.E.	± 0.11	± 0.11	± 0.17

(b) Immune Lactoglobulin Concentration (gm/100 ml whey)

	<u>Pool 1</u> (10 Samples)	<u>Pool 2</u> (16 Samples)	<u>Pool 3</u> (12 Samples)
Mean	7.64	6.72	8.66
S.D.	± 0.34	± 0.30	± 0.60
S.E.	± 0.11	± 0.08	± 0.17

It can be seen from the above table that the total protein and immune lactoglobulin concentrations in the wheys prepared from the random samples of colostrum fed to calves are reasonably consistent within each pool. The higher values present in Pool 3 are probably because the colostrum in this pool was obtained from cows calving in the

Veterinary Hospital. It was, therefore, definitely known that in each case the secretion was from the first milking post partum.

In order to minimise variations in immune lactoglobulin content of the pooled colostrum due to the freezing and thawing which was necessary in making up the colostrum pool, it was decided to dispose of any thawed colostrum that remained after a calf had been fed and not to freeze it for further use.

Also, to avoid variations in the thawing rate of the different constituents of colostrum, it was decided that all of the colostrum to be used had to be thawed before the first meal. The colostrum which was to be used for subsequent feedings was stored in a cool place until it was time to feed it. To minimise the possibility of souring occurring after rethawing, colostrum was never removed from the deep freeze longer than two hours before it was used, even in Section when "delayed" colostrum feeding was investigated.

(b) Colostrum feeding

It was felt at the outset the attempting to get calves to consume quantities as large as those consumed by the suckling calves in Part 2 (i.e. up to 7.5 lbs. per feed) would frequently result in failure and hence waste of the pooled colostrum. However, the amount offered probably needed to be greater than that generally offered by farmers when bucket feeding colostrum (i.e. 1-2 lbs.). The finding of McEwan (1968) that there was a linear relationship between the weight of immune lactoglobulin offered to calves and the subsequent serum immune globulin levels when the wide variations in bodyweights of calves were allowed for, obviously indicated that a fixed volume

of colostrum should not be fed to each calf. It was therefore decided to feed colostrum at the rate of 25 ml/lb. birthweight. This regime was subsequently found to be very suitable and was adhered to for all of the following experiments.

It was shown in Part I, Section III, that apparently there was, despite variations in immune lactoglobulin concentrations and amounts of colostrum ingested, a significant relationship between time of first suckling and the subsequent serum immune globulin concentration. However, in spite of this finding, it was decided to split the feeding of the colostrum into three meals. This was decided for the following three reasons.

(1) The above results were obtained with suckling when maximal amounts of colostrum were ingested and during these experiments smaller quantities were to be fed artificially.

(2) Kaeckenbeeck and Schoenaers (1964) had shown that when very small amounts of colostrum (i.e. 200 ml) were fed three times, the resulting E. coli titre was three times that obtained when one feed of 600 ml was given to a calf.

(3) It was assumed that it would be easier to get a calf at one hour of age to consume, for example, 700 ml, than it would be to get the same calf to consume 2100 ml of colostrum.

Feedings, in all save Section IV, were therefore carried out at one, five and nine hours post partum. At each of those times, one-third of the estimated total amount of colostrum for a particular calf was offered. In the experiments reported in Section IV, the calculated amount of colostrum was fed to each calf as one single meal.

A Rose-Miller teat bucket (see Figure 14) was used for calf feeding rather than the more conventional pail. It was used because with a pail, it was found to be impossible to get a calf to ingest all of the colostrum offered and also because some spillage was almost inevitable. With a teat bucket, it was possible to feed calves without any wastage at all, and also it was much easier to get one-hour-old calves to suckle rather than drink from a pail.

After colostrum had been thawed and the accurate amount measured out in a measuring cylinder, it was heated in a water bath to 40°C and then immediately offered to the calf. Every effort was made to measure the amount of colostrum to the nearest millilitre and to feed at the exact time.

Considerable difficulty was initially experienced in getting some calves to feed and several calves had to be excluded from the series (see Section I (a)). However, in time, little or no trouble was experienced except with the occasional very dull calf. Calves were fed either standing or in sternal recumbency, with the chin supported by the author's left hand.

(c) Preparation of colostrum whey

Colostrum whey was prepared by adding 0.25 ml of commercial rennet to 25 ml of colostrum in a universal bottle. The mixture was then incubated at 37°C in a water bath until it was seen to be well clotted. Following this, centrifugation was carried out for 20 minutes at 3,000 r.p.m. and then the almost clear whey was removed carefully with a Pasteur pipette. Care was taken at this point not to remove any colostrum fat with the whey sample. Total protein estimation and

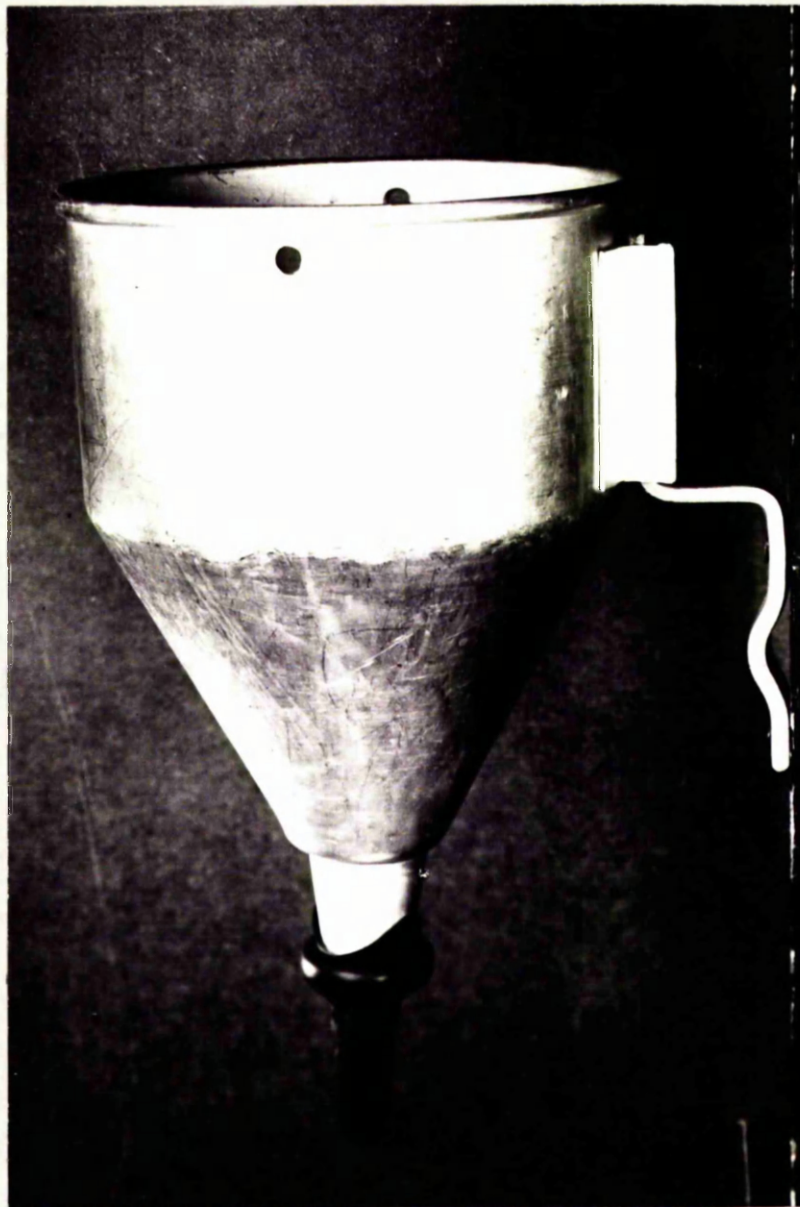


Fig. 14. A Rose-Miller teat bucket.

paper electrophoresis were then either carried out immediately or, if this was not convenient, the sample was stored at -4°C until several samples could be examined simultaneously.

Although total protein and immune lactoglobulin concentrations are frequently expressed in the following experiments as "grams/100 ml colostrum whey," it was felt that for the purposes of the studies in Part 4 of this thesis, the concentration of these constituents in colostrum might be a better form of expression. Consequently, experiments were carried out to find the relationship between colostrum whey and whole colostrum volumes. This work was carried out by Mr. D. P. Macdougall, B.V.M.S., M.R.C.V.S., of the Department of Veterinary Physiology, University of Glasgow, using ^{125}I labelled bovine fast IgG.

Colostrum samples were removed from nine different cows and to 100 ml aliquots of each, a known quantity of ^{125}I labelled bovine IgG was added. The radioactivity of this mixture was measured and then colostrum whey was prepared by the method described above. The radioactivity of the colostrum whey was then measured. The volume of whey present in each colostrum sample was found from the equation:

$$\text{Volume of whey (ml)} = 100 \times \frac{\text{(original volume of colostrum)}}{\text{Radioactivity of whey}} \times \frac{\text{Radioactivity of colostrum}}{\text{Radioactivity of whey}}$$

The following results were obtained for each of the nine samples:

TABLE 28

The Volumes of Whey Prepared from Nine Different Colostrum
Samples Following Treatment with Commercial Rennet

<u>Colostrum Sample</u>	<u>Volume of Colostrum (ml)</u>	<u>Volume of Whey In Colostrum (ml)</u>
Q12	100	89
Q13	"	85
Q16	"	86
Q17	"	89
Q18	"	78
Q20	"	87
Q21	"	79
Q23	"	84
Q25	"	80
Mean		84.1
S.D.		± 4.2
S.E.		± 1.4

These results indicate that when protein constituents need to be expressed as colostrum concentrations, the whey concentration should be multiplied by the conversion factor, 0.841.

(d) Ambient temperature

As already stated, the experimental area was heated thermostatically by three six kilowatt heaters. All calves, save those exposed to conditions of cold (see Section 1 (b)), were housed in this heated area.

Very careful note of the ambient temperatures was recorded while all of the calves in Sections 1 (a) and (b) were under test. Continuous temperature monitoring was carried out with a bimetallic thermograph (supplied by Baird and Tatlock (London) Ltd.) previously calibrated at 20°C in a water bath.

The lowest environmental temperature recorded while the calves in Section 1 (a) and the control calves of Section 1 (b) were under test was 16°C (60.8°F). However, this was an unusually low value and generally the ambient temperature stayed steady around 20°C (68°F). A full record of the maximum and minimum ambient temperatures for each of these calves appears in Appendix 3, Tables 4, 5, 6 and 7.

The management of those calves subjected to chilling has already been described in this section. No attempt was made to choose exceptionally cold weather for these calves. It was felt that if the seasonal variations in the serum immune globulin levels in market calves noted by Gay, et al. (1965b) could be explained more in terms of cold stress, then the temperatures usually encountered at any time between the months of November and April (inclusive) would show a significant result. There was a wide variation in the temperatures that these latter calves were subjected, but the range was from -10.5°C (13°F) to +5.6°C (42°F). Again, a full record of maximum and minimum ambient temperatures encountered by each of these calves appears in Appendix 3, Table 5.

When it became evident following the experiments reported in Section 1 (b) of this part, that chilling had no effect upon the immune lactoglobulin uptakes of chilled calves when compared to warm

control calves, temperature monitoring was discontinued. However, the experimental area continued to be heated as previously. The only apparent advantage of this was that the calves maintained in the warm environment appeared to be more comfortable, brighter and possibly easier to feed than the chilled calves.

(e) Weighing of calves

This was carried out on the same machine as that described in Materials and Methods, Part 2. However, these calves were all weighed at exactly 15 minutes post partum, and this was taken as their birth-weight. Even when calves were fed colostrum at five and nine hours post partum in Section IV, the amount of colostrum to be fed was calculated from the 15-minute weight (birthweight).

(f) Bleeding of calves

Blood sampling and serum preparation was carried out as described before in Part 1 of this thesis. However, a sample was also taken at 15 minutes post partum so that allowances could be made for the very slight (in most cases) turbidity seen when precolostral calf sera is subjected to the zinc sulphate turbidity test. This precolostral value was later subtracted from the 48-hour turbidity value so that a more accurate assessment of absorbed immune lactoglobulin could be made and the value so produced was termed the "absorbed immune lactoglobulin concentration."

(g) Estimation of the serum immune globulin concentration

This was measured by the zinc sulphate turbidity test described in Part 1.

(h) Estimation of colostrum whey total protein concentration

Colostrum whey was prepared by the method described above. The total protein concentration of this whey was then measured using the Biuret reaction (Weichselbaum, 1946).

(i) Estimation of colostrum whey immune lactoglobulin concentration

Colostrum whey was prepared by the method described above. Electrophoresis of the whey was then carried out using the technique described by Neil (1963). The resulting strips were evaluated automatically using a Chromoscan recording densitometer (Joyce Loebel and Company Ltd., Gateshead, England) again using the method described by Neil (1963). Two cellulose-acetate strips were completed for each colostrum whey sample. The mean for the two respective immune lactoglobulin fractions was used to determine the colostrum whey immune lactoglobulin concentrations from the previously determined colostrum whey total protein concentration.

(j) Corticosteroid preparation

The preparation used was a 1% solution of Dexamethasone trimethylacetate and prednisolone (Opticortone-S, CIBA Laboratories Ltd., Horsham, Sussex, England). Each calf treated with this compound was given 50 mgm intravenously and 50 mgm intramuscularly of the preparation at 15 minutes post partum.

Section 1

Attempts to Produce Consistent Immune Lactoglobulin Absorptions in Newborn Calves Using a Standardized Method of Colostrum Feeding and Management

The first two groups of calves were managed and fed by the standardized methods of management and feeding described in Materials and Methods, Part 3. Each calf was housed individually in a warm environment and fed pooled colostrum at the rate of 25 ml/lb. birth-weight. This total amount of colostrum was further divided into three equal meals and these were fed at one, five and nine hours post partum. Colostrum pool No. 1 was used in experiment 1 (a) and colostrum pool No. 2 was used in experiment 1 (b).

(a) The effect of breed of calf

It was decided to first compare the immune lactoglobulin absorptions of purebred Ayrshire and Ayrshire-cross-Friesian calves. This was because, due to the general trend towards using Friesian bulls in the West of Scotland, the latter type of calf was much more easily obtainable than purebred Ayrshire calves, at least with the type of cow dealt with by the supplier. It was felt that if no breed variation in immune lactoglobulin absorption could be demonstrated, there was little point in insisting on purebred Ayrshire calves for the subsequent experiments.

The results of this initial comparative study are presented in Table 29.

TABLE 29

The Absorption of Immune Lactoglobulin by Newborn Dairy CalvesThe Effect of Breed of Calf

<u>Ayrshire Calves (10)</u>		<u>Ayrshire-cross-Friesian Calves (10)</u>	
<u>Calf</u>	<u>48-Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>	<u>Calf</u>	<u>48-Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>
ST1	8	ST2	16
ST3	10	ST4	14
ST11	12	ST5	15
ST14	11	ST6	14
ST15	10	ST7	14
ST17	13	ST8	13
ST20	11	ST16	18
ST21	13	ST18	10
ST23	13	ST19	22
ST24	9	ST22	13
Mean	11.0		14.9
S.D.	± 1.7		± 3.1
S.E.	± 0.5		± 1.0

Significance: $p = < 0.01$

It can be seen from Table 29 that a significant difference ($p < 0.01$) was found to exist between the amounts of immune lactoglobulin absorbed under the conditions of the experiment. The purebred Ayrshire calves were found to have much more consistent 48-hour concentrations of absorbed immune lactoglobulin than the other group in spite of their poorer absorptions.

As the experiment proceeded, the impression was formed that the Ayrshire-cross-Friesian calves were larger and heavier at birth than the Ayrshires. This was investigated as it was felt that if a significant difference did exist between the birthweights of the two groups of calves, this might help to explain the higher 48-hour serum concentrations of absorbed immune lactoglobulin in the Ayrshire-cross-Friesian calves (i.e. they would have been given larger volumes of colostrum). The birthweights are presented in Table 30.

TABLE 30

The Birthweights of Ayrshire and Ayrshire-Cross-Friesian Calves

<u>Ayrshire Calves (10)</u>		<u>Ayrshire-cross-Friesian Calves (10)</u>	
<u>Calf</u>	<u>Birthweights (lbs.)</u>	<u>Calf</u>	<u>Birthweights (lbs.)</u>
ST1	59.0	ST2	63.25
ST3	70.25	ST4	65.75
ST11	75.5	ST5	80.5
ST14	75.5	ST6	70.5
ST15	86.5	ST7	75.25
ST17	78.25	ST8	71.0
ST20	88.5	ST16	72.25
ST21	64.5	ST18	71.25
ST23	65.76	ST19	76.5
ST24	65.0	ST22	78.75
Mean	72.9		73.3
S.D.	± 9.2		± 5.5
S.E.	± 2.9		± 1.7

Significance: n.s.

No significant difference could be demonstrated between the birthweights of the two groups of calves. It was, therefore, concluded

that the difference demonstrated in immune lactoglobulin absorptions in Table 29 was due to a breed difference in absorptive ability. It was consequently decided that further experiments would have to be restricted to Ayrshire calves only.

As more experience was gained in feeding very young calves, the number of "unfeedable" calves encountered became fewer. However, four calves in this first series were rejected because they were difficult to feed and failed to consume all of the colostrum offered to them. These four calves were all Ayrshire-cross-Friesians and, as Table 31 shows, three of the four were very large calves.

TABLE 31

The Birthweights of Four Calves Found to be Difficult Feeders

<u>Calf</u>	<u>Birthweight</u> (lbs.)
ST9	84.25
ST10	81.25
ST12	75.25
ST13	96.75

It was quite obvious when feeding the experimental calves that generally the best sucklers were those calves weighing less than the overall mean of 73.1 lbs.

(b) The effect of ambient temperature

Gay, et al. (1965b) found that a very marked seasonal variation existed in the serum immune globulin concentrations of Ayrshire bull calves obtained from markets in and around Glasgow. Although work already presented (see Parts 1 and 2) has indicated that the low mean serum concentrations of immune globulins in winter are basically associated with managerial variations, it was decided to investigate the effect of low ambient temperatures on immune lactoglobulin absorptions. A further ten Ayrshire calves were removed from their dams immediately after birth and placed in cold surroundings (a full account of their management is also described in Materials and Methods, Part 3). Apart from the fact that these latter calves were subjected to low ambient temperatures, their management was otherwise similar to the control calves. They were fed from the same pool of colostrum as the controls and at the same rates and times post partum.

The maximum and minimum temperatures encountered by the two groups of calves under test are recorded below in Table 32.

TABLE 32

<u>Control Calves (10)</u>		<u>Calves Subjected to Cold (10)</u>	
<u>Temperature (°C)</u>		<u>Temperature (°C)</u>	
<u>Max.</u>	<u>Min.</u>	<u>Max.</u>	<u>Min.</u>
21.2°	17.0°	5.5°	-9.0°

A full record of the maximum and minimum temperatures experienced by each calf is presented in Appendix 3, Tables 6 and 7. From the

figures presented in Table 32, however, it is quite clear that a wide division existed between the minimum temperature that the control calves were subjected to and the maximum temperature that occurred while the test calves were in unheated surroundings. It must also be remembered that the calves that were maintained in cold accommodation for 48 hours were initially placed there without first having been licked by their dams and consequently the evaporation of the amniotic fluid itself would be expected to give rise to considerable chilling. Moreover, all of these calves were kept on unlittered metal floors. In summary, it was felt that these conditions faithfully paralleled those that calves born in the byre in winter usually have to contend with in the West of Scotland. The results of this experiment are presented in Table 33.

TABLE 33

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves
The Effect of Ambient Temperature

<u>Control Calves (10)</u>		<u>Calves Subjected to Cold (10)</u>	
<u>Calf</u>	<u>48-Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>	<u>Calf</u>	<u>48-Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>
S1	10	C1	13
S2	11	C2	13
S3	14	C3	9
S4	12	C4	10
S5	7	C6	9
S6	8	C7	10
S7	12	C8	11
S8	11	C9	12
S9	6	C10	9
S12	12	C11	11
Mean	10.3		10.7
S.D.	± 2.4		± 1.5
S.E.	± 0.8		± 0.5

Significance: n.s.

It can be seen from the results presented in Table 33 that no significant difference was found to exist between the absorbed immune lactoglobulin of the two groups of calves. This was in spite of the fact that the calves maintained for 48 hours in a cool or cold environment certainly appeared to be less vigorous than the controls and were frequently more difficult to feed.

Section 11

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

The Effect of Mothering

The results obtained in Part 2, Section 11, indicated that high serum levels of immune globulins were uniformly present only when calves that suckled colostrum were left with their dams. When calves were separated from their dams between suckling spells, the 48-hour serum immune globulin concentrations were significantly lower even in the presence of similar large colostrum intakes. A similar phenomenon has been reported in rats by Halliday (1959) and in pups by Filkin and Gillette (1966).

From the results already obtained in Part 3, Section 11, it was quite obvious that disappointingly low levels of serum immune globulin were being obtained even when reasonably high colostrum intakes (i.e. up to two litres) occurred under what would generally be thought of as very good conditions for young dairy calves. It was therefore decided to investigate the effect of the dam's presence on ten calves maintained under the standard conditions already described. For the following experiments, colostrum pool No. 2 was used.

A full account of the management of the ten mothered calves is recorded in Materials and Methods, Part 3. In short, these calves were fed pooled colostrum at the same rates and times as the control calves, but were housed, not in individual pens, but in a much larger pen with their dams. These calves were muzzled to prevent suckling, and at the appropriate feeding times, these muzzles were removed, and

the calves were fed the appropriate amount of colostrum via the teat bucket. The amounts of immune lactoglobulin absorbed by the test and control calves are recorded below in Table 34.

TABLE 34

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

The Effect of Mothering

<u>Control Calves (10)</u>		<u>Mothered Calves (10)</u>	
<u>Calf</u>	<u>48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>	<u>Calf</u>	<u>48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>
S1	10	M1	16
S2	11	M3	25
S3	14	M4	20
S4	12	M6	19
S5	7	M7	16
S6	8	M8	14
S7	12	M9	14
S8	11	M10	18
S9	6	M11	19
S12	12	M12	16
Mean	10.3		17.7
S.D.	± 2.4		± 3.1
S.E.	± 0.8		± 1.0

Significance: $p = < 0.001$

It can be seen from the above table that a highly significant difference was found to exist between the amounts of immune globulin absorbed by the two groups of calves, even when the mothered calves did not suckle but were artificially fed. No significant difference

was found to exist in the mean birthweights of either group (see Appendix 3, Tables 6 and 8). This experiment therefore confirms the findings made in Part 2, but under far more exacting conditions. Both groups of calves in this experiment were bled at one, five and nine, twenty-four and forty-eight hours post partum, and the individual results are presented in Appendix 3, Tables 15 and 16. The rise in the mean serum concentrations of absorbed immune lactoglobulin in each group is shown in Figure 15. It can be seen that there would appear to be a higher globulin absorptive rate in the mothered calves than in the non-mothered controls. When the mean serum concentrations of absorbed immune lactoglobulin at the different times post partum were examined statistically, it was found that a significant difference existed between the concentrations at each sampling (see Figure 15) save the first.

It was once again noted when feeding the mothered calves with the teat bucket that they were considerably more alert and vigorous than the unmothered controls.

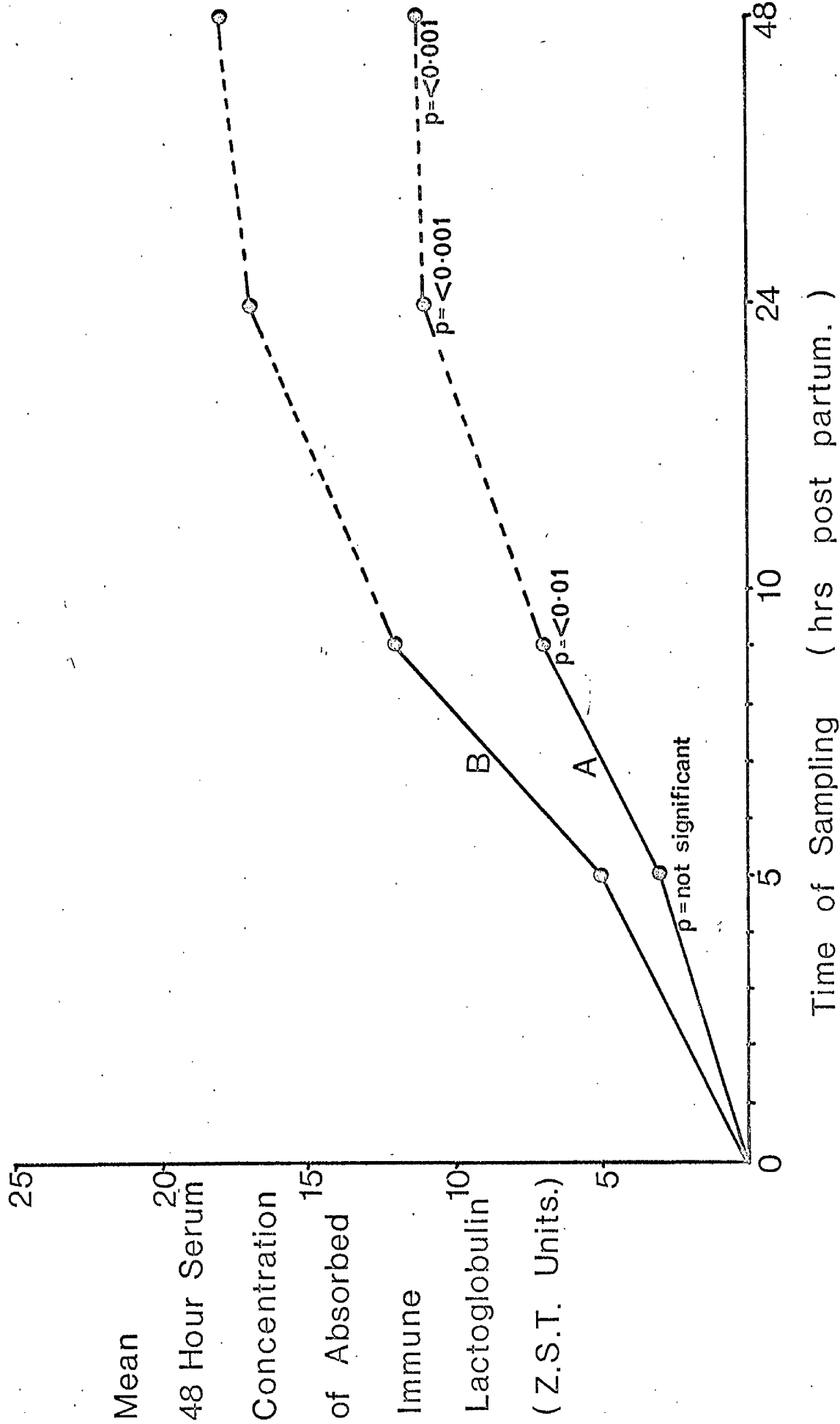


Fig. 15. The rise in serum concentrations of absorbed immune lactoglobulin in two groups of ten calves maintained under standard conditions of colostrum feeding and management. Line A joins the mean serum immune globulin concentrations of ten non-mothered calves at various times post partum. Line B joins the mean serum immune globulin concentrations of ten mothered calves at the same times post partum.

Section III

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

The Effect of Parenterally Administered Corticosteroids

Premature cessation of globulin absorption by the neonatal rat and mouse may be brought about by administration of corticosteroids (Moog, 1953; Halliday, 1959; Clark, 1959). It has therefore been decided to investigate this phenomenon in calves.

Ten newborn Ayrshire calves were therefore maintained under the standard conditions of feeding and management described in Materials and Methods, Part 3. However, at 15 minutes post partum, following weighing and blood sampling, the ten experimental calves each received a corticosteroid preparation (see Materials and Methods, Part 3) by both the intravenous and intramuscular routes. The results of this experiment are presented in Table 35.

TABLE 36The Absorption of Immune Lactoglobulin by Newborn Ayrshire CalvesThe Effect of Parenteral Corticosteroids

<u>Control Calves (10)</u>		<u>Corticosteroid Treated Calves (10)</u>	
<u>Calf</u>	<u>48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>	<u>Calf</u>	<u>48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>
S1	10	X1	15
S2	11	X2	12
S3	14	X3	12
S4	12	X4	15
S5	7	X5	16
S6	8	X6	16
S7	12	X7	16
S8	11	X8	17
S9	6	X9	12
S12	12	X10	17
Mean	10.3		14.8
S.D.	± 2.4		± 1.9
S.E.	± 0.8		± 0.6

Significance: $p = < 0.001$

Table 35 indicates that a significantly greater absorption of immune lactoglobulin occurred in those calves which received corticosteroid injections at 15 minutes post partum. The large doses that each calf received did not apparently exert any detrimental effects.

Section IVThe Absorption of Immune Lactoglobulin by Newborn Ayrshire CalvesThe Effect of Feeding Colostra of Differing Immune Lactoglobulin Concentrations

In the following experiment, a series of 20 Ayrshire calves were maintained under the standard conditions described in Materials and Methods, Part 3. The colostrum that was fed to these calves was obtained by machine milking their own dams as completely as possible between 15 and 30 minutes post partum. Samples were then removed from this colostrum after careful mixing for laboratory examination, and the colostrum was then fed to the calves, using a teat bucket, at the rate of 25 ml/lb. birthweight in three divided meals at one, three and five hours post partum.

Full records of this experiment appear in Appendix 3, Table 10, and Appendix 4, Table 1. Table 36 records the total protein and immune lactoglobulin concentrations of the wheys prepared from each individual colostrum sample, together with the 48-hour serum concentrations of absorbed immune lactoglobulin of the appropriate calves.

TABLE 36

The Absorption of Immune Lactoglobulin by Newborn Ayrshire CalvesThe Effect of Immune Lactoglobulin Concentration

<u>Calf</u>	<u>Whey Total Protein Concentration (gm/100 ml)</u>	<u>Whey Immune Lactoglobulin Concentration (gm/100 ml)</u>	<u>48-Hour Serum Concentration of Absorbed Immune Lactoglobulin (gm/100 ml)</u>
Q1	15.4	11.02	23
Q2	4.3	2.25	2
Q3	5.7	3.27	8
Q4	5.3	3.45	11
Q5	6.7	3.82	10
Q11	6.7	4.52	10
Q12	9.4	6.17	16
Q13	8.6	5.23	8
Q14	14.6	11.25	30
Q15	13.8	10.45	27
Q16	16.4	12.34	34
Q17	8.0	5.94	10
Q18	12.1	8.55	17
Q19	19.2	13.12	25
Q20	11.3	8.44	26
Q21	10.8	6.83	15
Q22	16.0	11.69	22
Q23	13.4	9.82	32
Q24	6.2	4.23	9
Q25	12.0	8.02	11
Mean	10.8	7.53	17.3
S.D.	± 4.2	± 3.33	± 9.1
S.E.	± 0.9	± 0.75	± 2.0

It can be seen from Table 36 that very wide variations were found to exist in the total protein and immune lactoglobulin concentrations

In the wheys prepared from each of the 20 samples of colostrum fed to the calves. The concentrations of these constituents in a large number of colostrum samples and the wide variations existing form the basis of a more detailed study on colostrum composition in Part 4 of this thesis. This will not be further discussed here, except in so far as the effect exerted upon the 48-hour serum concentrations of absorbed immune lactoglobulin in the 20 experimental calves.

The overall mean 48-hour serum concentration of absorbed immune lactoglobulin was found to be 17.3 ± 9.1 Z.S.T. units (S.D.). This is almost identical to the overall mean value of 18.3 ± 7.64 Z.S.T. (S.D.) units found in the ten non-mothered calves which were allowed to suckle their own dams at six hours and at twelve hours post partum.

A highly significant correlation ($r = 0.84$, $p = < 0.001$) was found to exist between the total protein concentration of whey prepared from samples of the different colostrums and the 48-hour serum concentration of absorbed immune lactoglobulin of calves fed these colostrums (see Figure 16). An even higher correlation ($r = 0.88$, $p = < 0.001$) was found when the colostrum whey concentrations of immune lactoglobulin were compared with each calf's 48-hour serum concentration of absorbed immune lactoglobulin (see Figure 17).

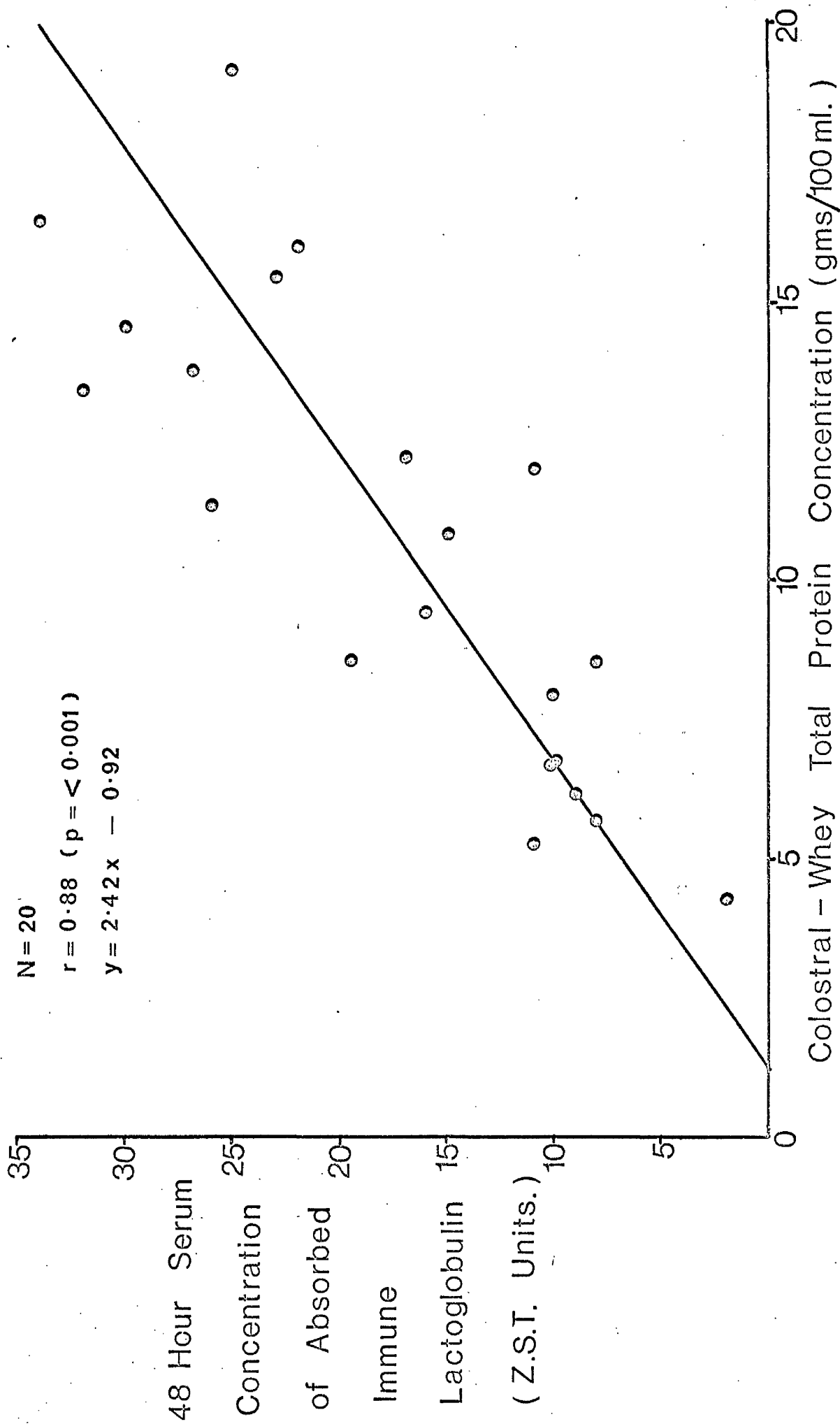


Fig. 16. The relationship between the colostrum whey total protein concentrations of twenty samples of colostrum and the 48-hour serum concentrations of absorbed immune lactoglobulin of twenty newborn calves fed these colostrum under standardised conditions of feeding and management.

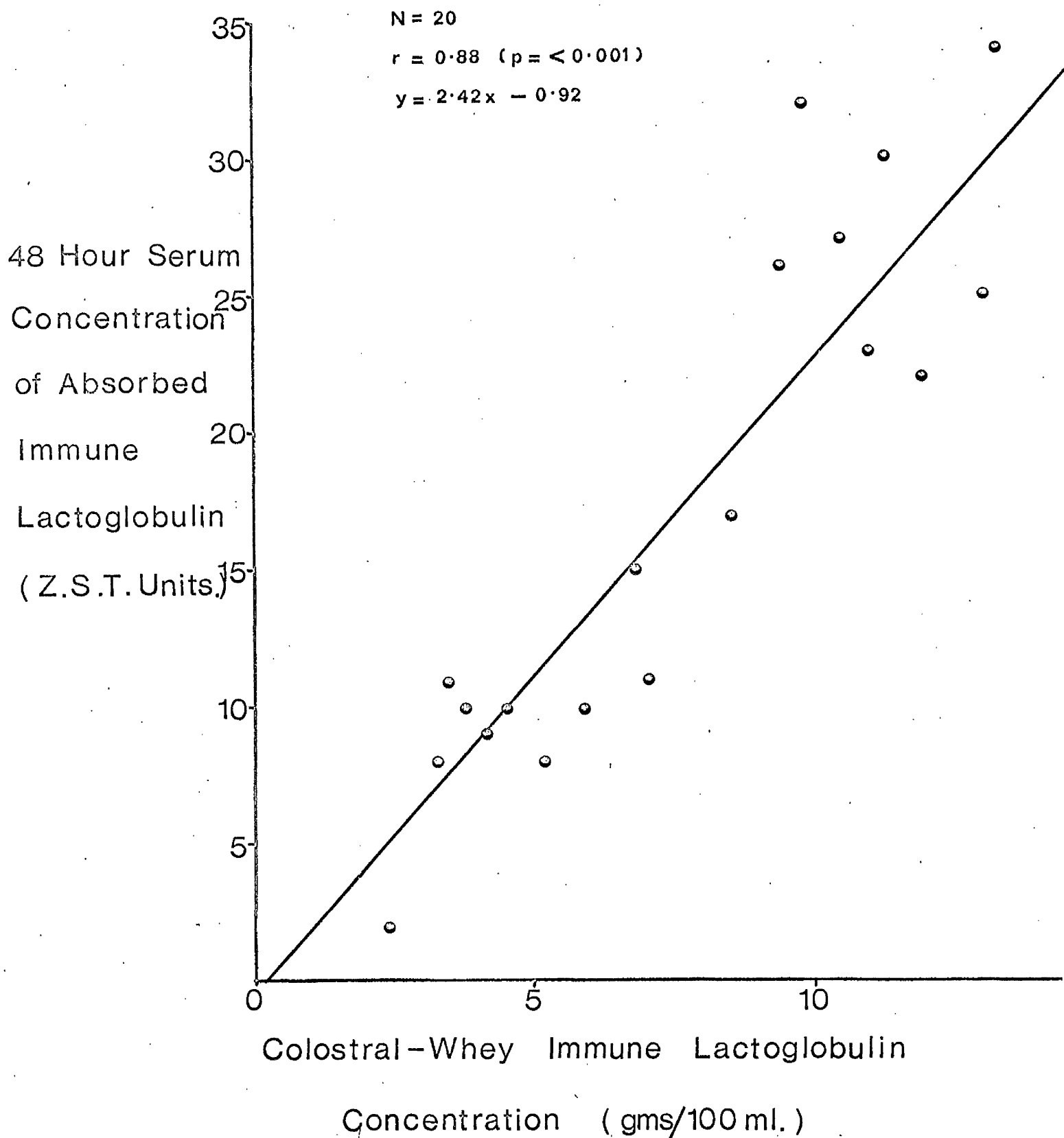


Fig. 17. The relationship between the colostral whey immune lactoglobulin concentrations of twenty samples of colostrum and the 48-hour serum concentrations of absorbed immune lactoglobulin in twenty newborn calves fed these colostrum under standardised conditions of feeding and management.

Section VThe Absorption of Immune Lactoglobulin by Newborn Ayrshire CalvesThe Effect of Feeding Colostrum at Varying Times after Birth

In the following experiment three groups of ten calves each were maintained under approximately the same conditions as described in Materials and Methods, Part 3, except that the times of first feeding were varied and that the amount of pooled colostrum to be fed was presented to each calf in one meal, not split into three separate meals as had been practised previously. All of the calves in this experiment were fed colostrum pool No. 3. The amount of pooled colostrum to be fed was calculated from the weight of each calf at 15 minutes post partum, even when colostrum feeding was delayed until nine hours post partum.

The birthweights, colostrum intakes and 48-hour serum concentrations of absorbed immune lactoglobulin are shown for the first group of calves in the following Table 37.

TABLE 37

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves
Calves Fed at One Hour Post Partum

<u>Calf</u>	<u>Birthweight</u> (lbs.)	<u>Colostrum</u> <u>Intake</u> (ml)	<u>48-Hour Serum</u> <u>Concentration of</u> <u>Absorbed Lactoglobulin</u> (Z.S.T. Units)
F2	63.5	1588	19
F6	45.0	1125	27
F8	71.75	1794	26
F10	67.5	1688	21
F13	81.5	2031	17
F18	77.5	1938	22
F21	76.75	1919	40
F27	72.25	1806	16
F29	70.0	1750	21
F33	64.25	1606	21
Mean	69.0	1725	23.0
S.D.	± 9.7	± 241.4	± 6.5
S.E.	± 3.1	± 76.4	± 2.1

Table 37 shows that it is possible, using a teet bucket, to successfully feed calves that are only one hour of age with large quantities of colostrum. These calves were consuming colostrum almost at the rate of 7% of their birthweights. It must be noted, however, that two calves had to be excluded from this group; one failed to ingest the whole amount offered and another was so incoordinated in its movements that some colostrum was spilled from its mouth. This experiment was carried out after the previous experiments had been concluded and consequently the author was far more experienced at feeding very young calves by this time.

Table 37 also clearly shows that the 48-hour serum concentrations of absorbed immune lactoglobulin of these ten calves, and hence presumably the absorptive efficiency, was considerably more variable than in previous experiments. In general, however, the serum concentrations of absorbed immune lactoglobulin are higher than those obtained from previous experiments.

The birthweights, colostrum intakes and 48-hour serum concentrations of absorbed immune lactoglobulin for the second group of calves were as follows.

TABLE 38

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

Calves Fed at Five Hours Post Partum

<u>Calf</u>	<u>Birthweight</u> (lbs.)	<u>Colostrum</u> <u>Intake</u> (ml)	<u>48-Hour Serum Concentration</u> <u>of Absorbed</u> <u>Immune Lactoglobulin</u> (Z.S.T. Units)
F1	83.75	2094	16
F4	84.5	2113	19
F5	71.5	1788	19
F7	106.0	2650	22
F12	70.25	1756	23
F19	63.25	1581	10
F20	50.75	1269	22
F25	74.5	1863	14
F26	74.0	1850	19
F28	66.5	1663	13
Mean	74.5	1863	17.7
S.D.	± 14.0	± 350.0	± 4.1
S.E.	± 4.4	± 110.7	± 1.3

It is evident from the data presented in Table 38 that the variation in 48-hour serum concentrations of absorbed immune lactoglobulin is not as marked as was the case with the first group of calves. No calves failed to consume their calculated amount of pooled colostrum when fed at five hours post partum.

The birthweights, colostrum intakes and 48-hour serum concentrations of immune lactoglobulin for the third group of calves are presented in Table 39.

TABLE 39

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

Calves Fed at Nine Hours Post Partum

<u>Calf</u>	<u>Birthweight</u> <u>(lbs.)</u>	<u>Colostrum</u> <u>Intake</u> <u>(ml)</u>	<u>48-Hour Serum Concentration</u> <u>of Absorbed</u> <u>Immune Lactoglobulin</u> <u>(Z.S.T. Units)</u>
F9	79.5	1988	14
F11	71.5	1798	11
F14	80.5	2013	12
F15	61.25	1530	14
F16	69.0	1725	13
F22	71.75	1794	10
F23	72.0	1800	13
F24	90.25	2256	8
F30	76.5	1913	14
F32	68.5	1713	16
Mean	74.0	1852	12.5
S.D.	± 7.6	± 185.5	± 2.2
S.E.	± 2.4	± 58.7	± 0.7

Reference to Table 39 shows that those calves fed at nine hours post partum still absorbed significant quantities of immune lactoglobulin and only one calf had a Zinc Sulphate Turbidity value of less than 10 units. As with those calves fed at five hours post partum, all of the calves in this group consumed their appropriate amounts of colostrum without a great deal of difficulty.

Far less variation was seen in the serum concentration of absorbed immune lactoglobulin in this group of calves than in the calves of the previous two groups.

A comparison of the mean 48-hour serum concentrations of absorbed immune lactoglobulin of each group of calves is presented below in Table 40.

TABLE 40

The Absorption of Immune Lactoglobulin of Newborn Ayrshire Calves

The Effect of Time of First Feeding

<u>Time of First Feeding (hours post partum)</u>	<u>No. of Calves</u>	<u>Mean 48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>	<u>S.D.</u>	<u>S.E.</u>	<u>Significance</u>
1	10	23.0	±6.5	±2.1	p = <0.05
5	10	17.7	±4.1	±1.3	
9	10	12.5	±2.2	±0.7	p = <0.01

A significant difference was found to exist between the 48-hour serum concentrations of absorbed immune lactoglobulin of the three groups of calves, thus inferring that colostral shutdown does not occur

at a specific time after birth but is an inexorable process initiated at the time of parturition. No significant difference was demonstrable between the mean birthweights of the calves in each of the three groups.

The individual 48-hour serum concentrations of absorbed immune lactoglobulin are presented graphically in Figure 18, and a simplified graph showing the mean values for each group of calves is also presented (Figure 19).

It can be seen from Figure 19 that a line joining the three points (i.e. the three mean 48-hour serum concentrations of absorbed immune lactoglobulin for each of the three groups of calves) is a straight line. This indicates that the rate of cessation of immune lactoglobulin absorption is constant at least over the time space from one to nine hours post partum. Extrapolation of this line shows that, given this constant rate of shutdown after nine hours post partum, no immune lactoglobulin absorption could occur at or after approximately $18\frac{1}{2}$ hours post partum. If this same line is extrapolated back, the point at which it intercepts the y-axis indicates the theoretically possible mean 48-hour serum concentration of absorbed immune lactoglobulin of a group of calves fed this same pool of colostrum at the same rate as the other calves, at the time of birth. This is clearly impossible, but would theoretically produce a mean value for the group of approximately 24 Z.S.T. units. Using this assumption, it is interesting to note that under the conditions of this experiment, by ten hours post partum, the mean absorptive efficiency of the calves is already less than 50% of what it was at birth.

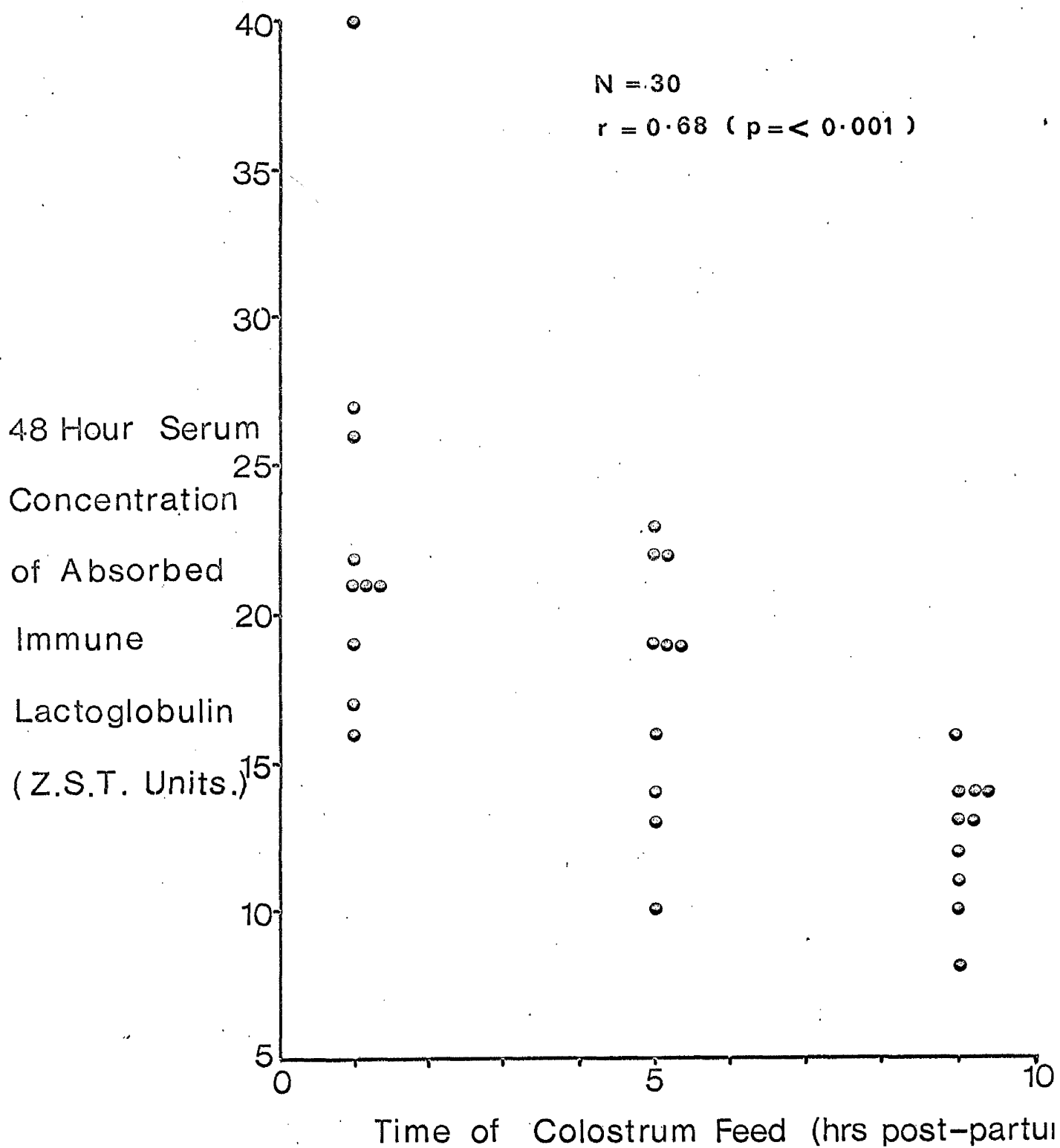


Fig. 18. The 48-hour serum concentrations of absorbed immune lactoglobulin in three groups of ten newborn calves fed pooled colostrum under standardised conditions of feeding and management at various times post partum.

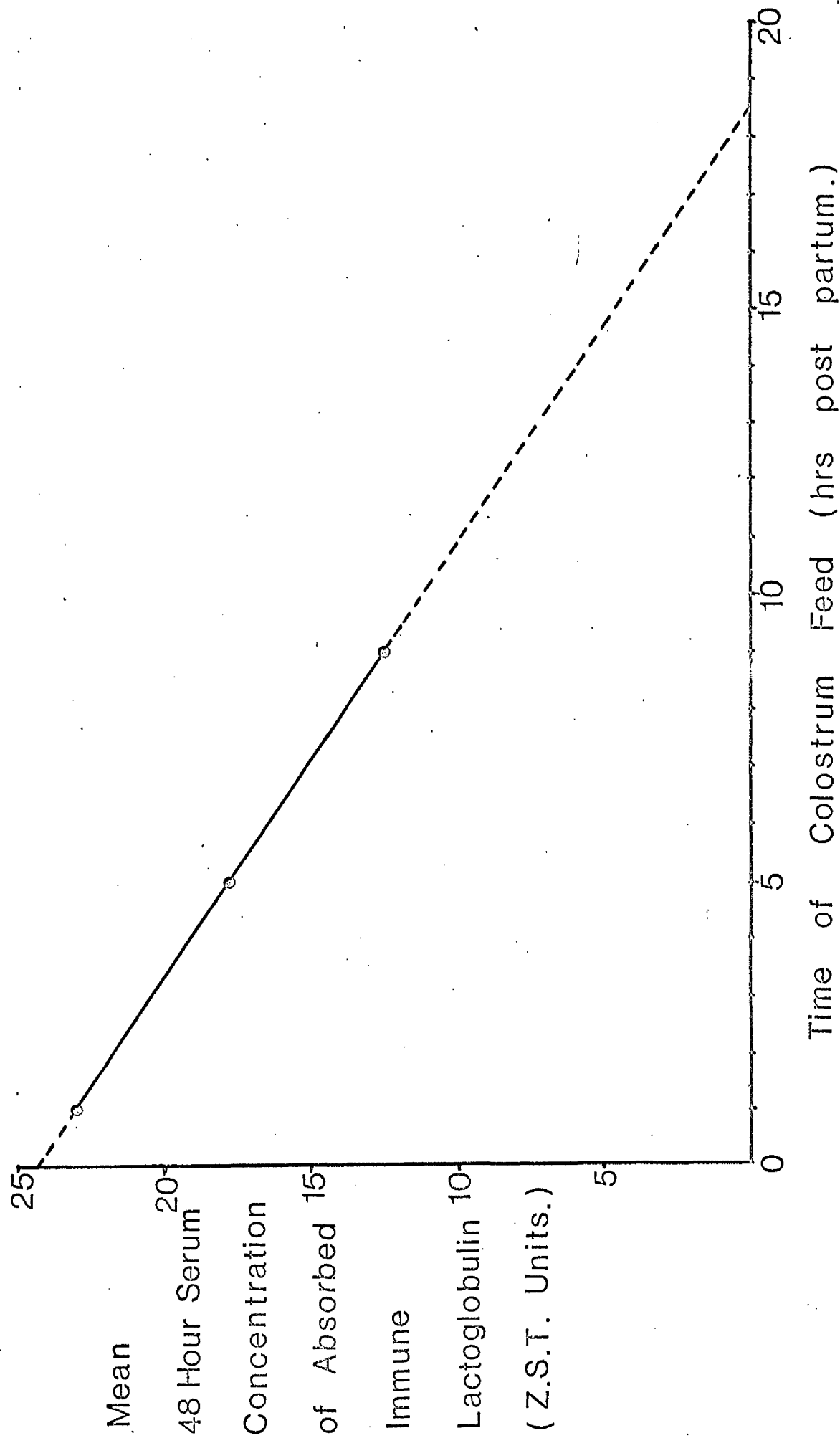


Fig. 19). The mean 48-hour serum concentrations of absorbed immune lactoglobulin of the three groups of newborn calves (see Figure 18) fed pooled colostrum under standardised conditions of feeding and management at various times post partum.

A very highly significant correlation ($r = 0.68$, $p = <0.001$) was found to exist between the time of first feeding of colostrum to calves and the serum concentrations of absorbed immune lactoglobulin at 48 hours post partum.

Section VI

The Absorption of Immune Lactoglobulin by Newborn Ayrshire Calves

The Effect of Divided Feeds of Colostrum

It was decided to investigate the effect of divided feeds of colostrum on the absorptive efficiency of newborn Ayrshire calves.

The calves in this experiment were fed colostrum pool No. 3 and were fed and managed under the standardised conditions of feeding and management described in Materials and Methods, Part 3. The results were then compared with those obtained in the previous experiment (Section V) for calves fed the calculated amount of pooled colostrum (i.e. 25 ml/lb. birthweight) in one feed and not in three divided meals as in this present study.

The results obtained are presented in Table 41.

TABLE 41

The 48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin in Ten Newborn Ayrshire Calves Fed and Managed Under Standard Conditions of Management

<u>Calf</u>	<u>48-Hour Serum Concentrations of Absorbed Immune Lactoglobulin (Z.S.T. Units)</u>
F34	20
F35	20
F36	19
F37	22
F38	15
F39	22
F40	18
F41	23
F42	16
F43	18
Mean	19.3
S.D.	± 2.5
S.E.	± 0.8

Table 41 demonstrates that the mean 48-hour serum concentration of absorbed lactoglobulin in the ten calves fed the calculated amounts of colostrum divided into three equal feeds was slightly lower than the mean value for the ten calves fed colostrum as one meal at one hour post partum. Thus, division of the calculated amount of colostrum into three equal feeds did not result in greater absorptions when relatively large quantities of colostrum were fed. A comparison of the mean 48-hour serum concentrations of absorbed immune lactoglobulin occurring in calves fed standard amounts of pooled colostrum as one single meal or divided into smaller portions is presented in Table 42.

TABLE 42

The Absorption of Immune Lactoglobulin by Newborn Ayrshire CalvesThe Effect of Frequency of Colostrum Feeding

(Calves Fed Single or Divided Meals of Colostrum at Standard Rate.)

<u>Times of Feeding</u> (Hours Post Partum)	<u>No. of</u> <u>Calves</u>	<u>Mean 48-Hour Serum</u> <u>Concentrations of</u> <u>Absorbed Immune</u> <u>Lactoglobulin</u> (Z.S.T. Units)	<u>S.D.</u>	<u>S.E.</u>
1 (single meal)	10	23.0	±6.5	±2.1
5 (single meal)	10	17.7	±4.1	±1.3
9 (single meal)	10	12.5	±2.2	±0.7
1, 5, 9 (divided meals)	10	19.3	±2.5	±0.8

The mean 48-hour serum concentrations of absorbed immune lactoglobulin of those calves fed single meals of colostrum at one or five hours post partum were not significantly different from the meal values for those calves fed at the same rate but where three equal feeds were consumed at one, five and nine hours post partum. However, there was considerably less variation in individual 48-hour serum concentrations of absorbed immune lactoglobulin in the latter group of calves.

Discussion

The first fact to emerge from the experiments carried out in Part 3 was that it was possible, with careful control of feeding and management, to produce very consistent immune globulin absorptions in a large number of newborn calves. In all, 120 calves were used, and 100 of these were fed pooled colostrum. Variations, of course, were found to occur from group to group depending on each group's treatment, but variations in the 48-hour serum concentrations of absorbed immune lactoglobulin within each group was minimal. When colostrum was fed in three equal meals, the widest standard deviation obtained was 3.1 Z.S.T. units in the mothered calves. When colostrum was fed as a single meal, the standard deviations from groups fed at one and five hours post partum were 6.5 and 4.1 Z.S.T. units respectively. The possible reasons for this greater variation are discussed below. The findings in Section III that a linear relationship existed between the whey concentrations of immune lactoglobulin in the colostrums presented to twenty calves and the amounts of this absorbed by these calves further suggests that, under a particular set of circumstances, a fixed proportion only of the immune lactoglobulin presented to a calf may be absorbed. The few calves which were excluded from the experiments were done so only because they were difficult to feed, and no calf which fed well absorbed abnormally low amounts of immune lactoglobulin when compared with the others in its group. These findings would seem to suggest that Ayrshire calves at least are born with an equal absorptive ability and that the view held by Fey (1962) that some calves are congenitally unable to deal with immune lactoglobulin is not likely to be a true assessment of the situation.

The significant difference in absorptive ability that was shown to exist between the pure Ayrshire calves and the Ayrshire-cross-Friesian calves is difficult to evaluate. As yet, no other investigation has been attempted to demonstrate a difference in the absorptive abilities of different breeds of cattle kept under more orthodox management systems. Several studies have shown that a difference does exist in the mortality rates of different breeds (Blakemore, et al., 1948; Withers, 1952-1953; and Leach, et al., 1968). All of these workers have shown that Channel Island and Ayrshire calves are more susceptible to neonatal disease than are Friesian, Shorthorn and cross-bred calves. However, Withers (1952-1953) pointed out that other variables (i.e. management and seasonal factors) might exert hidden influences upon these figures. Friesian calves, even when deprived of colostrum, were found by Lundquist and Phillips (1943) to survive under conditions which were invariably fatal for Guernsey calves. Later work, however, by Aschaffenberg, et al. (1952) showed that colostrum-deprived Friesian calves were just as likely to die as were Dairy Shorthorns (11 out of 12 calves died in each group). The only other published work that has been found which suggests a breed variation in absorptive ability was carried out by Halliday (1966) with sheep. In this work, a significantly higher 48-hour serum gamma globulin was found in Finnish Landrace lambs compared with Merino-cross-Cheviot and Scottish Blackface lambs kept under similar conditions. However, the amounts and immune lactoglobulin concentrations of the colostrum produced by these different breeds was

not investigated. In breeds of sheep as dissimilar as those mentioned, the colostrum may well have been markedly different in volume or immune lactoglobulin content.

Field observations that severe losses in beef calves due to colibacillosis are frequently linked with severely adverse weather conditions during the calving season have been made by several American workers (Amstutz, 1965; Moll, 1965 and Reisinger, 1965). Withers (1952-1953), in a survey of calf mortality in this country, was of the opinion that the very marked seasonal peak of mortalities occurring during the first quarter of the year was "due to the wide temperature variations and absence of sunshine common at this time." These workers have all linked colibacillosis with adverse climatic conditions, but only one (Moll, 1965) has suggested a link between cold weather and reduced globulin absorptive ability in young calves. In fact, no experimental work has shown that exposure to cold will initiate colibacillosis in colostrum-fed calves. The only work suggesting that reduced immune lactoglobulin absorption occurs during periods of bad weather is that by Halliday (1965a) working with lambs and the assumption was here that the lambs were affected in utero. This work has already been discussed in the introduction to this part, where it was suggested that the conclusions were not justified on the evidence presented, bearing in mind all of the other possible variables. Cold ambient temperatures did not depress absorption of immune lactoglobulin in the present study. Calves managed in this way did, however, seem duller and more difficult to feed. In view of this, it seems possible that exposure to severe chilling at birth might just

depress calves to such an extent that the standing times and times to first suckling are delayed, resulting (see Part 1, Section III) in lowered serum immune globulin levels. It must be also remembered that the calves in this series were being fed colostrum at one, five and nine hours post partum; the effect of chilling on calves not being fed colostrum until later might have been different.

It was found possible, using the standardised methods of feeding and management, to confirm the findings in Part 2, Section II, that suckled calves left with their dams were considerably more efficient at absorbing immune lactoglobulin than calves removed from their dams. In the present experiment, it was conclusively demonstrated that this increased absorptive efficiency occurred even when calves were not suckled but fed from a teat bucket in the presence of their dam. As the known literature dealing with the effects of mothering and maternal deprivation has already been discussed in Part 2, it will not be dealt with again. However, certain other interesting findings emerged from this particular experiment.

The actual reason for this increased efficiency of absorption seen in mothered calves is not clear. Some evidence has been presented (Figure 15) to suggest that the rate of absorption of immune lactoglobulin is increased in mothered calves, but it is not known whether or not the time available for absorption is altered. In addition, there is, as yet, no evidence to suggest that the effect of mothering is to somehow modify intestinal epithelial cells. As stated in Part 2, the difference between mothered and non-mothered calves may be simply a difference in oesophageal groove efficiency.

The calves maintained under the standardised conditions were groomed by their dams for a timed 15 minutes. Nevertheless, their 48-hour serum concentrations of absorbed immune lactoglobulin were found in Section 1 (b) to be not significantly different from those of the calves which were not thus groomed and which were exposed to a cold ambient temperature immediately on being born. The suggestion from this observation is that mothering for 15 minutes is not sufficient and must be continued for a longer time before it has a demonstrable effect on lactoglobulin absorption. It may be that grooming has to be continued until after the first meal of colostrum.

In spite of the finding that artificially fed, mothered calves absorbed greater quantities of immune lactoglobulin than non-mothered calves, it was decided to continue using calves removed from their dams at 15 minutes post partum and housed individually for subsequent experiments. This was mainly because far more consistent 48-hour serum concentrations of absorbed immune lactoglobulin were obtained in non-mothered calves, and hence comparison between groups was easier. It may be argued that a significant depression of absorptive ability might have been demonstrated had mothered calves been, for example, subjected to cold conditions. However, previous work (Parts 1 and 2) had shown that no seasonal variation in serum immune globulin concentrations occurred in suckled dairy calves housed with their dams in unheated loose boxes.

Moog (1953) and Halliday (1959) working with mice and rats respectively showed that oral or parenteral corticosteroids initiated premature cessation of immune lactoglobulin absorption. Reports of

similar work, unsupported by data, have suggested that a similar situation may occur in the newborn pig (Payne and Marsh, 1962), and it appears that the effect of corticosteroids on the absorptive efficiency of newborn ruminants has not been hitherto investigated. In the present experiments, large parenteral doses of a corticosteroid preparation significantly increased the 48-hour concentrations of absorbed immune lactoglobulin of non-mothered, newborn calves. In this context, it is interesting to note that Halliday (1959) found that although such injections initiated premature shutdown in suckling rats, the absorptive rate during this shortened period of absorption was apparently increased. Once again, it must be remembered that the experimental calves in this series were fed relatively large amounts of colostrum very early in life. Had the feeding times been delayed, then it is possible that decreased absorptive ability would have been noted.

A very wide variation was found in the immune lactoglobulin concentrations of wheys prepared from different cows' colostrums, and this will be discussed in more detail later in Part 4. As already stated, a very highly significant linear correlation was found to exist between the whey concentration of a given colostrum sample and the 48-hour serum concentration of absorbed immune lactoglobulin in the calf fed that sample. This work confirms observations made by McEwan (1968).

Reference to Table 36 shows that four of the twenty calves in the series were found to have absorbed less than ten Z.S.T. units of immune lactoglobulin by 48 hours post partum. In each of the four instances, colostrum with a very low immune lactoglobulin content had been fed to these calves. In one case, the 48-hour serum concentration of absorbed immune lactoglobulin was found to be 2 Z.S.T. units. This latter calf would normally be classified as completely agammaglobulinaemic in spite of receiving 1725 ml of its dam's colostrum during the first nine hours of life. These findings shed some doubt on the claim by Gay, et al. (1965a) that some of their calves were unable to absorb immune lactoglobulin by four to six hours post partum. Clearly any observations of this nature must include information regarding the time of the first colostrum feed, the volume of colostrum fed, and its immune lactoglobulin concentration.

The experiment investigating the effect of the immune lactoglobulin content of colostrum fed to calves involved 20 calves and extended over a period starting on April 16, 1968 and ending on June 7, 1968. In all cases, the calves were fed their own dam's colostrum. These 20 cows were all from different sources and it seems probable some had been housed for five months prior to parturition whereas others had been grazing for at least one month. In spite of these variations, a highly significant positive correlation ($r = 0.88$, $p = < 0.001$) was obtained when the calves' 48-hour serum concentrations of absorbed immune lactoglobulin were plotted against the appropriate colostrum whey immune lactoglobulin concentrations. Several studies have been carried out to investigate the importance of other factors

in colostrum which may accelerate the absorption of immune lactoglobulin by newborn calves (Deutch and Smith, 1957; Balfour and Comline, 1962; Hardy, 1968), and some have shown that certain factors do accelerate absorption in anaesthetised or decerebrate calves. However, in view of the findings quoted above, it would seem that the significant factor in colostrum is the concentration of immune lactoglobulin. Cows were purposely chosen for this experiment at different times of the year in the hope that acceleration factors might be present in varying amounts, but if they were, their effects were not readily obvious. It would seem that if these factors are involved in globulin transport through intestinal epithelium, their role in the conscious calf fed large quantities of colostrum early in its life are minimal.

It was found possible to feed relatively large volumes of colostrum (i.e. almost 7% of a calf's birthweight) by means of a teat bucket as early as one hour post partum. Two calves out of twelve failed to ingest their appropriate amounts of colostrum at this time and had to be excluded from the series. Nevertheless, these calves did ingest most of their respective amounts. Two further groups of calves comprising ten calves each were similarly fed at five or nine hours post partum, and no trouble was experienced in feeding their measured quantities of colostrum. The volumes of colostrum that were offered to these calves by teat bucket were not significantly different to the six-hour intakes of the mothered and non-mothered suckling dairy calves (see Part 2, Section 1).

The 48-hour serum concentrations of absorbed immune lactoglobulin in the ten calves fed at one hour post partum were on the whole reasonably high, but only one calf had a concentration in excess of 30 Z.S.T. units. The values for the calves similarly fed at five hours post partum were found to be significantly lower than those fed at one hour post partum, and a greater significant difference was demonstrated between those calves fed at five and at nine hours post partum. This indicates that the cessation of immune lactoglobulin absorption in newborn Ayrshire calves is an inexorable process which is initiated at the time of parturition. Further examination of the data presented in Table 40 reveals that the variation in the 48-hour serum concentrations of absorbed immune lactoglobulin in each of the three groups of calves is reduced as the time interval between birth and colostrum feeding is increased. This suggests that some non-mothered calves fed very early in life are less efficient at handling large amounts of colostrum than when fed slightly later. As first suggested in Part 2, it may well be that this is purely an effect of variable oesophageal groove efficiency.

Single feed at various times post partum indicated that colostral shutdown was a fairly rapidly progressing process initiated at the time of birth. Early work on the absorption of immune lactoglobulin by calves indicated that negligible absorption occurred by 24-48 hours of age (Kerr and Robertson, 1944; Comline, Roberts and Titchin, 1951; Deutch and Smith, 1957; Smith and Erwin, 1959 and Pierce, 1961). More recently, work by Kaeckenboeck, et al. (1961) suggested that the rate of absorption in calves was not constant over the first 24 hours of life. They claimed that absorptive efficiency was maximal only for

the first twelve hours of life and then declined rapidly so that by 16 hours post partum the intestine had lost 50% of its absorptive ability. The work presented in Section IV shows that the newborn calf is in an even more precarious position than was suggested by these latter workers. Reference to Figure 19 suggests that by ten hours post partum, an Ayrshire calf's intestinal epithelial cells had lost more than 50% of its initial absorptive ability. Furthermore, it seems likely that the time of complete intestinal shutdown under the conditions of this experiment would be at approximately 18½ hours post partum.

Kaeckenbeeck and Schoenaers (1961) showed that when a given amount of colostrum containing a specific E. coli somatic antibody divided and was fed in three equal amounts, the subsequent absorption of this antibody was almost three times that obtained when the colostrum was fed as one meal. However, these workers were dealing with a total quantity of 600 ml of colostrum. When, in the present experiments, much larger amounts of colostrum were fed to calves as single meals and the resulting 48-hour serum concentrations of absorbed immune lactoglobulin compared with those obtained from calves fed colostrum at the same rate but in three equal feeds, no additive effect in respect of repeated feeding occurred. It is interesting to note that those calves which were fed colostrum in divided amounts showed a much more consistent uptake of immune lactoglobulin than those calves fed large, single meals. Hence, from an experimental viewpoint, divided colostrum feeds are still obviously preferable to single large meals in that comparisons between groups are easier.

To sum up, the results from the experiments carried out in Part 3 of this thesis indicate that several different factors may affect the absorption of immune lactoglobulin by newborn calves. Carefully controlled conditions of management and feeding have confirmed the earlier finding that the mothering of calves increased their globulin absorptive efficiency, but this effect has now been shown to affect even artificially fed calves. It now seems certain that the marked seasonal variation in mean serum immune globulin levels of newborn Ayrshire calves in Scotland is due to variations in the early management of summer and winter born calves in this area. This question has already been fully discussed in Part 2 of this study. Exposure of artificially fed calves to cold temperatures did not reduce their globulin absorptive efficiency so that the possibility that the seasonal variation in mean serum immune globulin levels noted above is due to climatic variations is not likely to be true. Parenteral corticosteroids, when administered to newborn calves prior to colostrum feeding, resulted in significantly higher serum immune globulin concentrations in comparison to non-treated controls.

Colostrum shutdown is apparently initiated at birth and following this, the efficiency of absorption would appear to fall steeply. Consequently, it is necessary to re-emphasise the traditional advice with regard to the early feeding of colostrum and, moreover, to stress the fact that a very large initial volume should be given. Calves will usually ingest colostrum to the extent of 7% of their birthweight within the first few hours of life, and the impression

is that smaller calves would frequently ingest more if this were offered. Ingestion of large quantities such as this did not, in the present study, produce any serious adverse effects.

With careful standardisation of management and feeding, it was found possible to produce very consistent immune lactoglobulin absorptions within any groups of calves. It was found that this technique was very useful when studying factors affecting absorption of immune lactoglobulin. It is suggested that further studies should be carried out on the same or similar lines in order to rule out variables which in the past have frequently given rise to confusion. In any event, it would seem that any further experiments or clinical observations in this field should define:

- (a) The exact time of the first colostrum feed.
- (b) The amount of colostrum ingested.
- (c) The immune lactoglobulin concentration of the colostrum ingested.
- and (d) Whether or not the calf was mothered, and, if so, for how long.

PART 4

PART 4

STUDIES ON COLOSTRUM

Introduction and Review of the Literature

Colostrum has been defined as "that peculiar glutinous emulsion secreted by the mammary gland of all mammals in the period immediately following parturition" (McGirr, 1947). The Latin word "colostrum" suggests an emulsion of egg yolk in turpentine--a mixture which McGirr (1947) claims resembles bovine colostrum. However, the appearance and consistency of bovine colostrum may vary considerably and the secretion from one cow may resemble thin honey, whereas that from another may appear indistinguishable from normal cow's milk (Hill, Widdowson and Maggs, 1950).

The amounts of colostrum that can be removed from non-premilked cows shortly after parturition by general milking techniques may vary from $\frac{1}{2}$ - 22 lbs. (Hill, et al., 1950). Other workers have quoted a value of 20 lbs. (Kuttner and Ratner, 1923), and mean values of 12.7 lbs. (Sutton, Warner and Kaiser, 1947) and 14.7 ± 5.2 lbs. (Eaton, Johnson, Matterson and Spellman, 1949). Houdiniere (1945) suggested that bovine colostrum was "probably the major product of the mammary gland until man began to select and breed animals for more persistent milk production."

The difficulties of rearing calves deprived of colostrum were well recognised by the early 1900's but no clear-cut experimental evidence of this appears to have been produced until Smith and Little (1922a) compared two groups of calves (ten colostrum fed and twelve

colostrum deprived) in an experiment carefully designed to rule out other factors. All ten colostrum-fed calves survived, but nine of the twelve colostrum-deprived calves died from colisepticaemia (seven in less than six days). One of the three remaining calves became chronically ill and after slaughter on the 27th day, post-mortem examination revealed bacterial infection in a joint and both kidneys. The other two survived although one suffered a transient lameness and the other, a rhinitis.

This fundamental experiment demonstrated conclusively the importance of colostrum to the calf. Smith and Little (1922) concluded that "colostral function is essentially protective against the miscellaneous bacteria which are harmless later on when the protective functions of the calf have begun to operate and accumulate energy."

Since that time the search for the protective factor or factors has been concentrated mainly on research into two groups of compounds, first vitamin A and carotene and second, the proteins of colostrum.

The classical studies on vitamin A in mammals were carried out by Mellanby and Green (1929), who concluded that depletion of this vitamin predisposed to bacterial infection due to the effect on epithelial structures, hence facilitating bacterial penetration. Mainly as a result of their work, the term "anti-infective vitamin" was coined.

Dann (1933) compared the colostrum and milk of 14 Shorthorns and found that colostral Vitamin A levels were 10-100 times greater than those of later lactation milk. Several other workers soon verified his findings, but Stewart and McCallum (1938a) criticised

this work on the grounds that too few experimental animals were used. These latter workers (Stewart and McCallum, 1938a) studied the colostrum from 100 cows and heifers in central and western Scotland, stressing that almost all were of the Ayrshire breed and that all were similarly managed and fed since the samples were taken from animals calving in February, March, April and May, while being stall fed. They found that certainly there were high levels of carotene and vitamin A present in colostrum, but that the levels varied greatly (from 35-1181 International Units of vitamin A/100 ml colostrum). They also noted that there was a rapid fall in these levels following parturition so that by four days post partum, the values obtained were usually similar to those seen in milk (i.e. 30-50 International Units of vitamin A/100 ml colostrum). These observations have since been verified by other workers (Hansen, Phillips and Smith, 1946; Spielman, Thomas, Loosli, Whiting, Norton and Turk, 1947; Sutton, et al., 1947 and Thompson and McGillivray, 1957). As a result of their studies, Stewart and McCallum (1938a) advised early colostrum feeding to ensure the maximum vitamin A intake by calves.

In a later paper, Stewart and McCallum (1938b) fed colostrum of different vitamin A levels to calves and followed the performance of these calves. In all, 206 calves were used (95 in 1937 and 111 in 1938). It was found that where colostrum containing more than 250 Blue (Moore's) units of vitamin A was fed, the overall mortality rate due to "white scour and allied infections" (i.e. joint ill and navel ill) was 0.66%. The mortality rate from similar causes in calves fed colostrum containing less than 250 Blue Units vitamin A was 16%.

Unfortunately, no information was given regarding the amounts of colostrum fed or the times and methods of initial feeding. Nevertheless, for the first time since the classic experiments of Smith and Little (1922a) which showed the value of feeding calves colostrum, there appeared a highly significant difference ($p = <0.001$) in mortality rates related to a single constituent of colostrum. The above results naturally stimulated much research into the carotene and vitamin A levels of colostrum and the possible use of these compounds in the prophylaxis of colibacillosis in calves.

An early finding (Dann, 1933), Stewart and McCallum, 1938a) was that levels of colostral carotene and vitamin A varied markedly between individuals although later (Sutton, *et al.*, 1947) it was shown that this variation was at its greatest in the first milking post partum and within 24 hours became much less marked.

Dann (1933) showed that the levels of carotene and vitamin A were higher in the secretion of heifers than of cows. This was later verified by Hansen, *et al.* (1946) and Thompson and McGillivray (1957). However, the latter authors (Thompson and McGillivray, 1957) point out that although the concentration of these constituents was higher in their heifers, the total daily yield of the same constituents was highest in their cows. Stewart and McCallum (1938a) did not find cow-helifer differences but did show that the colostral concentrations of carotene and vitamin A were positively correlated with the length of the preceding dry period. Even when very close standardisation of feeding and management was carried out, Hansen, *et al.* (1946) still found a wide individual variation between cows of the same age.

Although Stewart and McCallum (1938a) recorded wide variations between the colostrum carotene and vitamin A levels of individuals of the same breed (i.e. Ayrshires), other workers, dealing with fewer animals, have claimed regular breed differences. Sutton, et al. (1947) found that carotene levels were very high in Guernsey and lowest in Friesian colostrum, and Parrish, Wise, Atkeson and Hughes (1949) agreed that colostrum carotene levels tended to be highest in the Channel Island breeds. Sutton, et al. (1947) found less breed variations in colostrum vitamin A levels, but that Brown Swiss cows produced the highest and Jerseys the lowest concentrations. Parrish, et al. (1949) failed to demonstrate definite breed variations in colostrum vitamin A.

The effect of diet on colostrum carotene and vitamin A levels was first investigated by Stewart and McCallum (1942). They found that supplementation of the winter rations of cows with 3 lbs. of carrots or 1/7 pints of cod liver oil daily for one to six months pre partum had no effect on colostrum carotene or vitamin A concentrations. Spielman, et al. (1947) found that diets deficient in carotene resulted in low colostrum carotene levels, carotene supplemented diets increased colostrum carotene but not vitamin A levels, and that vitamin A supplemented diets resulted in high colostrum vitamin A levels. In this experiment supplementation was at the rate of one million International Units of carotene daily and 1 million International units of vitamin A daily in the respective groups for the last 60 days of pregnancy. Henry, Houston and Kon (1940) and Parrish, et al. (1949) agreed with this finding in that

they found that access to pasture in the last few weeks of gestation results in higher carotene and usually higher vitamin A levels in colostrum. On the other hand, Sutton, et al. (1947) failed to demonstrate this.

Vitamin A can exist in two forms, namely, alcoholic and esteric vitamin A. Work carried out by Spielman, et al. (1947) showed quite conclusively that irrespective of the form of this vitamin in the diet, the esteric form was that which was almost exclusively present in colostrum. Thompson and McGillivray (1957) agreed on this point, stating that less than 2% of the total vitamin A in colostrum was in the alcoholic form.

Sampling and milking procedures are also very important factors in determining the concentrations of colostrum carotene and vitamin A obtained. This could, in part, explain the wide variations seen in the Stewart and McCallum (1938a) experiments, for example, where many different farmers were providing the necessary colostrum samples. Comparisons between the concentrations obtained in other experiments is difficult due to the varied sampling techniques. For example, Hansen, et al. (1946) pooled the first two milkings post partum, whereas Parrish, et al. (1949) used colostrum from the first milking only, and Spielman, et al. (1947) pooled the first pint taken post partum from each quarter.

Hansen, et al. (1946) showed that where calves were removed from their dams before the first milking, the drop in colostrum vitamin A between the first and second milkings was less marked than when the

calves remained with their dams for both milkings. In view of the fact that ten International Units of oxytocin administered intramuscularly to cows prior to the first milking produced similar results in the absence of their calves suggested that the calf's presence allowed greater initial evacuation of the udder (i.e. a more complete "let down"). Eaton, et al. (1949) showed that as colostrum was withdrawn from the udder, there was a significant positive trend in fat, carotene and vitamin A concentrations with each 2 lb. increment of colostrum. When carotene and vitamin A levels were expressed, not in terms of colostrum concentrations, but per gram of butterfat, there was still a positive trend on evacuation for carotene but not for vitamin A.

It may, therefore, be seen that in any experiment where levels of colostrum carotene and vitamin A are to be measured, it is imperative to evolve a standard routine of milking and sampling and that for comparative studies, the suggestion by Spielman, et al. (1947), that colostrum carotene and vitamin A concentrations should be expressed per gram of fat, should be adhered to. This, of course, will rule out variations due to varying butterfat percentages.

Thompson and McGilivray (1957) studied the drop in plasma carotene and vitamin A levels apparently associated with the drain of these compounds into the colostrum at the time of parturition. This was followed by a gradual rise in plasma levels over the following three weeks. The fall in plasma levels, they claimed, was irrespective of vitamin A intake. Walker, Thompson, Bartlett and Kerr (1949) had previously showed, however, that this drop in plasma levels at

parturition could be minimised if supplementary carotene and vitamin A were supplied in late gestation. Spellman, et al. (1947) demonstrated a highly positive correlation between plasma carotene and vitamin A concentrations 18 days pre partum, and colostrum concentrations at parturition.

Dann (1932) first discovered that calves were born with little or not vitamin A reserves in liver or plasma and since then, this finding has been substantiated by numerous workers (Phillips, Lundquist and Boyer, 1941; Hibbs and Krause, 1947; Hansen, et al., 1946). The fact that calves born to dams maintained during gestation on a diet deficient in carotene or vitamin A may be stillborn or show blindness and weakness suggests, however, that in cattle on a non-deficient diet some vitamin A must pass to the calf in utero (Spellman, Thomas, Leosli, Norton and Turk, 1946). These workers succeeded in increasing in carotene and vitamin A levels in precolostral calf serum by dietary supplementation of the dams for 60 days pre partum.

It is nevertheless obvious that calves have to rely upon a supply of colostrum for the major part of their vitamin A reserves and Sutton and Kaesser (1946) demonstrated a rapid rise in plasma vitamin A following ingestion of colostrum by calves. They also showed that carotene and vitamin A absorption occurred continuously at least over the first week when the feeding of colostrum was extended over this period.

In spite of the findings already mentioned (especially those of Stewart and McCallum, 1938b) resulting from the tremendous upsurge of interest in carotene and vitamin A between 1930 and 1950, the results of vitamin supplementation in attempts to prevent calf losses have been

mixed and on the whole, disappointing. Phillips, et al. (1941) demonstrated that colostrum-deprived calves fed on skimmed milk and a vitamin supplementation stood a better chance of surviving (mortality 25%) than did colostrum-deprived calves fed on skimmed milk alone (mortality 100%). However, their work really again underlined the value of colostrum feeding as no calves in their colostrum-fed control group died. Because most work on vitamin supplementation had been carried out either on calves from vitamin A deficient dams or on calves on low carotene and vitamin A diets, Hibbs and Krause (1947) investigated the value of vitamin A supplementation in traditionally managed, colostrum-fed calves. They found no decrease in the incidence of scouring, and this was in agreement with work carried out by Norton, Eaton, Loosli and Spielman (1946). The low plasma vitamin A levels associated with diarrhoea in calves were a result of the condition, presumably due to malabsorption (Sutton and Kaeser, 1946; Hibbs and Krause, 1947).

Final realisation that vitamin A deficiency was not the major factor involved in neonatal calf disease and mortality came with work by Blakemore, Davies, Eysenburg, Moore, Sellers and Worden, (1948). They initially failed to rear colostrum-deprived calves maintained on skimmed milk and supplementary vitamin A. Another group of calves was deprived of colostrum and maintained on skimmed milk but these latter calves were inoculated at birth with "precolostrum" and all survived. The precolostrum was shown to be rich in antibodies and the conclusion was that the protective factor was the globulins of colostrum, not the vitamin A content. Unfortunately, no detailed information was presented regarding these experiments.

Soon after this, work carried out by Aschaffenburg, Bartlett, Kon, Terry, Thompson, Walker, Briggs, Cotchin and Lovell (1949a) demonstrated conclusively that the protective factor in colostrum was carried in the aqueous and not in the fatty fraction. The mortality rate in eight calves initially fed the classified fatty fraction of colostrum was 50% compared with 8% in 24 calves fed the aqueous fraction. The cause of death in most cases was colisepticaemia. Once again, all 11 calves fed whole colostrum survived. In a later paper (Aschaffenburg, Bartlett, Kon, Walker, Briggs, Cotchin and Lovell, 1949b) the amounts of the aqueous fraction were reduced so that some calves received as little as 80 ml in place of colostrum. Nevertheless, all of these calves survived in spite of often profuse diarrhoea, whereas 83% of colostrum-deprived controls died.

In the light of these results it is very difficult to understand the highly significant results obtained by Stewart and McCallum (1938b). However, Rowland, Roy, Sears and Thompson (1953), while studying pre partum milking in cattle, demonstrated a significant correlation between the daily concentration of carotene and vitamin A and the daily weight of globulin in precolostrum.

Gay, et al. (1965a) have shown a definite relationship between the serum immune globulin concentration of young market calves and the subsequent fate of these calves, with low levels of immune globulin being associated with deaths from colisepticaemia and neonatal diarrhoea and very high levels with survival under the same conditions. More recently, McEwan (1968) has shown that under conditions of early colostrum feeding there is a significant correlation

between the amount of colostrum globulin presented to a calf and its subsequent serum immune globulin concentration. In short, it may be that findings of Rowland, et al. (1953) for precolostrum are also true for colostrum and that the high vitamin A colostrum of Stewart and McCallum (1938b) also contained high levels of immune lactoglobulin and vice versa. Hence, the protective factor may have been immune lactoglobulin and not vitamin A.

Elcheberg, cited by Famulener (1912), found that, compared with milk, cow's colostrum had a high globulin content. Crowther and Reistrick (1916) agreed with this finding, stating that the high total protein concentration of colostrum was due to its very high globulin content. Since that time, these observations have been substantiated many times using many different methods (Hill, et al., 1950, Rowland, et al., 1953, and Dixon, Weigle and Vazquez, 1961). Pierce (1962) discussed in detail the major colostrum proteins and, more recently, Gay (1965) reviewed the literature regarding the immune lactoglobulin of colostrum. According to Pierce (1962) the colostrum proteins and their relative proportions shown electrophoretically are as follows:

1	2	3	4	5
(Casein)	(Beta-lacto- globulin)	(Lact-albumin)		(Immune lacto- globulin)
2.5%	14.4%	6.5%	8.7%	67.8%

Gay (1965) suggested that the percentage of immune lactoglobulin in the total colostrum proteins was 54%-55%, while Dixon, et al. (1961) put the percentage much higher (at a mean of 78%). These variations are almost certainly due to sampling differences (for example, Pierce (1962) pooled the first three milkings post partum, whereas Dixon, et al. (1961) quote figures for the first milking only). The total protein content of precolostrum is even higher than that of colostrum at parturition (Rowland, et al., 1953), and this very high level of protein is due to the higher concentrations of globulin in precolostrum (Hill, et al., 1950). Dixon, et al. (1961) showed that in the one animal they sampled for precolostrum at one week pre partum, the percentage of globulin in the total protein was 95%.

The globulin "build up" in precolostrum starts at about one month pre partum, but reaches a peak during the last week of gestation (Larsen, 1958). The "build up" is apparently triggered off by the approach of parturition and not by the act of "drying off" (Larsen, 1958). Rowland, et al. (1953) showed a peak in globulin secretion by the cow's udder at about one week pre partum in spite of the fact that these cows were being pre milked twice daily.

Askonas, Campbell and Work (1954) using radioactive amino acids showed that casein and beta-lactoglobulin were synthesised by the udder of the goat; however, there has been much controversy regarding the source of immune lactoglobulin and whether or not it is identical to maternal serum gamma globulin (Smith, 1946, and Pierce, 1955). Largely as a result of work by Larsen and Kendal (1957), Dixon, et al. (1961) and Pierce (1962), the situation has been clarified and

most workers now accept that the two protein complexes (i.e. maternal serum gamma globulin and immune lactoglobulin) are closely similar, if not identical. Pierce (1962) has suggested, however, that some structural modification may take place during the phase of concentration in the udder.

Due to the cessation of gamma globulin secretion by the udder of the newly dried off cow, there results a phase of hypergamma-globulinaemia, (Dixon, et al., 1961). This is later followed by hypogammaglobulinaemia due to the drain of globulin into the udder (Larsen and Kendal, 1957). This drop in the serum gamma globulin complex has also been noted by Dixon, et al. (1961) and occurs during the last 30-40 days of gestation (Pierce, 1962). Quantitatively, the loss from the serum parallels the gain by the udder and during the last three weeks of pregnancy, approximately 34 gms of gamma globulin may pass daily to the udder (Larsen and Kendal, 1957). Garner and Crawley (1958) injected radio-iodinated gamma globulin intravenously into a four months' pregnant cow and failed to detect any concentration of this by the udder. However, they did note that in later pregnancy, a heifer concentrated the injected gamma globulin two or three fold. Also, secretion of the gamma globulin into the precolostrum ceased abruptly at parturition. Rowland, et al. (1953) suggested that the hormonal stimulus for the initiation of milk secretion also allowed globulin to pass from the serum into the udder. Dixon, et al. (1961) claim, in addition, that there is an increased rate of gamma globulin synthesis at parturition.

Wide variations are found in the concentrations of colostral globulin at the first milking following parturition and in the secretion from pre milked cows (Hill, et al., 1950). They postulated that the parturient cow's udder simultaneously produces two secretions. First, a honey-like secretion rich in protein and second, milk, is produced. This theory is partially borne out by their observations that the viscosity of at least precolostrum is proportional to its globulin content. Also, if this theory is true, then the globulin concentration is proportional to the milk yield at parturition and hence the weight of globulin in colostrum should be more constant than the concentration, and this was, in fact, found to be the case.

There appears to be no published work regarding the effect of diet on the immune lactoglobulin concentrations of bovine colostrum, although Humphrey and White (1966) stated that the serum globulin concentration of starved prisoners-of-war were not significantly reduced.

Eaton, et al. (1949) found a negative trend in protein concentration as colostrum was withdrawn from the udder during a particular milking but stressed that this was quantitatively insignificant.

Several studies have been carried out on the effect of pre partum milking on the quality of the secretion at and before parturition (Hill, et al., 1950; Aschaffenburg, Bartlett, Kon, Roy, Walker and Lovell, 1951a; Rowland, et al., 1953 and Dixon, et al., 1961). Unlike the others, Hill, et al. (1950) claim that the first post partum secretion from pre milked cows has no statistically

significant depression in globulin content compared with non-premilked cows. However, nine of their sixteen premilked cows were only milked for two or three days pre partum so that the secretion might well have been similar to ordinary colostrum. In the other experiments mentioned, the cows were milked twice daily for one to two weeks pre partum, and the globulin contents of the secretions at parturition were much lower than in the non-premilked controls.

Following parturition in the non-premilked cows, there is a steep fall in colostral protein, due entirely to the decrease in globulin content (McGirr, 1947; Hill, *et al.*, 1950). Consequently, by 24 hours post partum the total protein may have fallen from 14 gm% to 4 gm% (Hill, *et al.*, 1950).

Erlich (1892) actively immunised adult female mice against vegetable toxins (ricin and abrin). He found that it was possible to transfer this immunity from mothers to their young within ten days of birth. Fanulener (1912), working with goats, found that anti-sheep erythrocyte haemolysin was acquired by kids after suckling. He also found that the dam's colostral whey titre was higher than maternal serum, that no in utero transfer occurred, and that this passive immunisation of kids could only be carried out within the first few days of life. Gay (1965) made the point that most subsequent work in other ruminants has only confirmed these findings in other ways.

Howe (1921), using sodium sulphate precipitation methods, found that precolostral calf serum contained no euglobulin or pseudoglobulin I. However, a heavy precipitate was formed (i.e. these constituents appeared in calf serum) soon after suckling. He made the point that only when colostrum was ingested did this phenomenon occur.

Orcutt and Howe (1922) noted that the absorption of Brucella abortus agglutinins after a meal of colostrum occurred simultaneously with the absorption of globulins. Smith and Little (1923) found that a newborn calf absorbed Br. abortus agglutinins following a meal of adult cow serum. The same authors (Smith and Little, 1922b) had previously noted that the protective factor of colostrum was presumably present in adult cow serum in that calves injected and fed with this had almost as low a mortality as colostrum-fed calves. Jameson, Alvarez-Tostado and Soltor (1942) and Smith and Holm (1948) showed electrophoretically the appearance of a slow gamma globulin fraction in newborn calf serum following a meal of colostrum. Deutch and Smith (1957) similarly demonstrated the appearance by ultracentrifugation of a 6.5 S component in calf serum corresponding, they claimed, to colostrum and serum gamma globulin. Other workers have studied the absorption in the newborn calf of specific antibodies in colostrum. It has been thus shown that antibodies to Br. abortus (McDiarmid, 1946), E. coli (Kaeckenbeeck, Colinet and Schoenaers, 1961), Foot-and-Mouth disease virus (Graves, 1963), Trichomonas foetus (Kerr and Robertson, 1944) and Rinderpest (Brown, 1958), among others, are transmitted to the newborn calf solely via dams' colostrum.

Pierce (1962) underlined the speed and extent of immune lactoglobulin absorption in the newborn calf. He cited an instance where the serum gamma globulin level of a calf rose from 4% (precolostral serum) to 27% only 5 1/4 hours after suckling. The fact that immune lactoglobulin absorption can only take place within a short time of birth is made quite clear by the above workers. Kaeckenbeeck, et al. (1961) stated that such absorption is for only twelve hours following birth and is completely absent by 36 hours post partum.

Earle (1935) showed that newborn foals, kids, lambs and pigs are also agammaglobulinaemic at birth and receive globulins from their dams' colostrum.

The majority of the above workers have stressed that precolostral calf serum is agammaglobulinaemic and/or devoid of specific antibodies. However, recent studies using the more sensitive technique of immunoelectrophoresis have demonstrated low levels of immune globulin in some foetal calf sera (Kniazeff and Rimer, 1967 and Schimmel, 1966; Penhale, 1968). It is not clear from this work whether placental transfer or foetal synthesis of gamma globulin was taking place. However, foetal synthesis of antibodies to Leptospira saxkoebing has been demonstrated when calf foetuses are artificially infected after the 132nd day of pregnancy (Ingram and Smith, 1965). Foetal synthesis of 7 S gamma globulin has been demonstrated in lambs born to ewes experimentally infected with Border Disease (Gardiner, 1967).

Practically, the importance of very small amounts of precolostral gamma globulin can have no significant effect on immunity to colibacillosis in calves, since colostrum-deprived calves so frequently fail to survive artificial or natural challenge. Nevertheless, the presence of precolostral or antibodies must be considered when choosing precolostral sera for viral pathogenesis or reagent serum production (Kniazeff and Rimer, 1967).

Initially, the obvious advantages of feeding newborn calves colostrum were thought to stem from its high nutritive value and laxative effect (Howe, 1921). Later, the vitamin A content of colostrum was held to be the factor responsible for the protection of calves

against colibacillosis (Stewart and McCallum, 1938b). More recently, the protective factor has been demonstrated to be present in the aqueous fraction of colostrum (Aschaffenburg, Bartlett, Kon, Terry, Thompson, Walker, Briggs, Cotchin and Lovell, 1949a), more specifically, the immune lactoglobulin fraction (Pierce, 1962).

The protective factor appears to be immunological since protective factors other than the antibody content of the absorbed globulin have been investigated by some workers, but little evidence of other protective actions have been demonstrated. McCance and Widdowson (1959) working with newborn pigs claimed to have demonstrated plasma expansion due to the osmotic action of absorbed globulins. However, their control animals were fasted. Subsequently, work with calves (McEwan, Fisher and Selman, 1968) showed an equal plasma expansion in colostrum-fed calves and milk-fed controls. Fisher, Selman, McEwan and de la Fuente (1968) also showed no increased buffering capacity with increasing serum globulin levels in newborn calves.

Briggs (1951) suggested that the presence of K antigens was associated with the pathogenicity of certain strains of E. coli. He showed that colostrum fed to eleven calves which subsequently died of colisepticaemia contained no K antibody. Working with mice, he had previously shown that the presence of the respective K antibody was the death-preventing factor in antiserum and that O antibody conferred little protection. Briggs, Lovell, Aschaffenburg, Bartlett, Kon, Roy, Thompson and Walker (1951) extended this study and claimed that there was good evidence to suppose that colostrumal protection of

calves was linked to its K antibody content. Ingram, Lovell, Wood, Aschaffenburg, Bartlett, Kon, Palmer, Roy and Shillam (1956) studied 103 colostrum-deprived and 225 colostrum-fed calves. Of these, 94 colostrum-deprived and 59 colostrum-fed calves died. After studying the colostrum fed to these latter calves, they concluded that the protective factor in the colostrum fed to the survivors was the presence of the K antibody. However, Fay and Margadant (1961) found that they could confer immunity to experimental challenge with E. coli strain (078:K80B) by feeding colostrum containing no K antigen to this type.

Gay (1965) argued convincingly on two grounds that the work by Ingram, et al. (1956) did not demonstrate the protective value of K-antibody. First, no data was supplied regarding the K antibody of the colostrum fed to the surviving calves. Second, so many colostrum-fed calves died of colisepticaemia (29/59) that it seems likely that many of these did not absorb immunoglobulins from the colostrum with which they were fed (i.e. the assumption was made the colostrum feeding is synonymous with immune lactoglobulin absorption). Gay (1965) stated that colostrum-fed calves are resistant to experimental infection with serotypes of E. coli associated with colisepticaemia regardless of the presence or absence in their serum of specific agglutinins against these serotypes. In fact, it would seem that the only way to infect colostrum-fed calves with colisepticaemic strains of E. coli is either by infecting them before feeding colostrum (Fay and Margadant, 1961) or by introducing suspensions of the organism intra-peritoneally (Dam, 1967). As little as 80 ml of the

aqueous fraction of colostrum were found in one study to protect against death from colisepticaemia, although diarrhoea still occurred (Aschaffenburg, et al., 1949a).

Smith (1965) stated that neither O or K agglutinins in colostrum were responsible for protection against E. coli bacteraemia in calves. He claimed that the protective factors in colostrum were specific bactericidal antibodies (opsonins or bacteriotrophins) which, on absorption, were activated by complement, which is present in the serum of all calves at birth.

Fey and Margadant (1961) showed that 21/22 calves which died of colisepticaemia were either hypogammaglobulinaemic or agammaglobulinaemic. This was in spite of the fact that they were claimed to have been fed colostrum, although this feeding in many cases was delayed until after 12 hours post partum. Gay, et al. (1965a) demonstrated a relationship between the total serum immune globulin content of 178 young calves (as measured by the Zinc Sulphate Turbidity test) and the subsequent fate of these calves. Colisepticaemia only occurred in those calves with little or no serum immune globulin, and calves with very high levels of serum immune globulin survived under similar conditions. Deaths from the effects of diarrhoea also predominantly occurred in the low serum immune globulin calves. These 178 calves had no known history regarding colostrum feeding as all were purchased in local markets. The inference here that the serum level of specific antibody to E. coli bore no relationship to susceptibility to colisepticaemia but that the prime factor was a non-specific deficiency of serum gamma globulin was questioned by Penhale (1965). He pointed

out that a deficiency of serum gamma globulin implied a deficiency of specific antibodies. Penhale (1965) also claimed that E. coli antibodies are not gamma globulins, but electrophoretically faster β_2 M macroglobulins.

There is some evidence to suggest that specific agglutinating antibodies to pathogenic strains of E. coli will protect a calf from the enteric form of colibacillosis (Gay, 1965). However, experiments whereby dams have been vaccinated with pathogenic strains of E. coli with a view to boosting colostrum whey titres, and hence the resistance of their calves, have, on the whole, been disappointing (Reisinger, 1965). Schoenaers, Kaeckenbeeck and el Nagoh (1967) vaccinated 2610 cows at seven months of gestation on 172 farms, the vaccine consisting of five commonly pathogenic serotypes of E. coli and Freund's adjuvant. The overall mortality rate from colibacillosis in the previous year had been 24.5%, and they claimed to have reduced this to 3.7% (this figure includes 34 abortions which were possibly due to the vaccine). It is difficult to evaluate the efficacy of this vaccination programme for two reasons. First, from the initial 2610 cows, 538 calves (i.e. 21%) were eliminated for various reasons (e.g. sale of dam, no data, abortions, stillbirths, embryotomy and calves not receiving colostrum). Presumably, colostrum-deprived calves were included in the previous year's figures. Second, there is no information as to whether or not improved colostrum consciousness occurred in the farmers as a result of dam vaccination and the resultant underlining of the immunological properties of colostrum. The fact that so few of the recorded deaths from colibacillosis were due to colisepticaemia might suggest that colostrum feeding had been more conscientiously carried out than in previous years.

Ingram and Smith (1965) reviewed the literature concerning the immunological responses of young animals and concluded that the presence of relatively high titres of antibody acquired passively through colostrum interfered with the active production of antibody in young animals. McDiarmid (1946), working with Br. abortus, claimed that the disappearance of maternally derived antibody from a calf's serum depended on the colostrum whey titre and hence on the calf's initial serum titre of agglutinins to Br. abortus. Gay (1965) stated that the rate of decrease in antibody levels depends on the antigen. The situation has very serious implications when it comes to vaccination control of certain serious diseases. It is well known that the colostrum-deprived calf can respond antigenically within the first few days of life (Brown, 1958; Pierce, 1962). This response does depend though on the antigen and occasionally colostrum depresses antibody formation even when it does not contain antibodies to the challenging antigen (Smith and Ingram, 1965). However, maternally acquired antibodies have been found to depress a calf's antigenic response and consequently where a calf is born to a cow resistant, for example, to Rinderpest or Foot-and-Mouth disease, vaccination must be delayed and the delay may place this young animal at risk (Brown, 1958; Graves, 1963). A similar problem is encountered in the control of canine distemper (Baker, Robson, Gillespie, Burghar and Doughty, 1959) and the Colostrum disease of sheep (Cooper, 1967).

To sum up, the protective factor in colostrum is in the immune lactoglobulin fraction which is derived from the dam's serum globulins. This may be absorbed unchanged through the calf's intestinal wall in the early neonatal period. The nature of this protection is imperfectly understood and controversial. It may be due to the wealth of specific antibodies present in the fraction or to a non-specific effect of serum gamma globulin. Some early work suggesting that colostrum vitamin A has a protective function still cannot be discounted.

From the above discussion it is obvious that a great deal of work has already been carried out on the proteins of bovine colostrum. However, little interest has apparently been paid to the very marked individual variations in the immune lactoglobulin concentrations of different cows' colostrum at the first milking post partum. The work already presented in Parts 1, 2 and 3 of this thesis has indicated that the seasonal variation in the serum immune globulin concentrations of calves in the West of Scotland is due mainly to differences in early management between summer and winter born calves. However, under certain conditions a close relationship exists between the concentration of immune lactoglobulin in colostrum and the resulting serum immune concentration of a calf fed that colostrum. It was felt that if lower concentrations of immune lactoglobulin occurred during the winter months, the decreased absorptions resulting from winter management regimes might be accentuated. Consequently, it was decided to analyse a large number of colostrum samples from non-premilked cows and heifers calving at different times of the year in order to see if such a seasonal variation did, in fact, exist.

It has been shown that a correlation does exist between the vitamin A and the globulin concentrations of precolostrum. The opportunity was therefore also taken to measure the carotene and vitamin A levels in the samples in order to see if such a relationship existed in colostrum.

Materials and Methods

1. Milking technique

The cows and helpers which produced the colostrum samples studied in the following experiments were entirely free of clinical mastitis, and none had been premilked. As already stated (see Materials and Methods, Part 3), the cows and helpers were removed from their calving accommodation 15-30 minutes post partum, and they were milked at varying times after this, but never more than five hours post partum. An attempt was made to milk the animals by a standard technique at all times. To that end, the cows and helpers were all machine milked using a portable, electrically operated, single unit machine (Alfa Laval Co. Ltd., Cwmbran, Monmouthshire).

Every attempt was made to evacuate each animal's udder as completely as possible, but stripping was limited to machine stripping. At no time was a dam in the presence of its calf.

When the quantity of colostrum produced was measured, it was done so to the nearest 0.25 litre using a one litre measuring cylinder.

All colostrum samples were stored at -4°C until it was convenient to carry out the necessary determinations.

2. The determination of vitamin A and carotene in colostrum

The method used was an adaptation of the rapid procedure for the extraction and determination of vitamin A and carotene in milk as described by Boyer, Splitzer, Jensen and Phillips (1944). The modifications employed were first suggested by Parrish, *et al.* (1949) to compensate for the higher viscosity and greater vitamin A and carotene concentration of colostrum.

Boyer, et al. (1944) make the point that the method of determination employed below measures total carotenoids. However, the carotenoids in cows' milk (and presumably colostrum) are principally beta-carotene although other pigments (e.g. xanthophylls) are known to be present. In this method of determination, the carotene concentrations were directly measured in a photo-electric colorimeter (Evans Electroselenium, Halstead, England).

The determination of vitamin A utilized the Carr-Price reaction, which occurs when antimony trichloride reacts with vitamin A and which produces a transient blue colour. The Carr-Price reaction was carried out in a tube placed in the colorimeter as the blue colour is immediately produced and fades rapidly. An attempt was made to rule out the low grade Carr-Price reaction produced by the carotenoids.

(a) Reagents

Unless otherwise stated, all reagents used were of the analytical reagent (Analar) grade and were supplied by B.D.H. Laboratory Chemical Division, Poole, England.

(i) Potassium hydroxide solution

Ten millilitres of distilled water was added to 20 grms of potassium hydroxide pellets (May and Baker Ltd., Dagenham, England), and after the pellets had dissolved, the solution was mixed and shaken with 90 ml absolute ethyl alcohol.

(ii) Antimony trichloride solution

Twenty grams of antimony trichloride was rapidly weighed and then transferred to a brown, glass-stoppered bottle. One hundred ml of chloroform was then added, and the mixture was stirred until solution was complete. Before use, the antimony trichloride solution was filtered using a rapid qualitative paper.

(iii) Acidified alcoholic wash solution

One millilitre of concentrated hydrochloric acid was added to 100 ml of ethyl alcohol, and this mixture was then made up to 1 litre with distilled water.

(iv) Other reagents

- (1) Petroleum ether (40°C-60°C)
- (2) Absolute ethyl alcohol
- (3) Chloroform
- (4) Acetic anhydride

(b) Equipment

(i) A portable photo-electric colorimeter and matched tubes (Evans Electroselenium Ltd., Halstead, England).

(ii) A rapid delivery pipette was constructed whenever necessary simply by attaching a three-way, bulb pipette filter to a 10 ml glass pipette.

(iii) Glass stoppered, pear shaped separating funnels (250 ml).

(iv) Round bottomed flasks (250 ml).

(v) A pressure/vacuum pump (A.E.I., Motor and Control Gear Division, Newcastle, England).

(c) Extraction Procedure

Thirty millilitres of the alcoholic potassium hydroxide solution were added to 20 ml of colostrum in a separating funnel, and after shaking, the mixture was allowed to stand for three hours.

After standing, 50 ml of diethyl ether were added and the tightly stoppered separating funnel was briskly shaken for about one minute. Once again, the mixture was allowed to stand and separation into two layers occurred. The lower layer so formed was then drawn off into a second separating funnel. The residue in the second funnel was then shaken with a further 30 ml of diethyl ether for approximately one minute and after separation, the lower layer was discarded. On both occasions, while shaking was being carried out, any vapours produced were allowed to escape by carefully opening the funnel's stopper. Consequently, this part of the procedure was carried out in a fume cupboard.

One hundred millilitres of distilled water were added to the contents of the first separating funnel (i.e. the ether extract) and gentle mixing was accomplished by inverting the stoppered funnel once only. Following this, the water layer was drawn off into the second funnel, but in this latter case, the ether extract and the water were vigorously shaken for one minute. The water layer was then, following separation, discarded.

The ether extract in the first separating funnel was then shaken briefly with 40 ml of the acidified alcoholic wash solution. After separation, this wash solution was then drawn off into the second funnel and the above process was repeated. The wash solution was

finally discarded. Following this procedure, a repeat washing was carried out using 25 ml of the acidified alcoholic wash solution.

The ether extracts in both separating funnels were then combined and 15 ml of petroleum ether was added to decrease the water content. The combined solutions were then given a final wash by shaking with 25 ml of distilled water. The extract was then allowed to stand for 15-20 minutes, after which any further water, settling out was carefully drawn off.

The contents of the one remaining separating funnel were then removed to a round bottomed flask held in a water bath maintained at 60°C and evaporation of the solvent was carried out in a fume cupboard. This process was accelerated by blowing a current of air over the surface of the extract. The air current was produced by a vacuum pump. Eventually a small amount of a viscid, golden yellow residue remained in the flask.

The round bottomed flask, containing the residue, was then cooled in a beaker of cold water, and exactly 5 ml of diethyl ether was added. After the residue had dissolved, 5 ml of saturated sodium chloride solution was added, followed by 10 ml of petroleum ether (accurately measured). After a vigorous shaking, the contents were allowed to stand for several minutes. If by this time, the extract was not absolutely clear, the mixture was again thoroughly shaken.

(d) Analysis

A 10 ml aliquot was transferred to an EEL colorimeter tube and the total carotenoid concentration was measured using Filter No. 440. The colorimeter tube was then replaced in the water bath, and the solvent evaporated using the method already described.

The residue which remained following evaporation was then dissolved, once the colorimeter tube had been cooled, in exactly one ml of chloroform. Following this, a single drop of acetic anhydride was added to the contents of the tube to remove any traces of water.

After the colorimeter had been carefully shielded to prevent contact with the corrosive antimony trichloride solution, the tube containing the vitamin A solution was placed in the colorimeter. Nine millilitres of the antimony trichloride solution were then added as quickly as possible. The initial deflection which occurred immediately reduced slightly, and this latter deflection held steady for several seconds, but then faded. The deflection which held steady for several seconds was noted, and the filter used for the vitamin A determination was No. 620.

Two analyses were carried out simultaneously on each sample of colostrum, and in every case the higher of the two values for both carotene and vitamin A were taken and used in subsequent calculations.

(e) Calculations

The calibration of the colorimeter for the above determinations were based mainly on the methods of Boyer, et al. (1944).

Standard solutions of crystalline beta-carotene (Koch-Light Laboratories Ltd., Colnbrook, Bucks., England) using a 1:10 chloroform/petroleum ether mixture as the solvent were prepared and their respective colorimeter deflections using filter No. 440 were noted. A record of the deflection for each standard solution appears in Table 43.

TABLE 43

The Colorimeter Deflections Associated with
Standard beta-Carotene Solutions

<u>Standard Solution of beta-Carotene</u> (I.U./10 ml. chloroform)	<u>Colorimeter Deflection</u>
200	53.5
160	49.75
120	48.0
100	46.5
80	43.5
60	37.75
50	34.25
40	29.75
30	24.5
20	21.5

From the above values, a straight line graph was constructed on semi-logarithmic paper (see Figure 20). All deflections for the colostrum samples were then read off this graph, and the values so obtained were multiplied by a factor of 7.5 in order that each carotene concentration be expressed as International Units per 100 ml of colostrum.

Standard solutions of vitamin A palmitate in oil (B.D.H. Laboratory Chemical Division, Poole, England) were prepared using a 1:2 ether/petroleum ether mixture as the solvent and their respective colorimeter deflections using the method outlined above (see Analysis) using filter No. 620 were noted. A record of the deflection for each standard solution appears in Table 44.

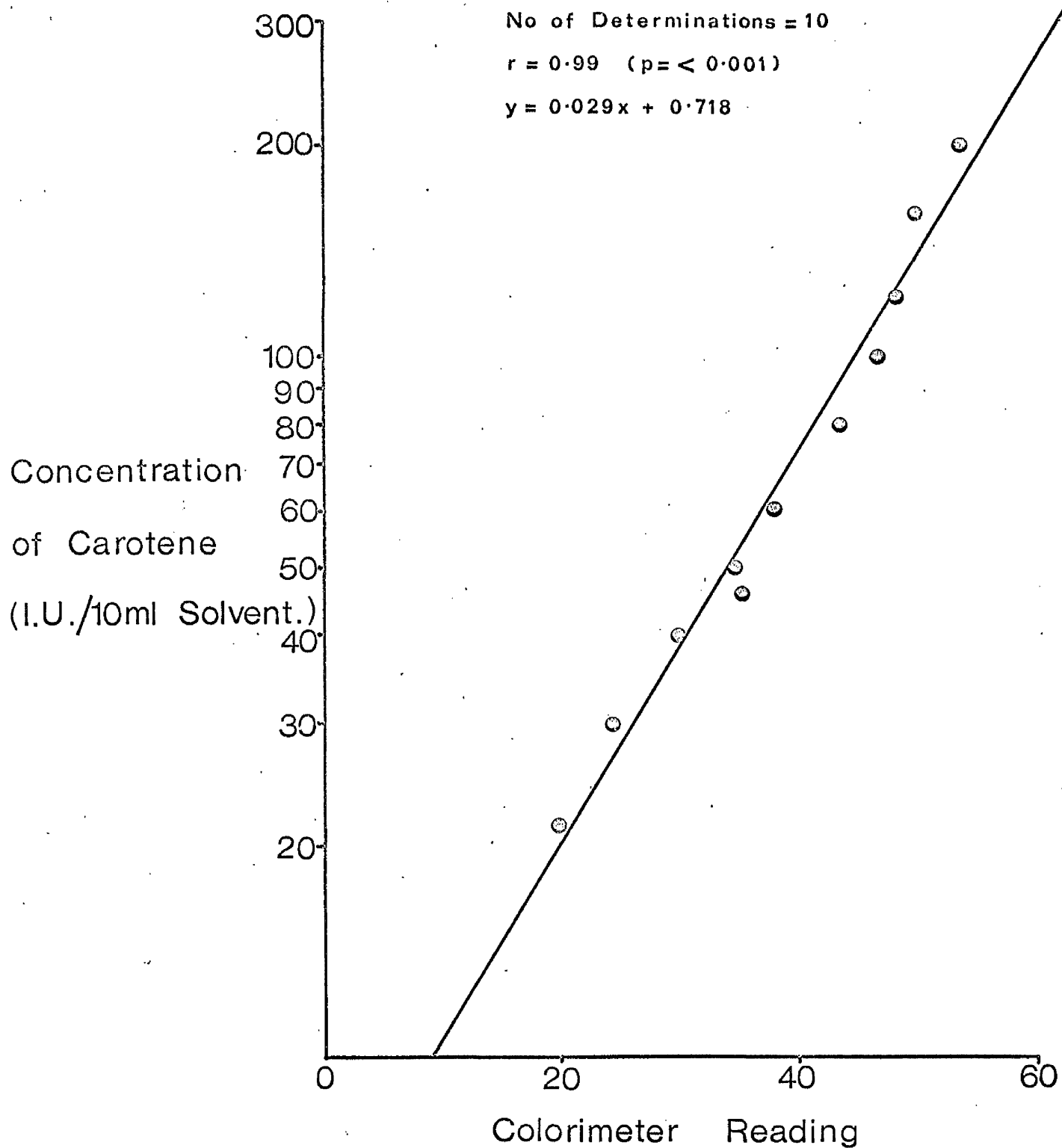


Fig. 20. The calibration of a colorimeter with standard solutions of carotene.

TABLE 14

The Colorimeter Deflections Associated with Standard Vitamin A Solutions

<u>Standard Solution of Vitamin A</u> (i.u./10 ml chloroform)	<u>Colorimeter Deflection</u>
230	98.0
200	95.0
180	91.0
150	86.75
130	82.25
100	77.5
75	64.5
50	49.25
40	41.75
25	28.5

Once again, a straight line graph was constructed on semi-logarithmic paper (see Figure 21). The deflections obtained from the samples of colostrum were then read off this graph. Due to the fact that carotene itself produces a mild Carr-Price reaction, the initial value obtained from the standardisation graph was termed the "apparent vitamin A concentration." A correction for the Carr-Price reaction of carotene had, therefore, to be found and the method of doing this is described below.

In order to estimate the Carr-Price reaction of carotene, standard solutions of crystalline beta-carotene were prepared as above. Ten millilitre aliquots of these solutions were then placed in colorimeter tubes and evaporated using a water bath and an air current until a small drop of residue remained. The residue was then dissolved in 1 ml of chloroform after the colorimeter tubes had been cooled. The tube was then placed in the colorimeter and the procedure for the determination of vitamin A as described above (see Analysis) was then closely followed, and the respective deflections noted. The values

Concentration
of Vitamin A
(I.U./10ml Chloroform.)

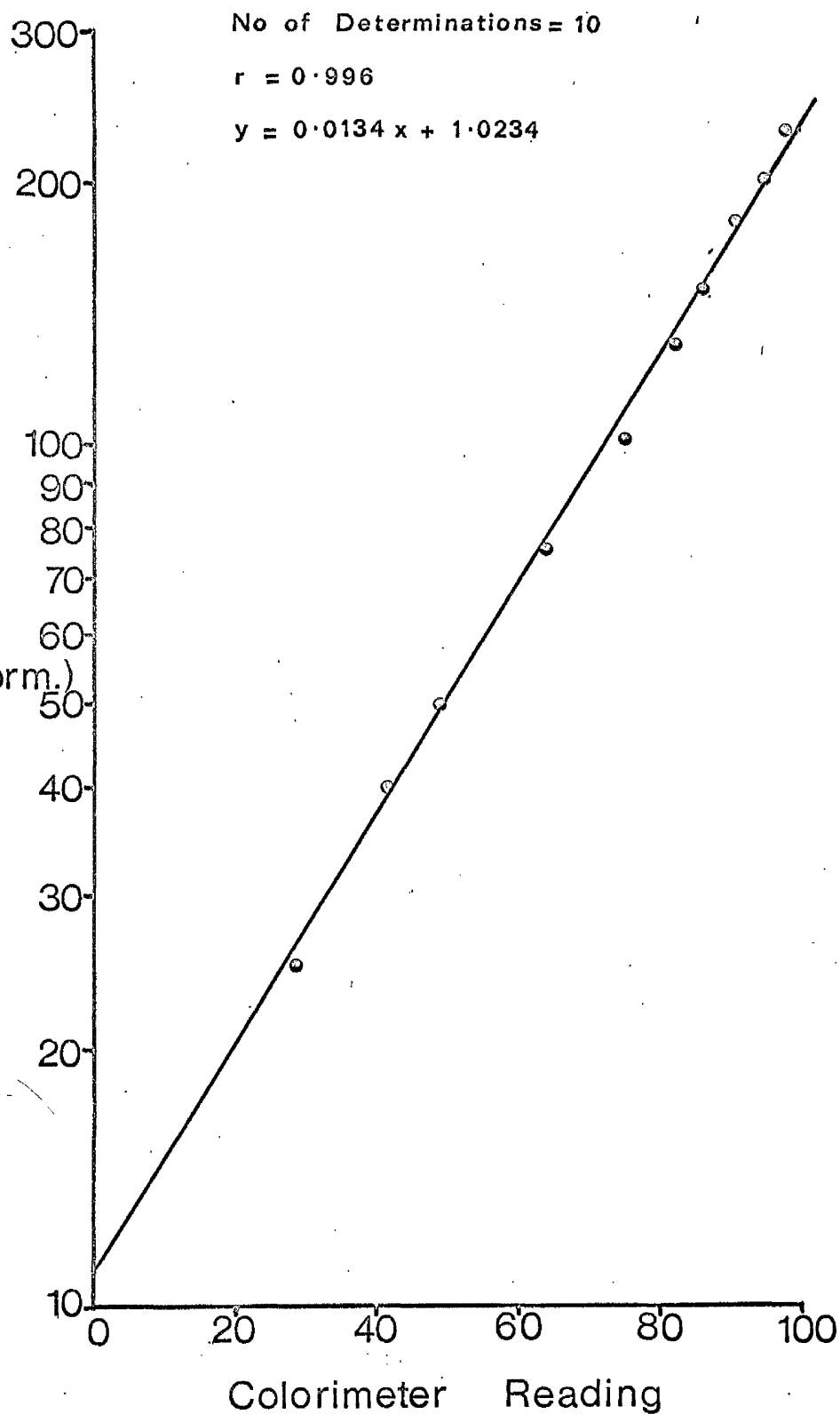


Fig. 21. The calibration of a colorimeter with standard solutions of vitamin A.

obtained are presented in Table 45.

TABLE 45

The Carr-Price Reaction of Standard Carotene Solutions

<u>Standard Solution of</u> <u>beta-Carotene</u> (i.u./10 ml chloroform)	<u>Colorimeter</u> <u>Deflection</u>	<u>*Equivalent Concentration</u> <u>of Vitamin A</u> (<u>*Equivalent Concentration</u> <u>of Vitamin A</u> (i.u./10 ml chloroform))
200	37.0	
160	35.0	31.0
120	24.5	22.5
100	21.5	20.5
80	17.5	18.2
60	14.5	16.5
50	11.0	14.8
40	10.0	14.3
30	7.5	13.25
20	7.0	13.00

* This value was assessed from the Vitamin A standardisation graph already described.

The concentrations of the standard beta-carotene solutions were plotted on plain graph paper against the equivalent vitamin A concentration and a linear relationship was found to exist (see Figure 22). When the carotene concentration of each colostrum sample became known, the Carr-Price reaction of this concentration of carotene and hence its equivalent vitamin A concentration were read off this latter graph. The equivalent vitamin A concentration was then subtracted from the apparent vitamin A concentration, and the result was termed the "corrected vitamin A concentration." Once again, in order to finally express vitamin A concentrations in International Units/100 ml colostrum, the corrected vitamin A concentration was multiplied by a factor of 7.5.

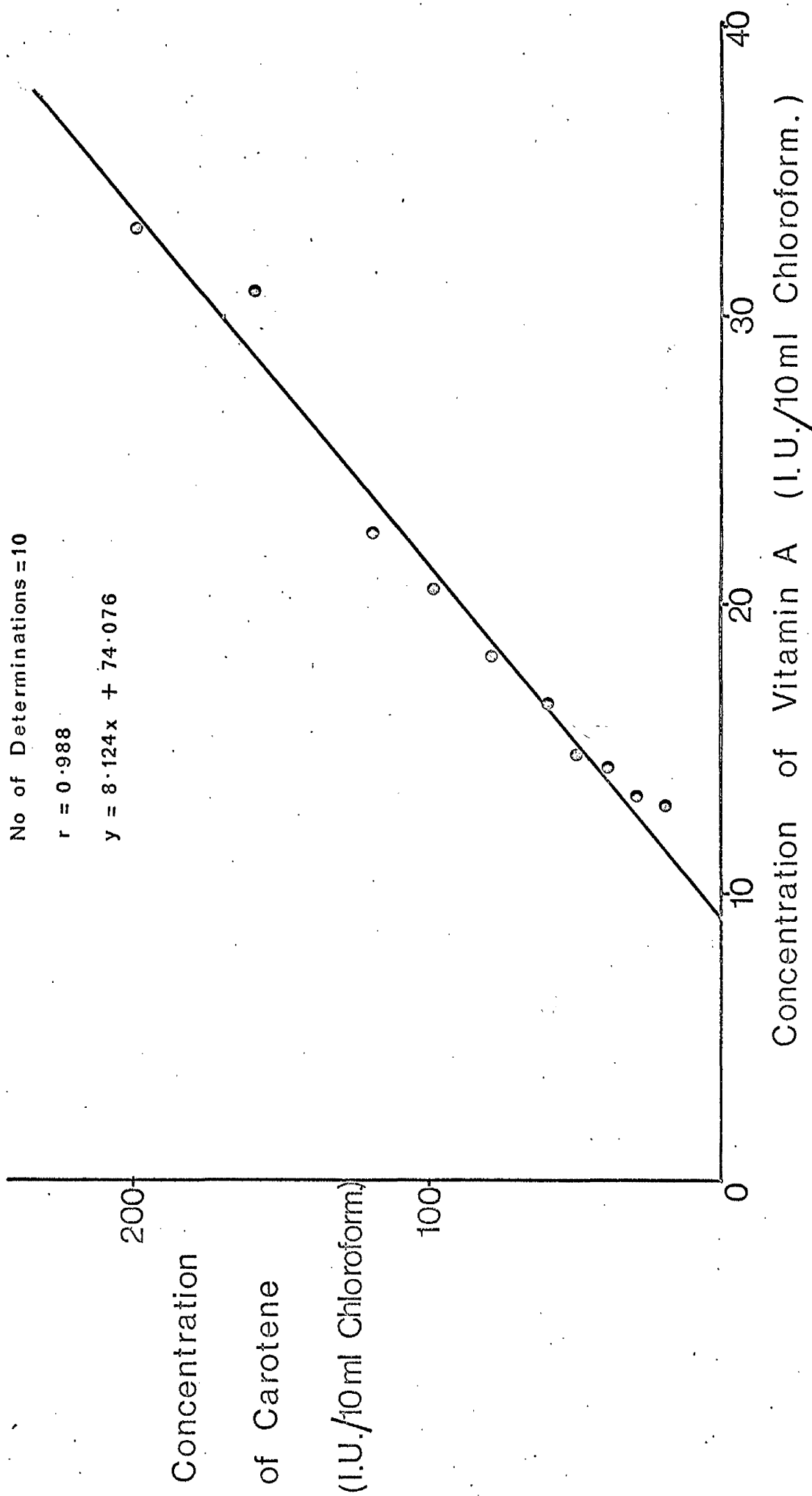


Fig. 22. The correction for the Carr-Price reaction of carotene.

3. Colostrum whey protein determinations

The method of colostrum whey production was as described previously (see Materials and Methods, Part 3). Total protein and immune lactoglobulin determinations were also carried out by the methods outlined earlier (see Materials and Methods, Part 3). Immune lactoglobulin concentrations in colostrum whey were converted to colostrum concentrations using a factor (0.841), the derivation of which has already been described (see Materials and Methods, Part 3).

Section I

The Immune Lactoglobulin Content of Colostrum

The original observation by Gay, et al. (1965) that a seasonal variation in the mean serum immune globulin concentrations occurred in Ayrshire bull calves obtained from markets in the West of Scotland made a study of the immune lactoglobulin concentrations of bovine colostrum imperative. It has already been shown (see Part 3, Section IV) that, under standard conditions, the 48-hour serum concentration of absorbed immune lactoglobulin of a calf was directly related to the immune lactoglobulin concentration of the colostrum that it had been fed during the first few hours of life. Consequently, in addition to studies on the immune lactoglobulin content of different colostrum samples, further studies were obviously indicated to investigate the possibility of a seasonal variation in the colostrum concentrations of this globulin.

Initially, the studies on bovine colostrum were restricted to the samples which were obtained from the dams used in Part 3, Section IV. Later, 100 samples of colostrum collected from the dams of calves used in the previous experiments were used in order to investigate the possibility of a seasonal variation in immune lactoglobulin concentration. The information obtained from the study on the colostrum samples of the 20 dams of the calves used in Part 3, Section IV, is presented in Table 46.

TABLE 46

Studies on the Colostra Produced by 20 Non-Premilked Ayrshire
Cows and Heifers at the First Milking Post Partum

<u>Dam</u>	<u>Volume of Colostrum Produced (litres)</u>	<u>Colostrum Whey Total Protein Concentration (gm/100 ml)</u>	<u>Colostrum Whey Immune Lactoglobulin</u>	
			(a) <u>Percentage</u>	(b) <u>Concentration (gm/100 ml)</u>
Q1	3.5	15.4	71.6	11.02
*Q2	4.5	4.3	52.3	2.25
Q3	16.0	5.7	57.4	3.27
Q4	14.5	5.3	65.1	3.45
Q5	4.25	6.7	57.0	3.82
Q11	13.25	6.7	67.5	4.52
Q12	6.5	9.4	65.6	6.17
Q13	8.0	8.6	60.8	5.23
Q14	5.25	14.6	77.1	11.25
Q15	3.0	13.8	75.7	10.45
Q16	16.0	16.4	75.2	12.34
Q17	3.5	8.0	74.3	5.94
Q18	3.5	12.1	70.7	8.55
Q19	4.5	19.2	68.3	13.12
Q20	6.0	11.3	74.7	8.44
Q21	6.0	10.8	63.2	6.83
Q22	5.0	16.0	74.3	11.89
Q23	4.75	13.4	73.3	9.82
Q24	6.0	6.2	68.2	4.23
*Q25	6.5	12.0	66.8	8.02
Mean	7.0	10.8	67.9	7.53
S.D.	±4.6	± 4.2	± 6.8	±3.33

* Ayrshire heifers

It can be seen from the above results that a wide individual variation was found to exist in the volumes of colostrum present in the bovine udder soon after parturition. Most (80%) of the dams produced between 3.0 and 8.0 litres on being machine milked. However, four Ayrshire cows were found to have produced as much as 13.25-16.0 litres. The two heifers in the series (Q2 and Q25) produced 4.5 and 6.5 litres respectively.

The consistency of the colostrum produced by the dams in the series varied from thin (milk like) to very thick indeed. The colour of the different colostrum samples varied from white to khaki brown.

A very wide individual variation was found to exist in the colostrum whey total protein concentrations of the 20 samples. However, the percentage of the total protein concentration which, on electrophoretic separation, was found to be immune lactoglobulin, remained relatively constant at $67.9 \pm 6.0\%$. Consequently, a similar, wide, individual variation in the colostrum whey immune lactoglobulin concentration was also found in these samples.

The relationship between the total protein and immune lactoglobulin concentrations was investigated, and this is illustrated graphically in Figure 23. It was found that an extremely close linear relationship existed ($r = 0.99$, $p = <0.001$).

It is interesting to note that one of the two Ayrshire heifers in this series (Q2) had, on fractionation, by far the lowest colostrum whey immune lactoglobulin concentration. However, the other heifer (Q25) resembled the other, more mature, dams in the group.

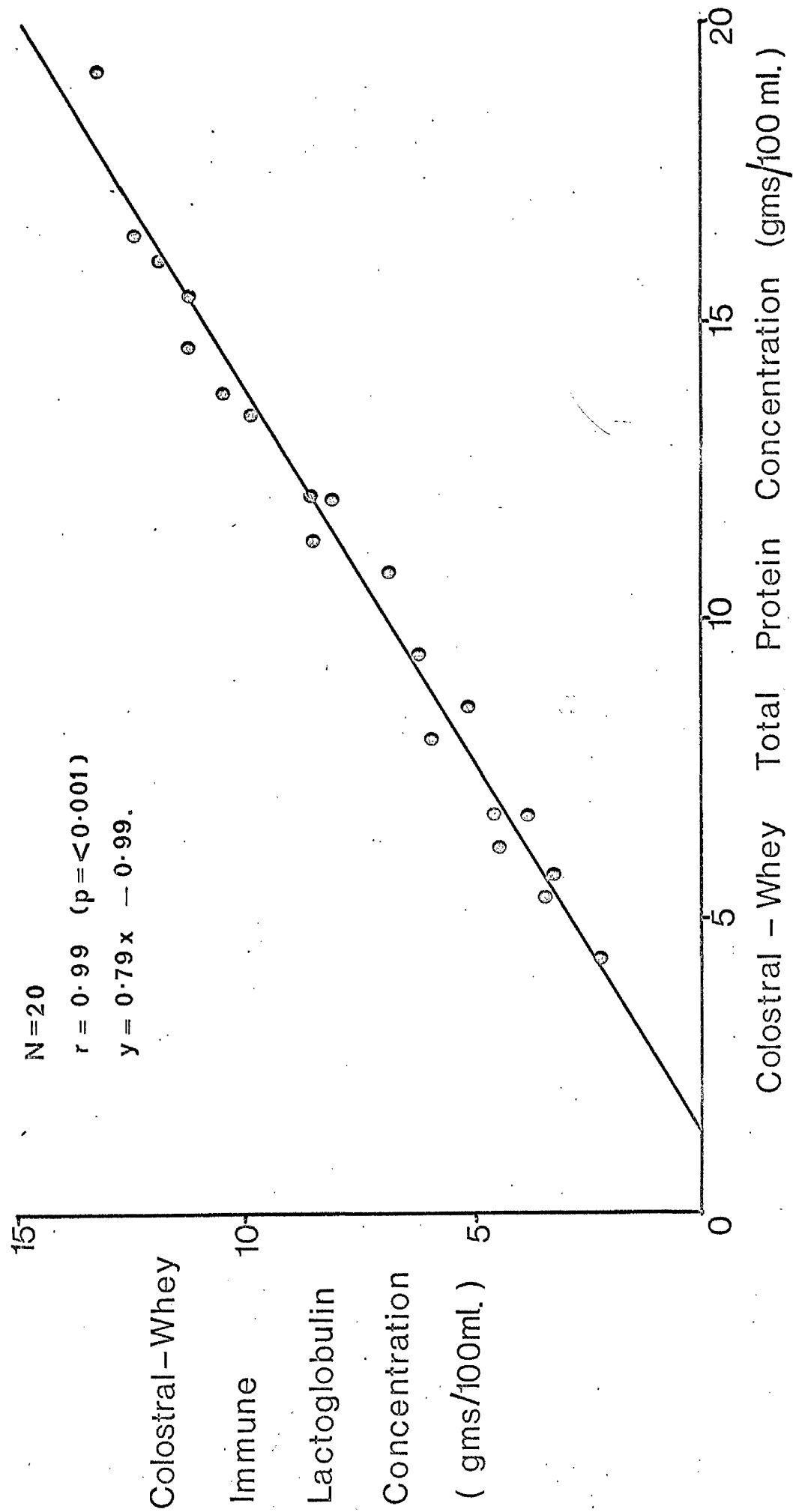


Fig. 23. The relationship between the total protein and the immune lactoglobulin concentrations of wheys prepared from twenty different colostrum samples.

The possibility that a relationship existed between the volume of colostrum present at parturition and its immune lactoglobulin concentration was investigated (see Figure 24). It was found that no correlation existed.

As already mentioned, 100 samples of colostrum obtained from the first milking post partum of Ayrshire cows and heifers were examined. The samples were collected during the months from February, 1968, to November, 1968. The immune lactoglobulin concentration of each sample is recorded individually in Appendix 4, Table 2, but the overall picture obtained is presented in Figure 25.

Once again, an extremely wide individual variation was found to occur, but this was present at all times of the year. No tendency towards a seasonal variation in the immune lactoglobulin concentrations of these samples of colostrum was found to occur.

$N = 20$

$r = -0.27$ (not significant)

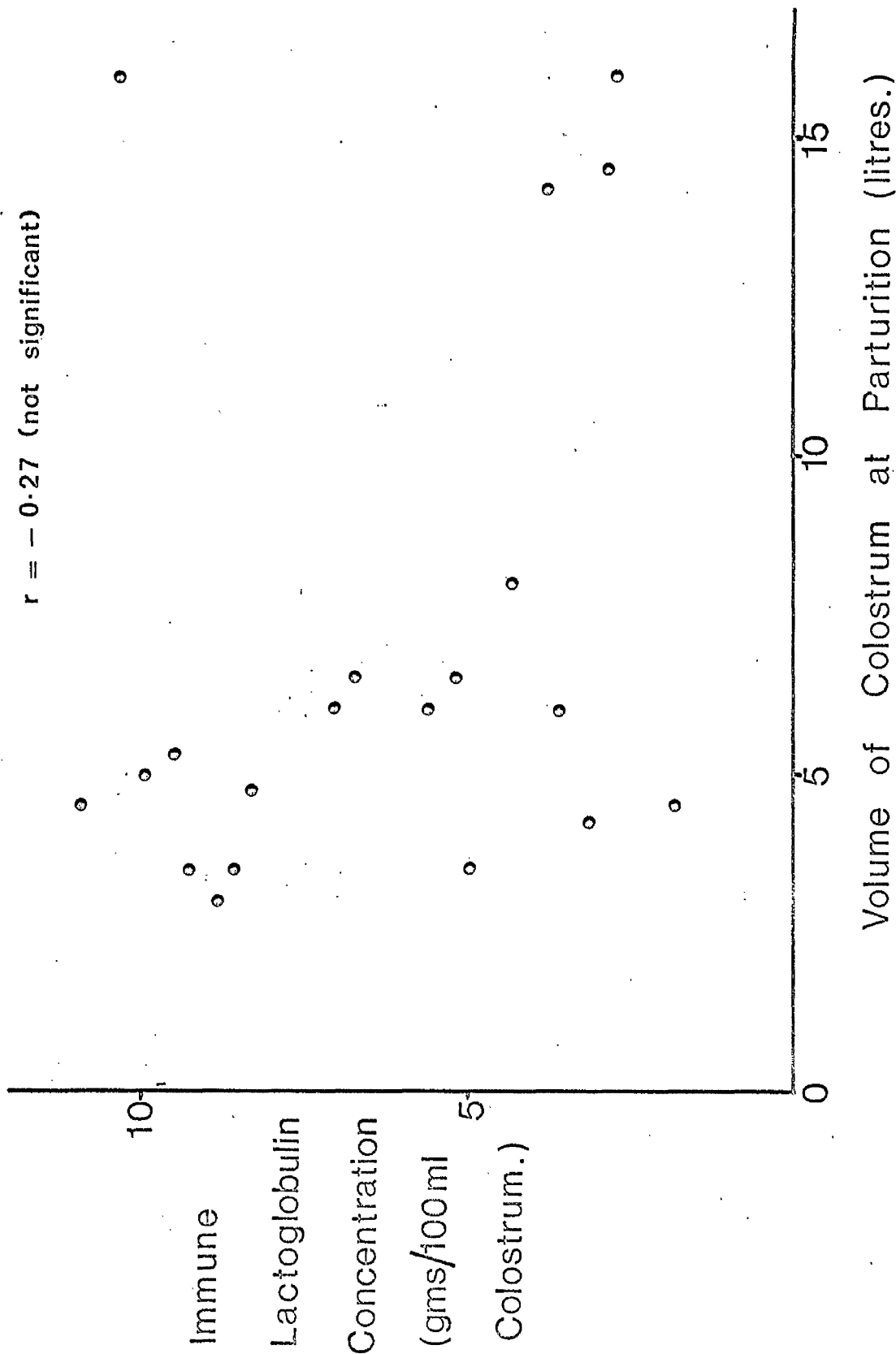


Fig. 24. The lack of correlation between the volumes of colostrum present at parturition in twenty Ayrshire cows and heifers and the immune lactoglobulin concentrations of those colostrum samples.

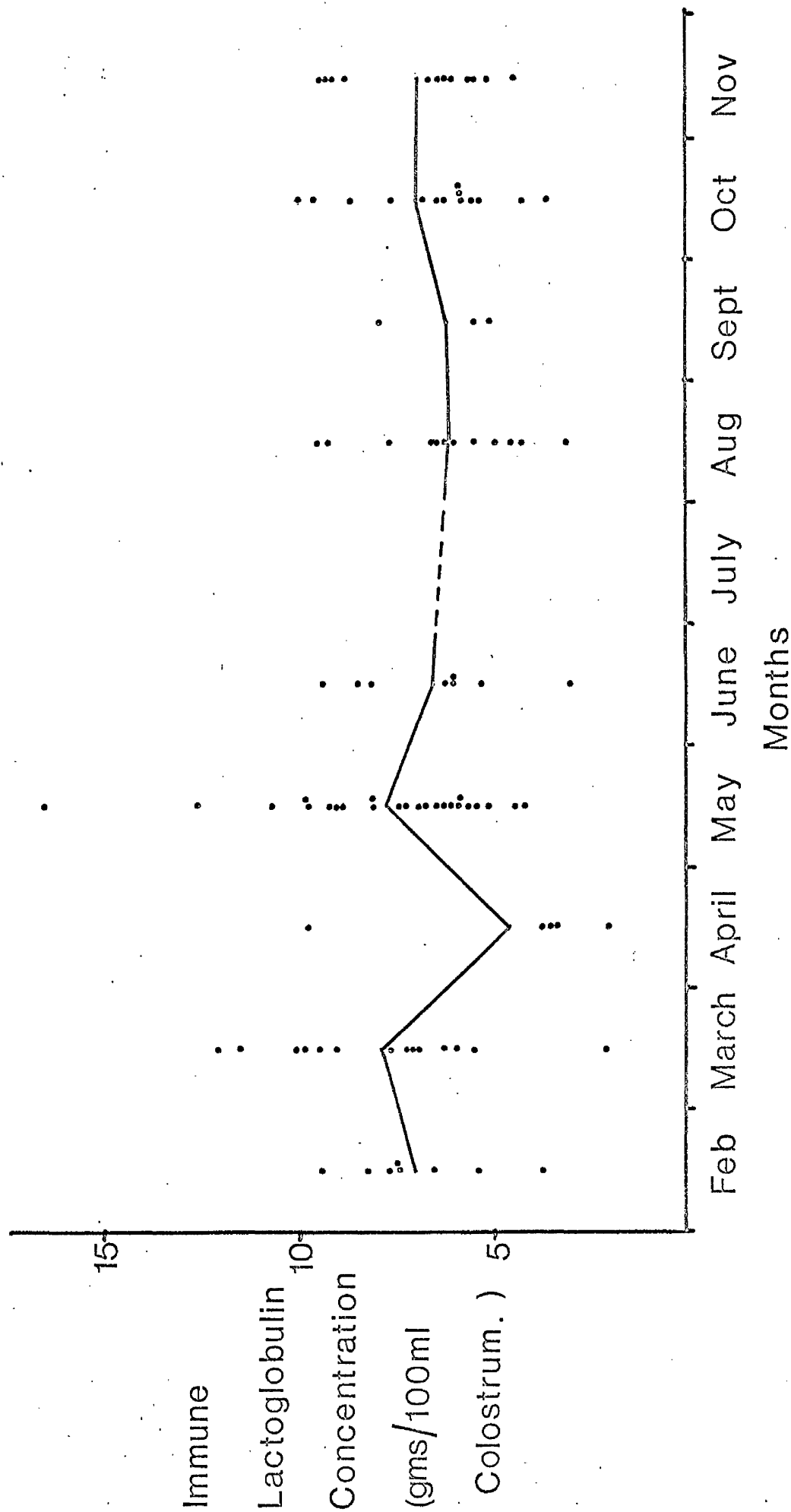


Fig. 25. The wide individual, and the lack of any seasonal, variations in the immune lactoglobulin concentrations of samples of colostrum obtained at parturition from 100 non-prenilked, Ayrshire cows and heifers.

Section II

The Carotene and Vitamin A Content of Bovine Colostrum

The work of Stewart and McCallum (1938a, 1938b) suggested that the vitamin A content of colostrum fed to calves had a very important bearing on the fate of many of those calves. However, most later work has not confirmed that vitamin A plays a very important role in calf survival (Norton, Eaton, Loosli and Spielman, 1946 and Hibbs and Krause, 1947). In view of the fact that vitamin A is the only colostrum constituent apart from immune lactoglobulin that has been linked with calf mortality, it was decided to study the concentrations present in 100 colostrum samples obtained from Ayrshire cows and helpers at the first milking post partum. This was attempted, partly to see whether a seasonal variation existed in colostrum vitamin A levels, and partly whether a correlation existed, as had been suggested by some work on precolostrum (Rowland, *et al.*, 1953), between the vitamin A and the immune lactoglobulin concentrations.

The carotene and vitamin A concentrations of each of 100 colostrum samples are recorded individually in Appendix 4, Table 2. Reference, however, to Figures 26 and 27 show several very interesting features.

First of all, extremely wide individual variations were found to exist in the colostrum concentrations of both carotene and vitamin A. Moreover, these variations were found to be much greater during the summer and early winter months. The minimum colostrum carotene concentrations encountered during the series were <75 International Units/100 ml colostrum and 0 International Units/100 ml colostrum,

respectively. The maximum carotene and vitamin A values attained were 1200 International Units/100 ml and 4888.5 International Units/100 ml colostrum respectively.

The lines in Figures 26 and 27 join the mean monthly carotene and vitamin A colostrum concentrations respectively. It may be seen that a tendency towards a seasonal variation in both constituents exists, with the highest mean concentrations occurring at the end of the grazing season and the lowest occurring during the later winter months. Due partly to the very wide individual variations in both constituents in any one month, and partly to the occasional small number of monthly samples, a statistical comparison of monthly values was not attempted. It was decided that a far better idea of variations due to season would be arrived at if the monthly values were grouped together into four seasons based on feeding practices in the West of Scotland. The results are presented below in Table 47.

TABLE 47

Seasonal Variations in Colostral Carotene Concentrations

<u>Season</u>	<u>Months</u>	<u>No. of Samples</u>	<u>Mean Colostral Carotene Conc. (I.U./100 ml)</u>	<u>S.D.</u>	<u>S.E.</u>
Late winter feeding period	Feb., March April	27	120.3	± 69.4	±13.4
Early grazing period	May	24	405.9	±371.0	±75.7
Late grazing period	June, Aug. Sept.	23	421.7	±283.5	±59.1
Early winter feeding period	Oct., Nov.	26	692.0	±343.8	±67.4

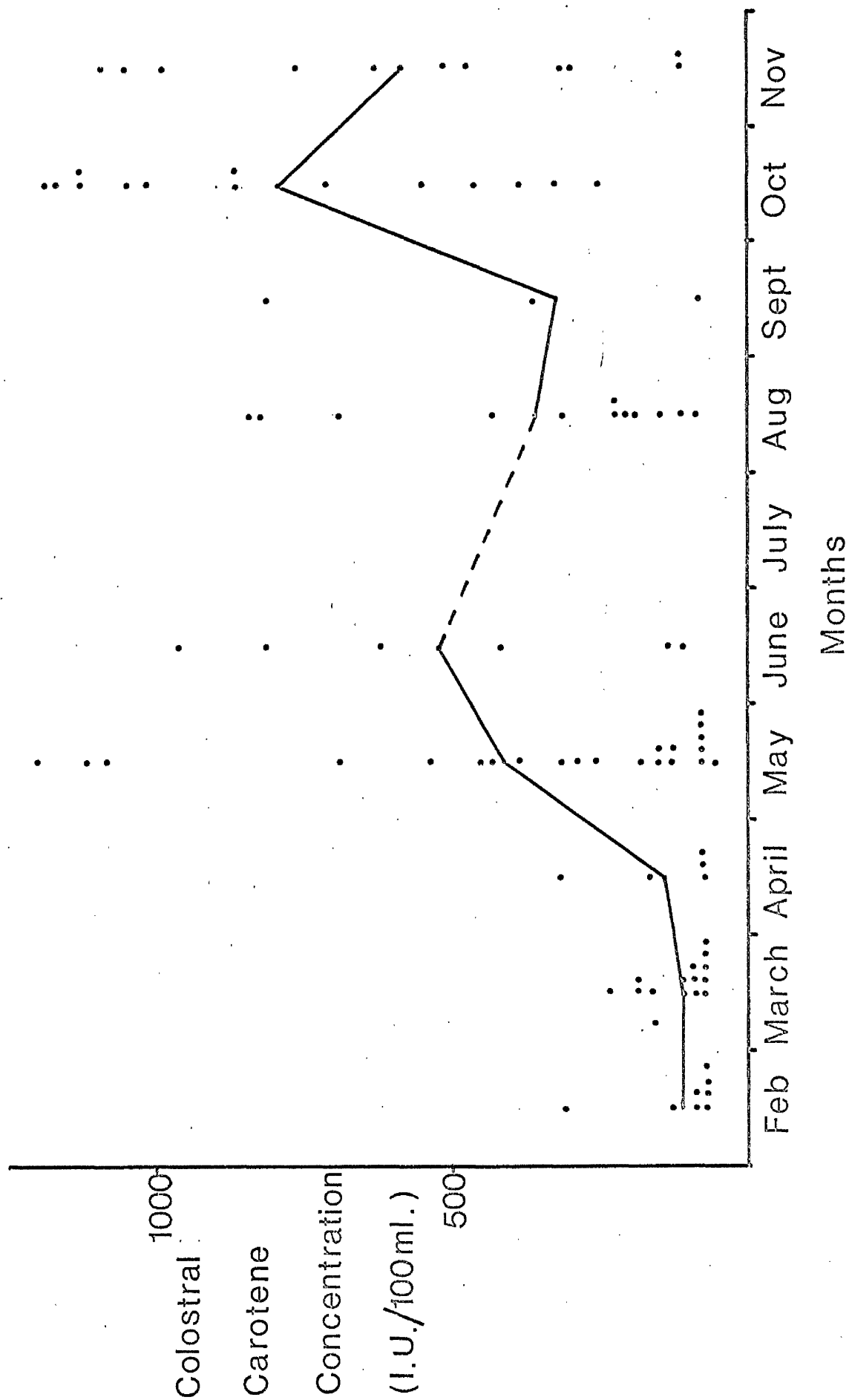


Fig. 26. The wide individual and seasonal variations in the carotene concentrations of samples of colostrum obtained from 100 non-prenilked, Ayrshire cows and heifers.

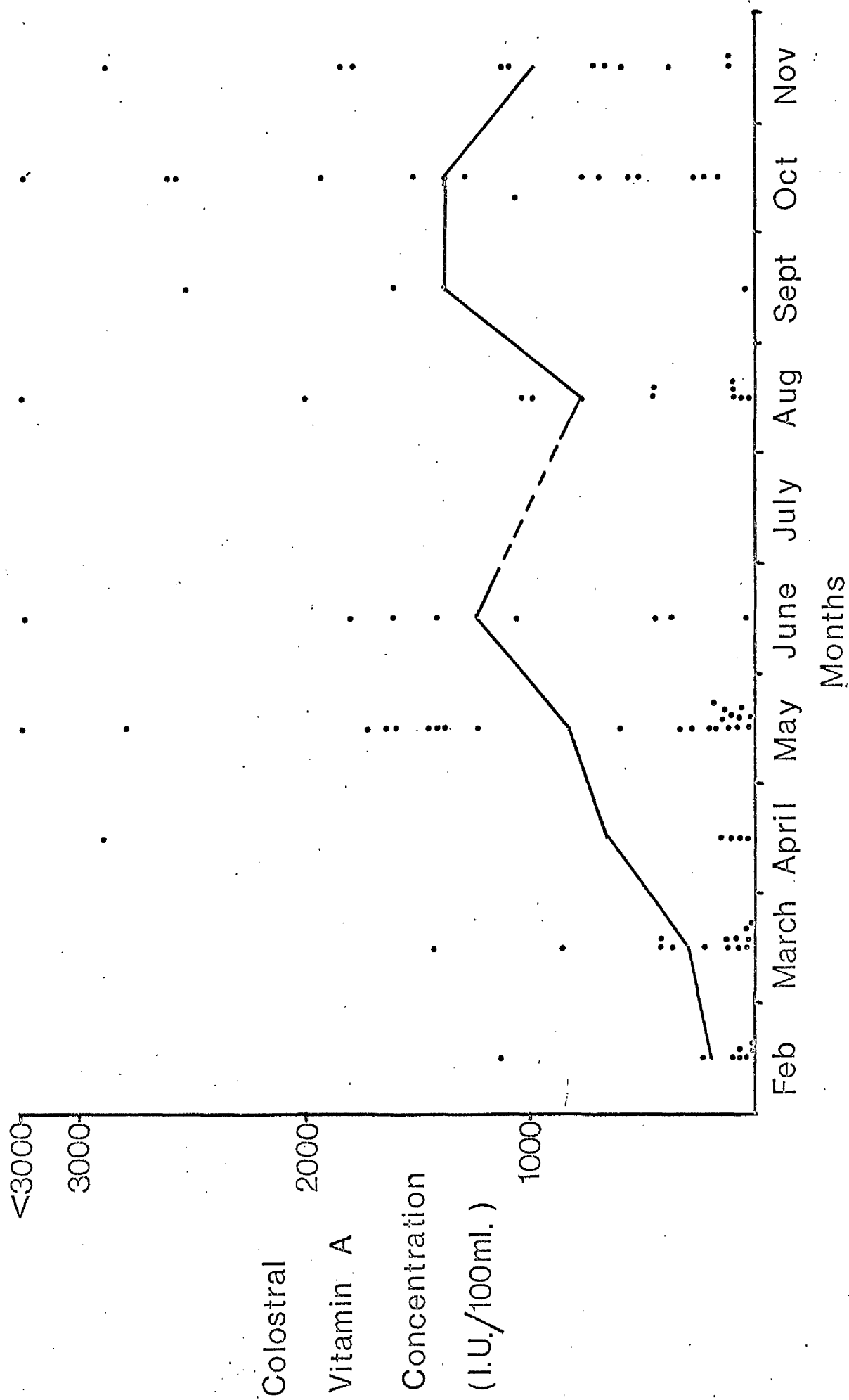


Fig. 27. The wide individual and seasonal variations in the vitamin A concentrations of samples of colostrum obtained at parturition from 100 non-premilked, Ayrshire cows and helpers.

Statistical examination of the values presented in Table 47 reveals the following differences were found to be present.

TABLE 48

An Analysis of the Seasonal Differences of
Mean Colostral Carotene Concentrations

Late winter feeding period	p = <0.001
Early grazing period	p = n.s.
Late grazing period	p = <0.02
Early winter feeding period	p = <0.001
Late winter feeding period	

It can readily be seen that the mean values of colostrum carotene increase through the grazing season to a peak at the end of this season. These values, which are high at the beginning of the winter, then fall markedly so that a highly significant difference exists between early winter and late winter values. The group of dams calving in May and thus probably having had access to pasture for up to four weeks were found to produce colostrum with significantly higher carotene concentrations.

The colostrum vitamin A concentrations were similarly grouped and are presented in Table 49.

TABLE 49

Seasonal Variations in Colostral Vitamin A Concentrations

<u>Season</u>	<u>Months</u>	<u>No. of Samples</u>	<u>Mean Colostral Vitamin A Concn. (I.u./100 ml)</u>	<u>S.D.</u>	<u>S.E.</u>
Late winter feeding period	Feb., March April	27	337.5	± 610	±117.3
Early grazing period	May	24	851.5	± 986.8	±201.4
Late grazing period	June, Aug., Sept.	23	1013.2	±1409.4	±293.6
Early winter feeding period	Oct., Nov.	26	1180.9	±1097.4	±215.2

A statistical analysis of the above results was carried out, and it was found that a highly significant difference ($p = <0.01$) occurred between the mean colostrum vitamin A concentrations of the early winter and late winter fed groups. Also, a significant difference ($p = <0.05$) was found to exist between the late winter fed and the early grazing group. Reference to both Figure 27 and Table 49 indicate that probable seasonal variations occurred at other times of the year. However, the extremely wide individual variations in colostrum vitamin A obviously made a statistical analysis of the difference in mean values for the other seasons impossible.

Finally, an investigation was undertaken to see whether a correlation existed between the immune lactoglobulin and vitamin A concentrations of the colostrum samples. This particular study was limited to those samples obtained from cows and helpers calving during the months of November, February, March, April and May, in order that

a comparison could be made with the results obtained by Stewart and McCallum (1938b). Earlier work established that there was a seasonal variation in the colostrum vitamin A, but not in the colostrum immune lactoglobulin concentration. It was therefore decided to compare each month's values separately, and the results are presented in Figures 28, 29, 30 and 31. A comparison of the values obtained from the colostrum of April-calving cows was not attempted because of the small number of samples obtained in that month. It was found that a correlation did not exist between these two colostrum constituents.

N = 8

r = 0.19 (p = not significant)

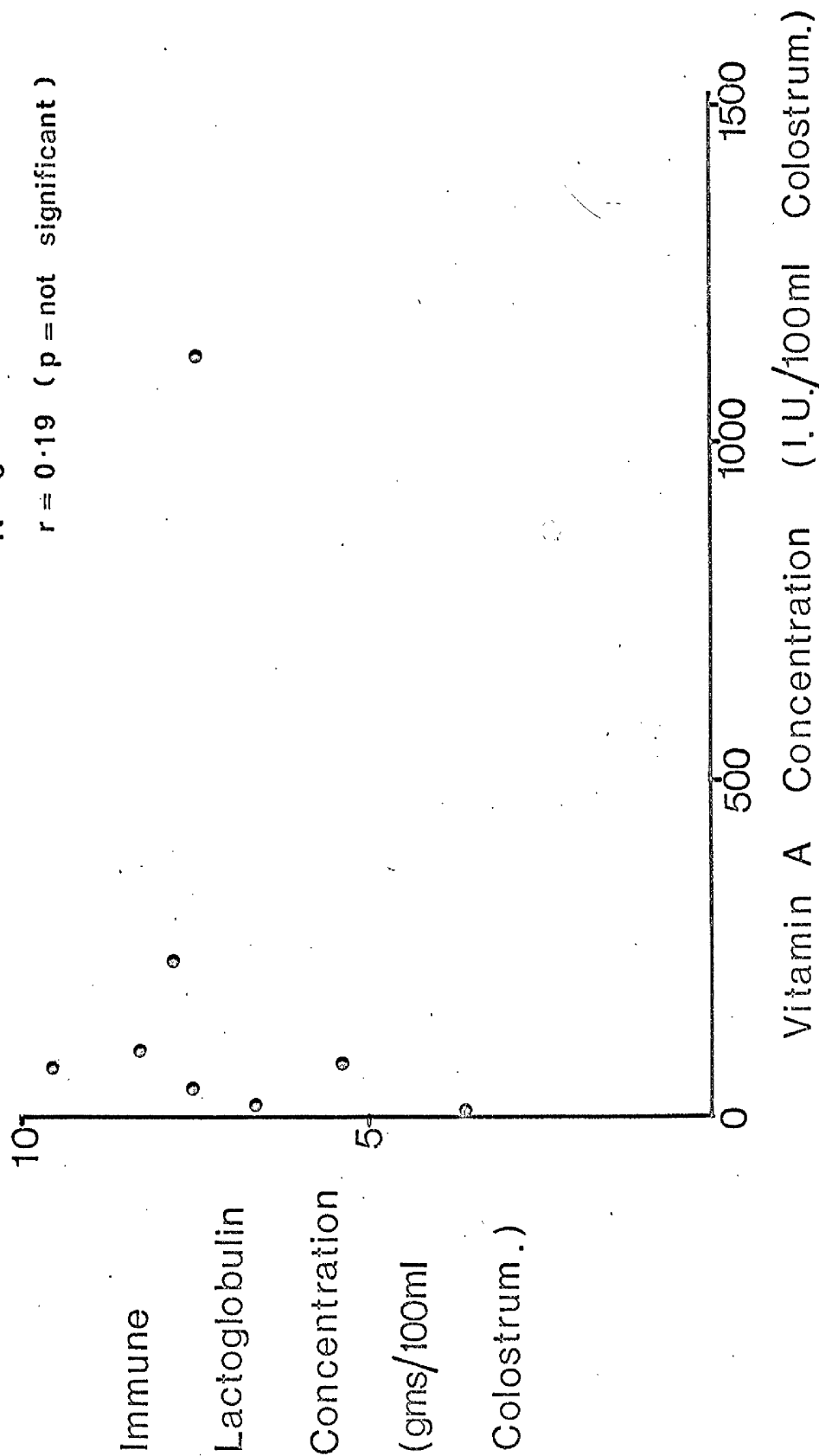


Fig. 28. The lack of correlation between the immune lactoglobulin and the vitamin A concentrations of colostrum samples obtained from eight Ayrshire cows calving in February, 1968.

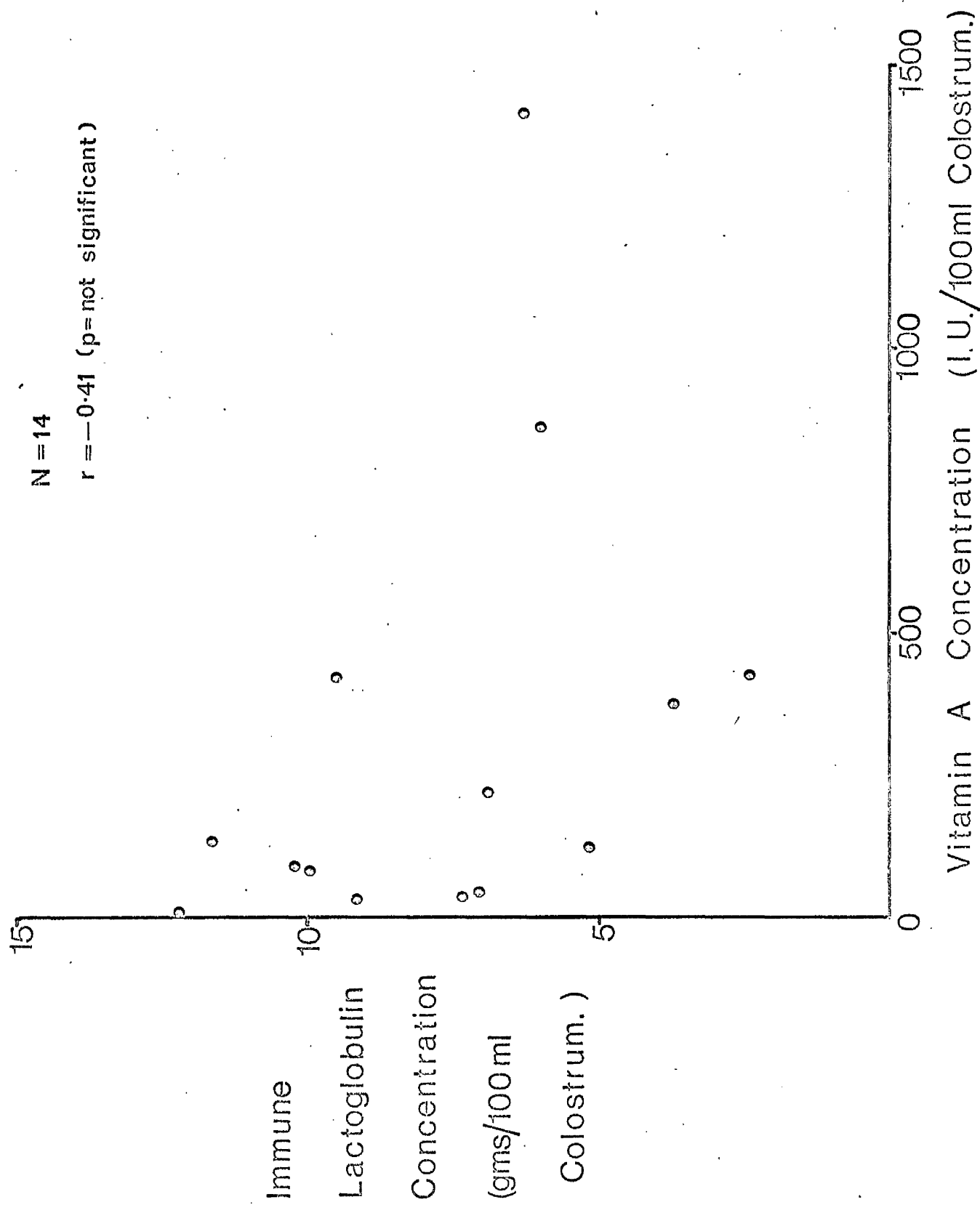


Fig. 29. The lack of correlation between the immune lactoglobulin and vitamin A concentrations of colostrum samples obtained from 14 Ayrshire cows calving in March, 1968.

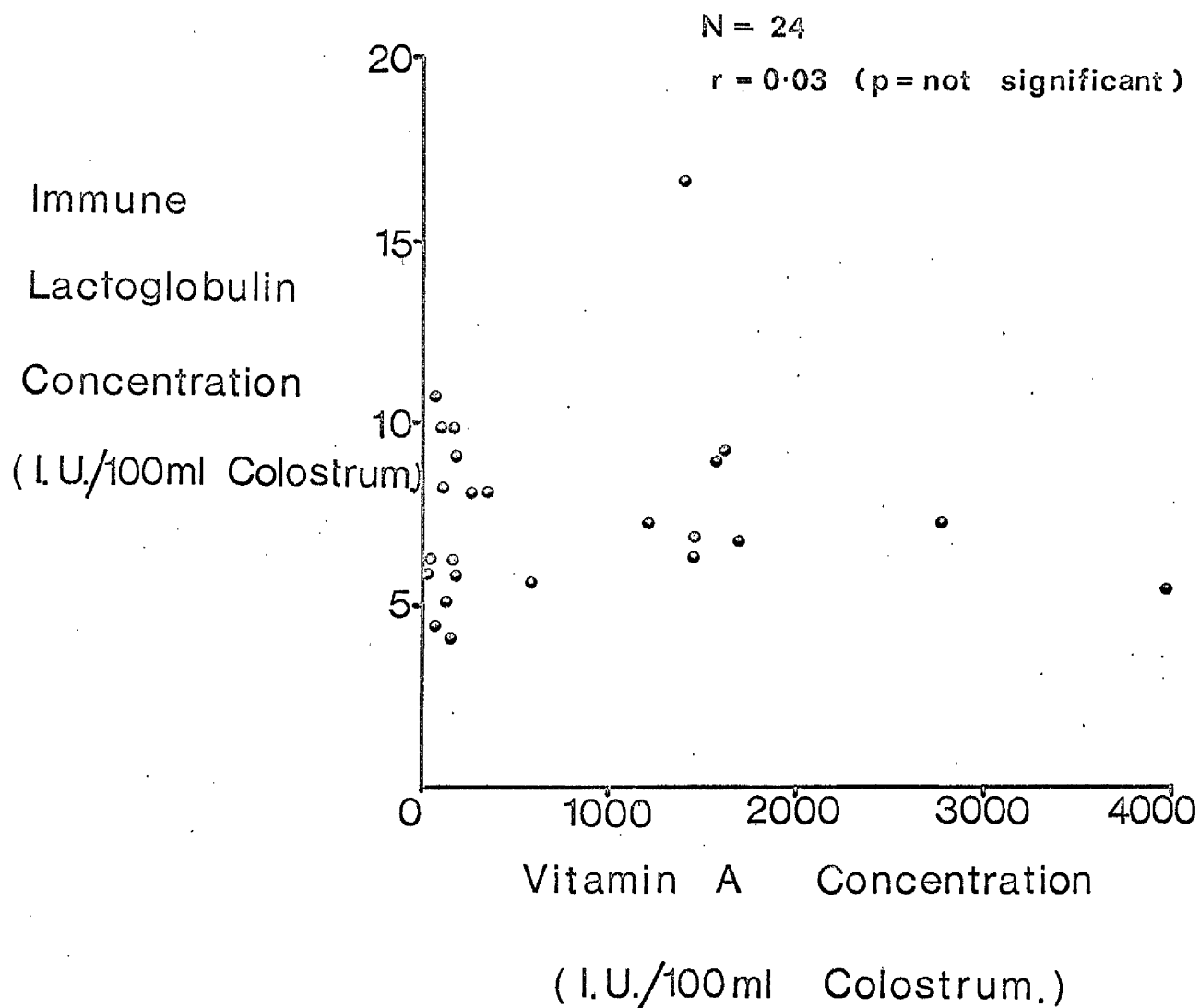


Fig. 30. The lack of correlation between the immune lactoglobulin and vitamin A concentrations of colostrum samples obtained from 24 Ayrshire cows calving in May, 1968.

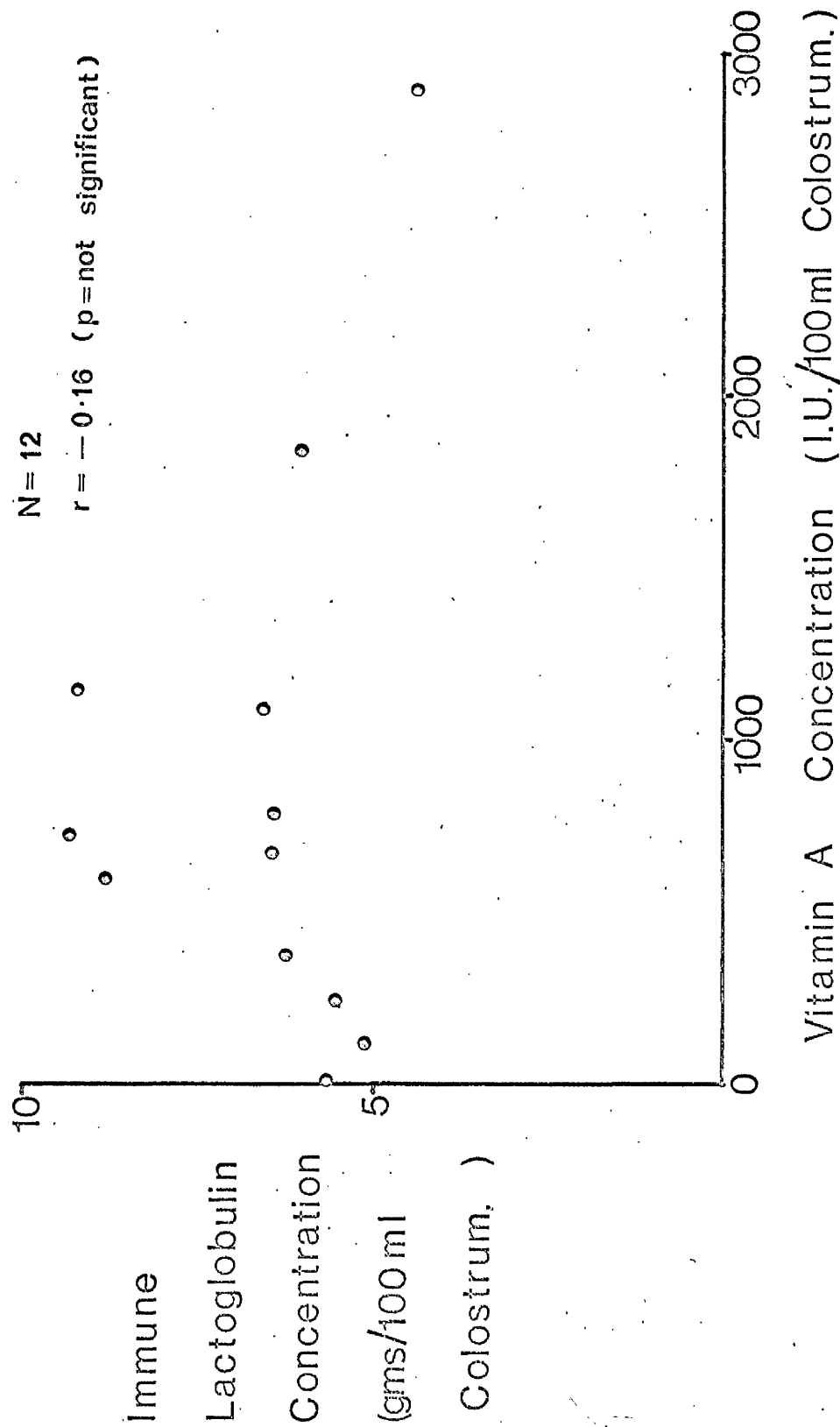


Fig. 31. The lack of correlation between the immune lactoglobulin and vitamin A concentrations of colostrum samples obtained from 12 Ayrshire cows calving in November, 1968.

Discussion

Information regarding the amounts of colostrum ingested by suckling by newborn dairy calves was presented in Part 2, Sections I and II, of this thesis. In that series of 30 calves, it was found that the amount of colostrum ingested during the suckling spell at six hours post partum ranged from 2.5 lbs. to 7.5 lbs. (i.e. approximately 1-3 litres). The present study indicated that the minimum quantity of colostrum obtained when the 20 cows and helpers were machine milked as completely as possible shortly after parturition was three litres. However, the point was also made in Part I, Section II, that the newborn calves did not initially suckle from all four teats, but usually preferred to suckle one teat only, or possibly, two on one side of the dam's udder. It is probable, then, that most calves do not, during the critical first few hours of life, ingest all of the available colostrum from their dams. However, it has been shown by Hansen, *et al.* (1946) that milking a newly calved cow in the presence of its calf produced a greater degree of udder evacuation than did milking while the dam and calf were separated. It may well be, therefore, that during suckling, calves obtain more colostrum from a particular quarter than would be available if that quarter were milked out in the absence of the calf. In any event, it would seem that in the dairy cow and helper at any rate, there is usually a great enough volume of colostrum to satisfy most calves' initial hunger drives.

It has also been shown (Part 3, Section IV) that under standard conditions the immune lactoglobulin concentrations of wheys prepared from different samples of colostrum and fed to newborn calves bore a close linear relationship to the 48-hour serum concentrations of absorbed immune lactoglobulin in those calves. In Part 2, Section II, of this thesis, very wide individual variations were found to occur in the 48-hour serum concentrations of immune globulin of dairy calves suckling at fixed times post partum in spite of high colostrum intakes. It has already been postulated that these variations were due to variations in the immune lactoglobulin concentrations of the different colostrum samples ingested by each calf. The results obtained during the investigations into colostrum composition would tend to support this hypothesis.

Despite wide variations in colostrum whey total protein concentrations, the proportion of this that was found on electrophoretic separation to be immune lactoglobulin was relatively constant. The mean value of $67.9 \pm 6.6\%$ (S.D.) agrees with that of 67.8% found by Pierce (1962) and lies almost exactly midway between the values of 54-55% and 78% suggested by Gay (1965) and Dixon, *et al.* (1961) respectively. The suggestion has already been made that these differences are probably related to the method of colostrum sampling.

A very highly significant relationship was found to exist between the colostrum whey total protein and immune lactoglobulin concentration of a colostrum sample. It would seem that for routine clinical investigational purposes, it is unnecessary to go to the

lengths of electrophoretic separation of colostrum whey proteins in order to measure the immune lactoglobulin concentrations in colostrum. A reasonably accurate assessment of this may be arrived at by measuring the colostrum whey total protein concentration using the Biuret reaction (Weichselbaum, 1946) and then reading the corresponding immune lactoglobulin concentration from a graph such as that presented in Figure 23.

The reasons for the wide individual variations in the immune lactoglobulin content of colostrum obtained under fairly standard conditions are not readily apparent. There is no seasonal variation in the concentration of this particular colostrum constituent so that nutritional factors may be fairly confidently ruled out. All of the colostrum samples were obtained from clinically healthy cows and heifers, and it is well known that the serum gamma globulin concentrations of healthy adult cattle do not vary to anything like the degree seen in the colostrum whey immune lactoglobulin concentrations (Dimopoulos, 1963). Hence, it seems likely that the recorded differences are due to variations in the globulin concentrating process which is carried out by the cells of the mammary gland during the last 30-40 days of gestation (Pierce, 1962) or to variations in the degree of dilution of the globulin secretion of the udder by "milk" (Mill, *et al.*, 1950). Both of these suggestions imply that possibly the number of alveolar cells is reduced either by disease (e.g. subclinical mastitis) or because of lack of proper development of the mammary gland.

It has been claimed that although wide variations were found to exist in the concentration of colostrum globulin at the first milking post partum, the actual weight of globulin produced at this time was

much more constant (Hill, *et al.*, 1950). This was not found to be the case in the present study (see Figure 24). Moreover, when the weights of immune lactoglobulin present at the first milking post partum were estimated, a range of 85.1 gm (Q2) to 1,660.8 gm (Q16) was found. It may be that two or more of the above factors nevertheless combine to produce these variations in immune lactoglobulin concentrations.

No seasonal variation in the colostrum concentrations of immune lactoglobulin was found. This is further proof that the seasonal variation in the mean serum immune globulin levels of calves in the West of Scotland as recorded by Gay, *et al.* (1965b) is purely managerial in origin.

The investigation into the carotene and vitamin A concentrations of 100 colostrum samples revealed very wide individual variations in both constituents, but more especially in the vitamin A concentrations. This was despite the fact that, first, a standard sampling routine was adhered to, and, second, all of the dams were of the Ayrshire breed. The fact that many other workers have noted this wide variation has already been mentioned, and the various suggestions as to the cause of this have already been discussed in the introduction to this part.

In summary, much of the variation in the present study may have been due to the fact that both heifers and cows were used in the series. Moreover, all were obtained from a dealer during the last few days of gestation, and no information regarding the type of feeding and management prior to admission was obtainable. Also, as part of the purpose of this study was to investigate a possible correlation between colostrum vitamin A and immune lactoglobulin concentrations, the

levels were expressed in terms of whole colostrum. Had the concentration of vitamin A been expressed in terms of butter fat, then variations due to the butter fat content of the colostrum samples would have been obviated.

Although the above-mentioned individual variations in carotene and vitamin A concentrations were found to exist at all times of the year, it was obvious (see Figures 26 and 27) that the greatest variation occurred during the summer months. In spite of this wide variation, a definite seasonal variation was found to exist which was most marked when the months were grouped together into four seasons based upon feeding practices in the West of Scotland. A build up in both concentrations occurred during the grazing months to a peak in October and then these concentrations declined through the winter. Access to pasture for the last few weeks of gestation (i.e. in the cows and heifers calving during the month of May) had a marked boosting effect on the mean colostrum carotene and vitamin A concentrations. This last observation concurs with that made by Henry, *et al.* (1940) and Parrish, *et al.* (1949).

It is interesting, in view of the work by Stewart and McCallum (1938b), to note that the lowest mean colostrum vitamin A concentrations recorded occurred during the months of highest calf mortality (Withers, 1952-1953).

Although a positive correlation was found by Rowland, *et al.* (1953) to exist in precolostrum between the vitamin A and immune lactoglobulin concentrations, no such relationship was found in the

present investigation, even when the concentrations of these two constituents were examined on a monthly basis in order to minimise the effect of the seasonal variation in colostrum vitamin A concentrations.

The work by Stewart and McCallum (1938b) is very hard to evaluate since little information is given regarding the animals under test. Nevertheless, it is plain from the results of their studies that calves receiving high concentrations of vitamin A in the colostrum fed to them stood a significantly better chance of survival than those receiving low concentrations. It was claimed that all of the colostrum samples were obtained from Ayrshire cows calving between November and May in well-managed dairy farms in the West of Scotland and also that the feeding practices were similar on all of the farms.

If normal farming practices for the area were followed on the farms in their series, then most of the calves born in May would have been born out of doors, and thus most would have been likely to have acquired reasonably high serum immune globulin levels. The present study has indicated that a seasonal variation in colostrum vitamin A concentrations does occur with low mean levels during the late winter months and higher mean levels in the early winter, spring and summer. It may well be therefore that a proportion of the calves received high levels of serum immune globulin and hence would have been more likely to survive than those calves born during the late winter months. Calves fed low vitamin A colostrum were more likely from the present studies to have been born during the late winter months, and this is just the time when low serum immune globulin levels occur (Gay, *et al.*, 1965b), and calf mortality is at its highest.

Many of the calves receiving high vitamin A colostrum, again because of the seasonal variations in colostrum vitamin A content, are likely to have been born during the early winter months. In general, high mortality rates in calves are not experienced at this time of the year in this area, even though serum immune globulin levels are low (Gay, *et al.*, 1965). It may be that this is due to the fact that the pathogenic challenge at this time of the year is not as great as it is during the late winter months, due to the relatively small number of calves born during the summer in the West of Scotland. A relationship between disease incidence and length of time that a building has housed susceptible calves has been demonstrated by Roy, *et al.* (1955).

To sum up, it has been shown that a seasonal variation in the concentrations of immune lactoglobulin in colostrum does not occur in at least Ayrshire cattle. Wide individual variations do occur in the concentrations of this constituent and the reason or reasons for this are as yet not known. A seasonal variation in the colostrum concentrations of both carotene and vitamin A was demonstrated, but, again, wide individual variations were found. The work purporting to show that vitamin A was the protective factor in colostrum has been questioned. However, it is felt that further work should be carried out on the vitamin A content of colostrum if only to show that the occurrence of lowest mean colostrum vitamin A levels at the time (i.e. late winter) of highest calf mortality is purely coincidental.

GENERAL SUMMARY AND CONCLUSIONS

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Dairy calves left with their dams in a loose box for the first two days of life usually attained high serum concentrations of immune globulin as long as suckling occurred before eight hours post partum. However, further work established that in dairy calves the large amounts of colostrum generally consumed were not necessarily commensurate with the absorption of large amounts of immune lactoglobulin. To produce consistently high serum immune globulin concentrations in newborn dairy calves allowed to suckle to satiation at six and twelve hours post partum, it was essential to leave them with their dams between sucklings. Control calves which were removed from their dams between sucklings at the same times post partum were found to have absorbed significantly less immune lactoglobulin. No significant difference was demonstrable between the colostrum intakes of each group.

The decrease in the absorptive efficiency of newborn calves following separation from their dams was investigated further under very strictly controlled conditions of colostrum feeding and management. Despite the fact that both mothered and non-mothered calves in this experiment were artificially fed (i.e. with a teat bucket), a highly significant difference between the mean 48-hour serum concentrations of absorbed immune lactoglobulin of each group of calves was again demonstrated.

The reason for this decrease in the efficiency of immune lactoglobulin absorption of non-mothered calves is as yet unknown. It may be that the mothered calf has a more efficient oesophageal groove mechanism. Limited evidence from other species, such as rats and lambs, suggests that this may be the case. However, decreased globulin absorption following removal from the dam has also been recorded in very young monogastric animals, such as rats and pups. In these cases it has been postulated that it is the result of increased adrenocortical activity and this, too, may occur in the non-mothered calf.

The exact part played by the dam in producing a calf with a high absorptive efficiency is also not yet known. Although it seems more likely that the grooming activity of the dam is the conditioning factor, the possibility that the mere presence of the dam alone produces a calf with a high absorptive efficiency cannot as yet be discounted. Clearly there is a need for further study into the effects of maternal deprivation on the very young calf.

Initial observations on suckling dairy calves, followed later by more critical experiments using non-mothered calves maintained under controlled conditions of colostrum feeding and management, established beyond doubt that intestinal shutdown in calves is a progressive process initiated at birth. In the suckling dairy calves, a significant negative correlation was found between the time of first suckling and the 48-hour serum immune globulin concentration. The marked variation in the times to first suckling in these calves was the major cause of the wide individual differences between the serum immune globulin concentrations at 48 hours post partum. The importance of factors

delaying the times to first suckling in newborn calves must therefore be stressed, and some of these factors have been defined. In non-mothered calves fed by teat bucket, it was found that intestinal shutdown progressed so rapidly that by ten hours post partum, the ability to absorb immune lactoglobulin had decreased by more than 50%.

Experiments carried out on 120 newborn calves maintained under very strictly controlled conditions of colostrum feeding and management completely failed to demonstrate a single calf with a decreased absorptive ability. Within each group of calves, constant and predictable serum concentrations of absorbed immune lactoglobulin were attained on every occasion.

Due to this highly consistent absorption of immune lactoglobulin by dairy calves under controlled conditions, it was also possible to study the effect of varying certain managerial procedures. It has been suggested that adverse weather may affect the ability of the young lamb and calf to absorb immune lactoglobulin. This work has already been reviewed, and it has been pointed out that the conclusions were not based upon very sound evidence. In the present study, no decrease occurred in the absorptive ability of newborn Ayrshire calves, when these calves were subjected to cold, uncomfortable conditions from birth.

The amounts of immune lactoglobulin absorbed from summer and winter colostrum samples did not differ. The important factor was the colostrum concentration of immune lactoglobulin and the wide variations in this were individual and not seasonal. It seems unlikely, therefore, that under practical conditions, the presence or absence of absorption acceleration factors exert a significant effect upon the rate or extent of globulin absorption.

Finally, the amount of immune lactoglobulin absorbed from a sample of colostrum was not increased by dividing the sample and feeding smaller amounts of this more frequently. Other workers have increased the absorption of antibody by splitting the colostrum to be fed into smaller meals. However, they were offering very small amounts of colostrum to their calves on the assumption that the newborn calf while suckling, repeatedly ingests very small quantities of colostrum. In the course of these present studies, this was found not to be the case. During the first eight hours post partum, the majority of calves suckled only once or twice. The mean colostrum intake of dairy calves allowed to suckle to satiation at six hours post partum was approximately 7% of their birthweight.

The marked seasonal variation in mean serum immune globulin concentrations of newborn Ayrshire calves obtained from markets in the West of Scotland first recorded by Gay, *et al.* (1965b) has been shown in this study to be managerial in origin. The high mean concentrations recorded during the summer months are the result of most calves being born out of doors and hence mothering and, in the majority of cases, suckling occurring without interruption. The traditional method of early calf management in this region involves calving cows in a byre while tied by the neck, removal of calves from their dams as soon as possible after birth, and feeding such calves from a pail after the next milking. Such a routine, from the results outlined above, can produce only a population with a decreased absorptive efficiency. Then these calves are offered usually less than two pints of colostrum for their first meal. The net result of such a routine is to produce calves with low or negligible amounts of circulating immune globulin.

For many years, surveys have emphasised the extremely high calf losses in dairy herds in the West of Scotland. Several workers have suggested that this is due to climatic and managerial factors and the susceptibility of the Ayrshire calf to neonatal disease. However, very few studies have attempted to disassociate these three factors in an effort to indicate the importance of each individually or even to define what constitutes good or bad calf management. Earlier work showed that at the time that losses were at their peak in this region, that is, during the first quarter of each year, the mean serum immune globulin concentrations of newborn calves was extremely low. Moreover, calves with low serum concentrations of immune globulin were highly susceptible to death from either colisepticaemia or neonatal diarrhoea.

The results of the investigations reported in this thesis have defined the reasons for the low serum immune globulin concentrations of winter-born dairy calves in the West of Scotland. It has also been shown that it is possible through modifications of the traditional methods of early calf management in this area to produce high serum concentrations of immune globulin in dairy calves born during the winter months. It is, therefore, suggested that the institution of these modifications on dairy farms during the winter would reduce the high mortality rates occurring at this time of the year.

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APPENDIX I

APPENDIX I

Individual Records of the 30 Dams and Calves Used in the Investigations Into the Behaviour of Cattle During the First Eight Hours Post Partum

BC1: 20.1.67

Dam: Beef Shorthorn; mature adult; quiet. Shape - good.

Calf: Angus X Shorthorn; bull; small and strong.

- 00.25 Calf born. At 00.22 hours, cow was straining in recumbent position and calf was being pushed against manger. As shoulders of calf were passed, the cow leapt to its feet, circled rapidly once, with calf hanging from hips and then, after one powerful strain, the calf was dropped from the standing position. Cow bellowed, leapt at calf, hesitated, and then started to roughly lick it. Rough, continuous licking directed at calf's trunk--no interest in umbilicus. Cow continually grunting at calf.
- 00.29 Calf (lying on back) making first attempts to right its position. Head shaking since birth. Mouth breathing.
- 00.31 Calf into sternal recumbency, head held up. Now breathing through nose. Continually wrinkling nose, sneezing and head shaking.
- 00.36 Struggling to rise (rearing up on to knees). Calf still wet, but most of shiny amniotic liquor has been licked off.
- 00.51 Calf still struggling to rise, frequently makes "praying dog" position, which it may hold for some minutes, but then kicks with hind feet and falls on to nose. Cow still licking very roughly.
- 00.54 Calf standing, very shaky. Immediately shuffled towards cow, but cow still licking it (now concentrating on head) and circles away. Calf persisting and pushing right axilla.
- 00.57 Cow finally stood to allow teat seeking to take place. Calf "pushing" at udder and "butting."
- 01.00 Calf pushing now at axilla for short time, but soon back at udder (left side). Eventually found left ant. (L.A.) teat, but only nibbled side.
- 01.02 Teat seeking broken off. Calf shuffling around box; cow following and licking calf.
- 01.05 Calf teat seeking again (left side). Cow standing quietly. L.A. teat in mouth twice, but did not suckle.

APPENDIX I (continued)

ECI: 20.1.67

- 01.10 Calf suckling L.A. strongly. Immediately, cow started cuddling. Everything very quiet apart from occasional moan from cow and suckling noises from calf.
- 01.24 Calf left L.A. (now no longer erect). Cow still cuds.
- 01.26 Calf slumped down.
- 01.33 Cow eating straw contaminated by uterine fluids, breaking off occasionally to lick calf, especially when calf stirs.
- 01.37 Cow down and cuddling. Calf two feet from cow's left shoulder.
- 01.57 Cow startled, bled twice, then rose. Immediately licking calf and calf (easily) up too. Calf now very strong and gives an occasional bleat and buck. Milk running down nostrils. Mild teat seeking advances to cow, but does not persevere.
- 02.00 Calf down (quite easily this time). Cow licking calf occasionally and still eating contaminated bedding.
- 02.10 Cow down.
- 02.11 Cow up and licking calf (again 2 bleats before rising). Eating straw.
- 02.25 Cow down and cuddling. Calf two feet away from cow's right shoulder.
- 02.55 Cow up, licking calf, eating straw. Calf up, teat seeking.
- 03.00 Calf suckling L.A. teat (empty?) so discontinues after three minutes and teat seeks elsewhere.
- 03.07 Calf suckling right anterior (R.A.) teat.
- 03.10 Off R.A. teat (no longer erect). Calf bucking; cow eating straw.
- 03.19 Calf down (cow licking it and moaning).
- 03.23 Calf up.
- 03.28 Cow now circling to catch end of placenta, now hanging almost to ground. Calf now almost forgotten but for occasional lick in passing.
- 03.31 Calf down, cow down; calf up, cow down.

APPENDIX 1 (continued)

B01: 20.1.67

- 03.34 Cow up. Seems unable to settle because of sight and smell of placenta. Cow up and down several times; cow occasionally eating straw. When cow down never further than six feet away from calf.
- 03.56 Cow down, cuddling, then "sleeping." Calf one foot from cow's left shoulder.
- 05.17 Cow up, still trying to get at placenta.
- 05.28 Calf up, cow licking it and eating straw.
- 05.33 Calf down.
- 05.40 Cow, after several attempts, managed to catch end of placenta in mouth and started eating it. Soon after strained once in micturition position and passed it all.
- 06.35 Has now almost finished eating placenta, but much difficulty encountered. Choked several times, yawned occasionally. Cow down and cuddling. Calf one foot away from cow's right shoulder.
- 07.15 Cow up and eating straw, later clearing up cake.
- 07.30 Calf up, urinated for first time, then down.
- 07.49 Cow licking calf.
- 07.45 Calf up--fairly determined attempts to suckle but occasionally breaking off to run and buck.
- 07.46 Suckling R.A. teat.
- 07.53 Off teat. Bucking then fell down and stayed down.
- 08.00 Calf up, weakly pushing at cow's sides.
- 08.04 Calf down; cow down and cuddling but very alert. Calf one foot from cow's left side.
- 08.19 Cow (alarmed) up, but still cuddling; calf up and urinating.
- 08.25 Cow and calf down. Calf one foot from cow's right shoulder.
- Observation ended.

APPENDIX 1 (continued)

BC2: 27.1.67

- Dam: Beef Shorthorn X Highland; mature adult; nervous. Shape - good.
- Calf: Angus cross; helper; small and strong.
- 16.16 Calf born to hips, cow then rose and calf immediately dropped from the standing position. Cow immediately licking calf, but not so obsessively as Cow 1. Cow licking mainly around back and hindquarters of calf.
- 16.20 Calf mouth breathing still.
- 16.21 Calf nose breathing and snuffling, stirring. Cow consistently bellowing and grunting. Cow already beginning to take interest in contaminated bedding.
- 16.23 Cow left calf altogether to lick and eat straw. However, returns to lick calf whenever it moves.
- 16.24 Calf into sternal recumbency.
- 16.28 Calf attempting to avoid cow's licking. Head shaking, etc.
- 16.31 First attempt by calf to rise, several attempts and everytime calf falls, cow gets excited and licks it, all the time grunting. Calf shivering now, very miserable.
- 16.59 Cow now only interested in contaminated bedding.
- 17.00 Calf still attempting to rise and cow no longer licking it.
- 17.01 Calf up but very unsteady. Cow once again licking it.
- 17.02 Calf slumped down. One almost successful attempt to rise, then up quite easily and walking unsteadily.
- 17.04 Calf starts pushing at inguinal region of cow momentarily.
- 17.06 Fleeting interest in axilla and xiphoid area, then pushed back along body to groin, hindleg and then right past cow. Cow now licking calf's anus and perineum.
- 17.07 Calf down and then immediately up. Raising corner between manger and wall for one or two seconds only. Pushing cow's underside, from axilla to udder.
- 17.09 Calf now mostly pushing at udder. Nibbling L.A. teat once. Cow still licking calf's perineum. Occasionally cow circles hindquarters towards calf with the result that she captures its nose in her inguinal region.

APPENDIX 1 (continued)

BC2: 27.1.67

- 17.14 Calf at axilla again for a moment, then suckling R.A. teat.
- 17.16 Now suckling R.P. teat.
- 17.17 Now trying all teats from right side of cow.
- 17.20 Cow now circling away from calf.
- 17.21 Occasional quick sucks from R.P. teat ($\frac{1}{2}$ minute in all).
- 17.26 Suckling L.P. teat from left.
- 17.29 Off L.P. teat, teat seeking and wandering. Fell.
- 17.32 Calf up (easily), attempting to buck.
- 17.34 Cow down. Calf wandering, milk drooling from lips. Bleating, head shaking and head butting. Occasionally calf runs a few steps. Obviously getting stronger all the time.
- 17.45 Calf down, two feet from cow's right shoulder. Cow lying without cuddling, occasionally moans and smells straw.
- 18.30 Cow up, smelling contaminated straw. Eating small amounts. Much more interested in straw than in its calf at the moment. Calf is very alert. All the time it has lay with its head up and has not slept at all. Cow moaning to itself.
- 18.32 Calf up, stretching. Teat seeking momentarily, then wandering around box.
- 18.45 Cow down.
- 18.50 Cow suddenly up, startled. Calf suckling R.P. teat.
- 18.55 Off teat.
- 18.58 Calf suckling R.P. teat.
- 18.60 Off teat--cow now eating end of placenta until it broke.
- 19.12 Calf down.
- 19.16 Cow down.
- 19.24 Cow cleansed and immediately up, eating it, until
- 19.45 Finished eating cleansing, then cleared up remaining bedding contaminated by discharges. Occasionally licks herself.

APPENDIX I (continued)

BC2: 27.1.67

- 19.59 Cow down, but uneasy. Calf 7 feet from cow's right shoulder.
- 20.17 Cow up and wandering around, occasionally eating a little straw.
- 20.30 Calf up, stretching. Making weak teat seeking advances, but does not really seem too interested in suckling. Cow now licking calf's perineum.
- 20.33 Cow down, calf bucking and wandering around.
- 20.36 Cow up and licking her own hindquarters and escutcheon.
- 20.46 Calf down, cow prowling around.
- 20.55 Cow drinking continually until 21.01. Cow licking calf, calf up.
- 21.04 Cow down on opposite side of box from calf.
- 21.11 Calf down. Cow appears to be very sleepy now and is continually nodding with eyes half closed.
- 22.03 While staring at calf, cow emitted several sharp biales. Suddenly arose and started licking calf and, occasionally, herself.
- 22.15 Cow eating hay and occasionally licking calf.
- 22.40 Calf up; now seems very fresh and alert. Running around box.
- 22.44 Calf went straight to right side of udder and nibbled at R.A. teat.
- 22.45 Suckling R.A. teat but agitatedly bunting udder and searching around--obviously the right quarters (shrunken looking) are empty (actually suckling about $1\frac{1}{2}$ mins.).
- 22.49 Still attempting to suckle right teats.
- 22.51 Calf wandering around; cow mainly interested in eating straw but occasionally licks calf's perineum.
- 23.00 Calf down; cow licking it and eating hay.
- 23.05 Cow down (has still not yet cudded). Calf 8 feet from cow's right shoulder.
- 23.32 Cow takes up "sleeping" position (in sternal recumbency, with head to flank and chin on ground). Shivering occasionally.
- 00.16 Observations ended.

APPENDIX 1 (continued)

BC3: 28.1.67

- Dam: Beef Shorthorn; mature adult; quiet. Shape - good.
- Calf: Angus cross Bull; small and strong.
- 11.01 Calf dropped from standing position. Cow bellowing noisily and frenziedly licking calf.
- 11.02 Calf sitting up in sternal recumbency. Cow, while licking, keeps thrusting its nose into calf's flank (rather like a bitch).
- 11.05 Calf rose into praying dog position, then fell. Cow still very excited and noisy.
- 11.08 Calf sitting like dog, and cow's nuzzling action tends to force calf to rise. Calf up momentarily several times, then fell.
- 11.11 Cow now occasionally taking interest in the contaminated bedding. Whenever calf struggles, though, the cow rushes back to it and starts licking, all the while blaring.
- 11.17 Calf finally up and cow now concentrating on licking its belly and anal area.
- 11.19 Cow now less keen to stay with calf. Calf pushing at cow.
- 11.21 Calf fell and cow got very excited and while blaring, licking phase started again. Calf up again quite easily and teat seeking. Cow too excited to stand now and keeps backing and circling away in order to lick calf.
- 11.23 Calf pushing at right udder but gently kicked by cow. Again cow circling away from calf, licking and occasionally feinting at calf with head.
- 11.26 Calf knocked over by a "feint" but immediately arose.
- 11.28 Cow now quieter and calf teat seeking.
- 11.36 Calf suckling L.A. teat ($\frac{1}{2}$ min.) until cow circled away.
- 11.37 Calf suckling L.A. teat ($\frac{1}{2}$ min.) and L.P. ($\frac{1}{4}$ min.) but cow still fidgetting and calf now getting excited, tail wagging, etc.
- 11.40 Calf pushing left side, gently kicked by cow. Cow eating a little straw.
- 11.42 Bleat from calf--cow very excited again and kicked calf as it pushed at left side.

APPENDIX 1 (continued)

BC3: 20.1.67

- 11.44 Calf suckling R ? teat (out of sight); cow standing quietly and licking calf's perineum (suckling $\frac{1}{2}$ min.). Cow circled away with calf literally chasing its udder.
- 11.49 Cow moaning and eating bedding.
- 11.50 Cow down but calf still wandering around, teat seeking.
- 11.54 Cow up, licking and moaning at calf.
- 12.03 Calf suckling R ? teat (out of sight). Cow much quieter now.
- 12.10 Off teat.
- 12.11 Cow down, calf still exploring, occasionally running and bucking. Cow appears slightly worried by outside noises but still stayed down.
- 12.15 Cow up, calf immediately teat seeking on left side.
- 12.16 Suckling L.A. teat and cow now very quiet and eating hay contentedly.
- 12.21 Calf stopped suckling, changed to right side and immediately started suckling again (which teat not known).
- 12.26 Off teat because cow circled away. Calf still appeared hungry though.
- 12.29 Calf suckling R.P. teat ($\frac{1}{2}$ minute only), then walked under cow. Cow licking calf.
- 12.34 Calf running about, upsetting cow.
- 12.37 Calf suckling R.P. teat.
- 12.38 Off teat because cow kicked calf. Once again, calf running and playing looks very strong and abdomen very distended. Frequently Falls, but immediately rises.
- 13.04 Cow down, licks calf when it passes near her head. Calf now very tired.
- 13.20 Cow up; calf unable to lie down.
- 13.32 Calf urinated.
- 13.35 Cow down. Calf still keeps trying but cannot coordinate legs well enough to do so. Extremely tired now.

APPENDIX I (continued)

BC3: 20.1.67

- 13.55 Finally calf slumps to ground, thus upsetting cow. Cow up and for next 70 minutes both cow and calf go down and rise (latterly calf has no trouble going down).
- 15.05 Cow down, licking calf as it wanders by.
- 15.07 Cow very uneasy, blaring as calf goes near box door. Eventually cow up.
- 15.12 Again cow and calf go down and rise frequently. Occasionally calf tries to suckle but is obviously not too keen to do so. Once again gently kicked by cow. Cow eating dry straw. Whenever cow lies down, it does so near calf.
- 15.55 Cow (while licking her own escutcheon) has seen end of placenta which seems to worry it. Ate a small part of placenta.
- 16.10 Cow down, cuddling quietly now. Once more, cow and calf go through an unsettled phase and continually rise and lie down.
- 17.26 Cow now in sleeping position, occasionally awakens and cuds for a few minutes. Calf sleeping about two feet from cow's right shoulder.
- 18.50 Cow once again sleeping.
- 19.01 Observations ended.

APPENDIX 1 (continued)

BC4: 30.4.67

- 12.30 Calf appeared to want to lie down but (like last calf) seems unable to do so. Cow again drinking for a few seconds.
- 12.31 Cow standing for suckling but calf pushing at right axilla. Occasionally suckling cow's skin (and making sucking noises).
- 12.36 Calf's first head butting (not strong). Consistently pushes too high for teats.
- 12.51 Calf still pushing at cow's side but pushing slightly lower now, occasionally at udder.
- 12.54 For the first time (apart from the short drinks) cow is occasionally leaving calf and taking an interest in contaminated bedding.
- 12.56 Cow down and calf still wandering--cow seems worried about calf.
- 12.58 Calf down right under the cow's chin. Calf lying in left lateral recumbency and shivering violently.
- 13.28 Cow suddenly licking calf again and grunting. Calf up into sternal recumbency.
- 13.29 Calf up, closely followed by cow (grunting).
- 13.44 Cow drinking again. Calf making quite powerful sucking movements into air.
- 13.46 Cow down, calf wandering.
- 13.47 Calf down easily only two feet away from cow. Cow occasionally eating small amounts of contaminated straw.
- 14.17 Both still down.
- 14.40 Cow now in the sleeping position.
- 14.42 Cow suddenly into half lateral recumbency and straining powerfully several times, immediately cleansing. After several minutes, cow still down but making odd licking movements towards her hindquarters.
- 14.45 Cow up.
- 14.46 Cow up and completely cleansed. Immediately exhibited Flehman. Cow not so interested in placenta as previous cows but frequently licks and smells it. Calf seems keen to suckle now.

APPENDIX 1 (continued)

004: 30.4.67

- Dam:** Beef Shorthorn; mature adult; quiet. Shape - very poor; very large pendulous abdomen, pendulous udder.
- Calf:** Ayrshire heifer; average size and not very vigorous.
- 11.40** Calf born (dropped from standing position). Cow immediately whirled and bellowed at calf. Started licking quite roughly. Calf lying in lateral recumbency kicking occasionally. Occasionally cow suddenly stopped its licking and exhibited Flehman.
- 11.45** Calf moved into sternal recumbency. Cow still very interested in calf and continually licking. Then making quieter noises.
- 11.54** Calf now very bright, lying with head up. So far no attempts made to rise.
- 11.55** Calf moved into sitting position on knees. Fell. Cow still licking and bellowing louder again.
- 12.00** Calf struggling to rise but roughly butted and licked by cow. (Very excited.)
- 12.01** Calf again rose into praying position but fell.
- 12.06** Calf up and walking very unsteadily. Cow still licking it about head and both circling (cow away from calf).
- 12.08** Calf pushing at cow and aiming straight at udder (not axilla). Cow licking calf's perineum.
- 12.09** Calf pushing cow's axilla.
- 12.11** Cow again nosily, licking especially at calf's perineum. Occasionally exhibiting "Flehman". Occasionally both cow and calf circle.
- 12.15** Calf pushing at shoulder, triceps, udder, femoral fold, flank and groin--all on the left side of the cow. Calf now appeared to be absolutely dry. Occasionally cow and calf circle again as (i) calf is trying to push at cow and (ii) cow still wants to lick calf's head.
- 12.20** Calf still mainly pushing at cow's shoulder. Cow still only interested in calf and not in contaminated bedding.
- 12.22** Cow took short drink.

APPENDIX I (continued)

BC4: 30.4.67

- 14.49 Cow now eating cleansing and calf pushing at cow's body. Fell on placenta but quickly up. Calf tail wagging violently.
- 15.10 Cow occasionally kicks gently at calf, mainly when calf attempts to suckle its femoral fold.
- 15.14 Calf down. Tends to lie in a more hunched-up position than previous calves (cold?, hunger?), occasionally coughs. Cow still eating placenta.
- 15.37 Cow finished eating placenta.
- 15.41 Cow now grunting and licking calf again.
- 15.43 Cow drinking. Her teats are more erect now than they were at parturition.
- 15.44 Calf up, standing finally in good suckling position, but still concentrating on sucking the inside of cow's femoral fold. Period now of weaker teat seeking advances by calf, mainly directed along cow's underbelly, groin and flanks., but especially anteriorally.
- 15.45 Cow drinking.
- 15.46 Calf down. Seems very tired.
- 15.48 Calf (coughing) up. Teat seeking but still sucking at femoral fold. Cow stands very well for teat seeking advances but occasionally kicks at calf's head.
- 16.00 Cow down.
- 16.02 Calf down, five feet behind cow.
- 16.07 Cow up, eating straw.
- 16.21 Cow down, cuddling for a short while, then very sleepy but in sleeping position for only one or two minutes.
- 18.02 Cow suddenly up, licking calf.
- 18.04 Calf up, straight to left side of cow and finally udder. Sucking the femoral fold. Cow grunting and calf occasionally coughing. Cow still exhibiting flehman occasionally. Calf tail wagging and (weakly) bunting.

APPENDIX 1 (continued)

BCA: 30.4.67

- 18.15 Due presumably to cow's persistence in licking the calf's perineum, the calf is now passing large amounts of meconium. Cow exhibits Flehman again.
- 18.17 Calf wandering but pretty soon takes up teat seeking again. Mainly concentrating on udder and femoral fold now.
- 18.40 Calf down (seems very weak now).
- 18.44 Cow down only one foot from calf. Cuddling.
- 18.54 Calf sucking at wet straw on floor of box.
- 19.12 Cow still cuddling and very sleepy.
- 19.30 Calf now sleeping.
- 19.40 Observations ended.
- N.B. Although this calf did not suckle during the observation period, it was found to be suckling when blood sampling was first carried out at 10 hours post partum.

APPENDIX 1 (continued)

DC5: 2.2.67

- Dam: White Beef Shorthorn cow. Mature adult, quiet. Shape - very good.
- Calf: Angus cross; helper; small and strong.
- 12.14 Calf born to hips with cow in sternal recumbency (quite a difficult birth).
- 12.15 Cow up, blaring and circling. Calf dropped and immediately cow spun around and started licking calf (in left lateral recumbency). Calf head shaking. Cow licking body of calf and occasionally legs.
- 12.16 Calf rose into sternal recumbency, sneezing and head shaking. Cow still circling calf and licking roughly (very attentive).
- 12.20 Calf attempted to rise and several times made a dog-sitting position; whenever calf fell, cow got very excited.
- 12.37 Calf almost stood at one point.
- 12.40 Cow eating straw. Then calf rose and cow got very excited. Blaring, licking, etc. Several times, calf fell and got up.
- 12.47 Calf up again and immediately started pushing at wall and manger. Tail wagging. Cow still very attentive.
- 12.52 Calf pushing initially at cow's brisket, then at udder.
- 12.53 Calf suckling L.A. teat (getting milk). Cow standing quietly, occasionally grunting and licking calf's perineum.
- 12.53 Calf suckling L.P. teat, but cow fidgetting a little, kicking a little. However, calf very persistent.
- 13.07 Calf seemed to have emptied both left quarter, but still trying on left side.
- 13.13 Cow moved away from calf. Calf bucking and running. Twice cow presented right side to calf at this point, and still licking calf occasionally. Calf looked very full.
- 13.20 Calf down and cow down. Over next hour or so both cow and calf got up and down repeatedly. The cow was never at ease and was never down for more than five minutes. Cow and calf always within a few feet of each other when down.

APPENDIX 1 (continued)

BC5: 2.2.67

- 14.36 Cow cleansed, after several strains. Immediately rose and started eating it. Calf up, but seemed very tired. Not teat seeking, just standing or wandering.
- 15.17 Cow finished eating cleansing, but still interested in contaminated bedding. Occasionally licking calf. Calf occasionally coughing.
- 15.38 Cow down, comfortable and cuddling.
- 15.42 Calf down, sleeping.
- 15.49 Cow up, cuddling.
- 15.54 Cow down with calf one foot from cow's right shoulder. Cow cuddling.
- 16.27 Cow up, smelling and licking calf.
- 16.34 Cow drinking for $\frac{1}{2}$ minute, then licking calf again.
- 16.48 Cow down, one foot from calf.
- 16.50 Cow up, smelling contaminated straw.
- 16.55 Cow down, again only one foot from calf. Cow sleeping, calf sleeping.
- 17.10 Cow up, after spell of blaring, then licking calf.
- 17.16 Cow down, one foot from calf, cuddling.
- 18.00 Cow cuddling, calf sleeping.
- 18.13 Cow cuddling, calf sleeping.
- 18.22 Cow sleeping.
- 18.36 Cow up (startled by outside noise). Stumbled twice, blaring, looking at door. Then licking calf.
- 18.37 Calf up, stretching.
- 18.42 Calf suckling L.A. teat. Kicked away once by cow but calf immediately started suckling again.
- 18.49 Again, cow's left quarters appear empty and (for the second time) cow presents right side to calf but calf not interested). Calf running and playing, cow grunting.

APPENDIX 1 (continued)

REG: 2.2.67

- 19.09 Both cow and calf roaming about. Cow licking and grunting.
- 19.06 Cow down, calf wandering near box door so cow up quickly and standing where she could see calf. Cuddling.
- 19.12 Calf standing at right udder but not interested in suckling. Cow standing well though, cuddling. Calf then wandering around box again.
- 19.19 Cow down momentarily but "worried" as calf too far away. Up. Cuddling, but still occasionally licking calf.
- 19.30 Cow down, cuddling. Calf still wandering.
- 19.34 Cow up. Mainly because she was not able easily to see the calf (still wandering around box).
- 19.35 Calf down; cow eating straw.
- 19.55 Cow down and cuddling.
- 20.15 Observation ended.

APPENDIX 1 (continued)

BGG: 9.2.67

Dam: Black Galloway; mature adult, nervous and occasionally aggressive. Shape - poor.

Calf: Hereford cross; helper; small but active calf.

19.45 Calf dropped from standing position after being born to hips from cow in recumbent position. Cow very attentive mother. Blaring and licking. Calf struggling to rise almost immediately.

20.05 After several strong attempts, calf up. Cow backing away, licking its head initially, then settling down a little (initially very excited). Calf not too keen to teat seek.

20.11 Cow down, calf wandering about (not pushing wall).

20.15 Calf down quite easily.

20.22 Calf up, wandering about aimlessly at first, then teat seeking at still recumbent cow. Cow showing very little interest in calf.

20.42 Cow occasionally licking calf.

20.43 Calf down, then immediately up again. Standing at cow's head, being licked.

20.45 Cow up, standing well for teat seeking. Calf at right side of udder, initially teat seeking, then nibbling cow's (very large) teats. Occasionally cow grunted, especially while licking calf's perineum.

20.48 Calf butting and tail wagging. Calf tried unsuccessfully to suckle cow from behind.

20.49 Calf nibbling cow's teats again from left. Occasionally wandering, finally teat seeking at cow's right axilla.

20.56 Cow down, interrupted calf while it was nibbling her teats. Calf wandering and whenever it neared cow's head, she would lick it.

21.01 Calf down, then up again, and running about box.

21.02 Calf down, two feet in front of cow. Cow licking calf, occasionally grunting.

21.12 Calf up, cow up. Cow standing well for suckling, but calf not teat seeking initially. Calf nibbling teat, but not suckling as it was having difficulty getting cow's very large teats into mouth. Cow eating hay.

APPENDIX 1 (continued)

DC6: 9.2.67

- 21.18 Calf gave up, wandering around box for $\frac{1}{2}$ minute. Then teat seeking on left side but nibbling R.A. teat.
- 21.26 Calf pushing along cow's underbelly, then pushing at left axilla.
- 21.28 Calf pushing at right side of udder, but mainly concentrating on left teats.
- 21.38 Calf wandering, then pushing at left udder. Cow eating hay--not at all interested in contaminated bedding.
- 21.40 Cow down.
- 21.44 Calf down (easily) one foot from cow's left shoulder. Cow licking it, then cuddling.
- 22.03 Cow (grunting) up, licking calf.
- 22.05 Cow cleansed very easily, began eating it.
- 22.21 Cow finished eating cleansing, eating a little contaminated bedding, then licking calf. Calf sleeping. Cow grunting.
- 22.26 Calf up, pushing at cow's brisket initially, then nibbling right teats.
- 22.27 Calf pushing right underbelly, then right axilla.
- 22.31 Calf pushing left underbelly, then left axilla. Calf pushing left udder, then wandering. Calf running, fell then rose again, wandering. Cow still eating contaminated bedding.
- 22.45 Calf suckling R.A. teat for 2 minutes.
- 22.49 Calf suckling R.A. teat for 1 minute. Cow eating straw.
- 22.51 Cow down, licking calf, then cuddling. Calf occasionally nosing cow, but mostly wandering around box.
- 22.56 Calf running and bucking a little.
- 23.26 Cow (grunting) up.
- 23.28 Cow down, lying quietly; calf down.
- 00.04 Cow (grunting softly) up.
- 00.05 Calf up, wandering. Not interested in suckling apparently. Running around box.

APPENDIX 1 (continued)

BEG: 9.2.67

- 00.12 Calf standing at left udder.
- 00.13 Cow drinking for nearly one minute. Calf teat seeking, pushing forcefully and tail wagging but cow a bit restive.
- 00.19 Calf suckling L.P. teat for $\frac{1}{2}$ minute, bunting and tail wagging all the time.
- 00.21 Cow down, calf wandering.
- 00.28 Cow up.
- 00.33 Calf pushing at cow's brisket.
- 00.35 Calf down; cow eating but grunting occasionally at calf.
- 00.45 Cow down, lying quietly.
- 01.56 Calf up; cow up, then eating.
- 02.05 Calf down, being licked by cow. Then calf up and down repeatedly for a spell. Cow eating and standing quietly.
- 02.24 Cow urinated, then down right next to calf.
- 02.26 Cow blared loudly twice, then up.
- 02.30 Cow down, again right against calf.
- 03.31 Cow (grunting) up; eating. Calf still sleeping.
- 03.41 Calf up, stretching, then wandering.
- 03.46 Observation ended.

APPENDIX 1 (continued)

BC7: 10.3.67

Dam: Beef Shorthorn. Mature adult, extremely wild and aggressive cow (dangerous). Shape - good.

Calf: Hereford Cross; bull; large, very strong calf.

23.45 Calf born. Some difficulty in passing hips. Calf born from cow in recumbent position and amnion only ruptured at last minute. Cow immediately up, glaring and licking calf. Very gentle with calf. Licking concentrated mainly on back and hindquarters.

00.17 Calf seemed a little dull, and still in lateral recumbency where born. Breathing well but not head shaking, etc. Cow still licking and grunting. Calf then struggled into sternal recumbency.

00.30 Calf attempted to rise. Several attempts.

00.40 Calf up. Immediately shuffled towards cow and started teat seeking. Cow stood very well, grunting and licking calf's head initially then as calf reached udder, its perineum.

00.51 Calf attempting to suckle on cow's right side.

00.55 Calf suckling R.A. teat for one minute.

00.59 Calf suckling LA. teat.

01.00 Calf suckling R.A. teat.

01.04 Calf suckling R.P. teat.

01.05 Calf going from teat to teat on right side. Stopped suckling at 01.08. Calf wandering, looking very full. Cow eating contaminated straw.

01.16 Calf, after several attempts, down and, very soon, sleeping.

01.30 Cow down, two feet away from calf. Occasionally, cow licked calf but mostly, cow just lying quietly.

02.27 Cow up, licking calf and grunting. Soon, though, eating hay for a time then cuddling. Calf sleeping.

02.46 Cow down, cuddling, about eight feet from calf.

APPENDIX 1 (continued)

BC7: 10.3.67

- 02.50 Cow up; eating contaminated bedding (seemed worried by placenta hanging down about four feet now).
- 03.09 Cow down, two feet from calf, cuddling.
- 03.35 Cow up, calf up. Calf running and frisking around box (very lively).
- 03.36 Cow cleansed and immediately started eating it.
- 03.52 Calf definitely suckling on left side but out of sight.
- 03.57 Calf stopped suckling test and suckling at left axilla. Calf then wandering.
- 03.59 Calf again unseen, but suckling on left side for two minutes.
- 03.51 Cow finished eating cleansing and turning to contaminated bedding. Calf wandering and occasionally coughing.
- 04.14 Cow down, calf down.
- 04.16 Cow up, grunting and licking calf.
- 04.26 Cow down, two feet from calf.
- 04.40 Calf up, wandering.
- 04.41 Cow up.
- 04.46 Calf stood idly at cow's left side but not test seeking. After one minute, wandering.
- 05.00 Cow down, cuddling; calf wandering.
- 05.09 Calf down, soon sleeping.
- 05.58 Cow up. First smelling, then eating contaminated bedding.
- 06.07 Cow down, right next to calf.
- 06.08 Cow up, eating hay.
- 06.13 Cow down.
- 06.22 Calf up and urinated. Wandering, then occasionally nuzzled the still recumbent cow.

APPENDIX 1 (continued)

BC7: 10.3.67

06.26 Cow up. Calf running around box. Not keen to suckle.
06.37 Calf down, cow eating.
07.06 Cow down, four feet from calf.
07.45 Observation ended.

APPENDIX 1 (continued)

BCR: 13.3.67

- Dam: Belted Galloway; mature adult, quiet. Shape - poor (large abdomen, pendulous udder).
- Calf: Hereford cross; bull; large and active but slight contraction of foreleg tendons.
- 16.30 Cow eventually calved with little effort. Calf dropped from standing position. Until hips reached, calving had been in recumbent position. Cow fairly quiet but licking calf immediately after calving (concentrating on body of calf). Cow grunting at calf constantly. Calf spent a long time lying in a large pool of amniotic fluid not really attempting to rise, just head shaking occasionally.
- 16.52 Calf into sternal recumbency.
- 17.35 After many unsuccessful attempts over the last 15 minutes (due mainly to calf slipping in amniotic fluid) calf up. Calf may have been tired at this point due to struggling, but from outset appeared rather dull and sluggish. Since calving, cow grunting at calf especially as it teat seeked.
- 17.55 Calf teat seeking, but not too aggressively. Cow standing well and licking calf's perineum.
- 17.56 Calf sucking the side of the R.A. teat for a few seconds, then wandering around box. Later pushing at cow's axilla in a jerky, hesitant sort of way. Then wandering again. Cow now very quiet.
- 18.04 Calf fell and stayed down. Cow standing quietly and eating clean bedding. Frequently grunting at calf. Cow also taking occasional quick drinks. Calf sleeping.
- 18.27 Calf (very easily) up, stretching. Pushing at left side of udder and nibbled teats but did not suckle.
- 18.32 Calf fell but immediately rose and started pushing on left side of udder again. Throughout this teat seeking, spell cow was licking calf's perineum.
- 18.33 Calf pushing at right axilla and brisket.
- 18.34 Calf pushing at right side of udder, then wandering and giving the occasional buck. The calf back pushing at left side of udder.
- 18.43 Calf pushing at left udder.
- 18.45 Cow moved away from calf.
- 18.47 Calf pushing right udder.

APPENDIX 1 (continued)

BCB: 13.3.67

- 18.50 Calf suckling L.P. teat for $\frac{1}{2}$ minute, but cow moved away. Then Calf pushing left udder again for $\frac{1}{2}$ minute.
- 18.53 Calf fell but immediately up again and pushing at left udder. By this point, calf had nibbled and mouthed all four teats, cleaning them but has only suckled for $\frac{1}{2}$ minute.
- 18.58 Calf wandering around box.
- 19.03 Calf sucking skin of right udder, making suckling noises but not on a teat.
- 19.08 Calf suckling L.A. teat for $2\frac{1}{2}$ minutes.
- 19.11 Calf very unsteady and tired, fell but got up again.
- 19.12 Cow moved away from calf to smell contaminated bedding (placenta now hanging to ground). Calf still seemed keen to suckle and was pushing cows underbelly, especially the right axillary area.
- 19.15 Cow eating hay quietly, calf wandering.
- 19.17 Calf down, sleeping.
- 19.36 Cow down, one foot away from calf.
- 19.58 Cow up, eating bedding.
- 20.02 Cow down.
- 20.09 Cow up (very unsettled) moved by placenta and smell of contaminated bedding.
- 20.13 Calf up after cow blared excitedly. Calf wandering. Cow drinking.
- 20.23 Calf stumbled to its knees, fell and stayed down. While up this last time, it showed considerable interest in its dam but only made one short, fruitless attempt to teat seek.
- 20.24 Cow (grunting) down.
- 20.31 Calf (bleating) up. Then cow up (worried by calf). Calf attempting to run around but kept stubling due possibly to its contracted tendons. Fell finally and stayed down.
- 20.40 Calf up, standing quietly.

APPENDIX I (continued)

DC8: 13.3.67

- 20.42 Calf down.
- 20.44 Calf up.
- 20.47 Calf and cow down (calf two feet away from cow). Calf sleeping but cow very restive and for a spell got up and down repeatedly. Not eating, not cuddling.
- 21.28 Cow (blaring) up. Circled to get at placenta, and in doing so trod on calf. Grunting, took occasional lick at calf. Calf up, wandering not teat seeking.
- 21.34 Calf down. Cow beginning to eat end of placenta even though she had not cleansed.
- 21.43 Cow cleansed and immediately set to eating it.
- 21.47 Calf down.
- 22.02 Cow down almost on calf, therefore calf (startled) up. For a time, cow and calf unsettled, and up and down repeatedly.
- 22.16 Calf pushing left udder.
- 22.17 Calf suckling L.A. teat for $3\frac{1}{2}$ minutes, then pushing left axilla.
- 22.24 Calf suckling L.? teat (out of sight) for one minute.
- 22.27 Calf fell and stayed down, passing meconium. Up again and almost immediately started pushing the left udder.
- 22.56 Cow and calf down. Initially cow cuddling, then took up the sleeping position. Both quiet.
- 23.59 Cow (blaring) up, eating hay. Calf up, pushing left udder.
- 00.01 Calf down--seems keen to do so after a little wandering.
- 00.05 Calf up and wandering for a further two minutes.
- 00.18 Cow down, two feet from calf.
- 00.30 Observation ended.

APPENDIX I (continued)

BC9: 16.3.67

Dam: Beef Shorthorn; mature adult. Shape - poor, quite a large, pendulous udder and large teats. Very quiet. Small.

Calf: Shorthorn; heifer; small and strong.

22.18 Calved. Cow bore calf to hips in semi-lateral recumbency, then rose slowly and dropped calf from the standing position. Cow very excited and noisy (loud blaring) licking strongly.

22.20 Calf rose into sternal recumbency, sneezing and head shaking.

22.21 Calf struggling to rise. Cow mainly licking head and neck.

22.22 Calf rose on to knees several times. Several unsuccessful attempts to rise; cow very excited.

22.55 Calf standing. Cow still very attentive. Calf fell after $\frac{1}{2}$ minute.

22.56 Calf up (easily) but immediately knocked down by dam's forceful licking.

23.00 Calf up and moving towards cow. Kept reaching udder but cow (excited) circling away.

23.05 Calf "pushed" on past cow and ended up pushing at wall and manger. Fell and immediately up. Still cow circles away whenever calf reaches udder.

23.08 Calf pushing left axilla and then udder. Cow again circling away. Then again, calf pushing cow's left axilla.

23.12 Calf pushing left udder with cow standing, but then calf fell. up immediately.

23.13 Calf pushing right udder. Cow fidgetting. Calf then wandering away.

23.18 Calf then at right udder but nibbling L.A. teat. Cow standing well and licking calf's anus. Calf having trouble getting teat ends into its mouth.

23.24 Calf suckling R.P., then L.P., then R.P. for a total of $4 \frac{3}{4}$ minutes, all from R. side.

23.33 Calf nosing teats but not suckling.

23.37 Calf wandering and occasionally bucking. Cow grunting.

23.38 Calf pushing left axilla and then left udder.

23.42 Calf (easily) down. Immediately tried to rise, but fell, then stayed down.

APPENDIX I (continued)

BC9: 16.3.67

- 23.48 Calf (looking tired) up and stood quietly. Cow licked it. Calf then suckling R.P., L.A. and R.A. teats for a total of $4\frac{1}{2}$ minutes. (all from right side.)
- 23.55 Calf then wandered off and seemed to have trouble lying down. Finally knocked down by cow (accidentally).
- 00.05 Calf sleeping. Cow worried but cleansing but not eating contaminated straw. Still grunting at calf and licking it.
- 00.15 Calf up. Cow grunting and licking it. Calf not keen to suckle but running about.
- 00.20 Calf attempting to go down but unsuccessful.
- 00.21 Calf finally slumped down but immediately rose as cow licked it.
- 00.23 Calf (very dull and tired) down. Licked by cow, then cow moved away and started eating contaminated straw.
- 00.45 Cow (grunting) down right next to calf.
- 00.47 Cow up, standing quietly.
- 01.06 Cow licking calf and grunting.
- 01.16 Cow down next to calf. Cuddling.
- 01.35 Cow up, cuddling. Calf up, cow very excited.
- 01.36 Calf wandering, then ran about and bucked a little. Finally attempted to go down several times.
- 01.42 Calf down for $\frac{1}{2}$ minute. Cow straining slightly on cleansing. Calf up and down several times--acted very tired. Cow continually licking it and grunting, later eating contaminated bedding.
- 01.48 Cow cleansed, began eating placenta (very excited).
- 02.17 Calf urinated.
- 02.19 Calf down and sleeping.
- 02.58 Cow still eating placenta (having great difficulty).
- 03.18 Cow still eating placenta (having great difficulty).
- 03.52 After finishing all of cleansing and more contaminated bedding, cow down and cuddling.

APPENDIX I (continued)

BC9: 16.3.67

- 03.55 Cow (worried by calf wandering away) up and grunting. For a few seconds, calf pushed at left axilla, then ran around box, bucking and jumping.
- 03.58 Calf pushing left udder for a few seconds then wandered off.
- 04.11 Calf down (easily). Cow drinking. Calf then up. Cow smelling contaminated straw and then showed "Flehman."
- 04.24 Calf down; cow eating dry bedding.
- 04.45 Cow down right next to calf. Calf sleeping.
- 05.00 Cow and calf both sleeping.
- 06.03 Cow up, cuddling quietly, calf still sleeping.
- 06.18 Observation ended.

APPENDIX 1 (continued)

BC10: 6.5.67

Dam: Old Blue-Grey cow, quiet. Shape - very poor indeed. Udder pendulous, teats large and hind teats very nearly touching ground.

Calf: Angus cross; helper==small but strong.

19.20 Calved. Little difficulty. Calf born to hips from recumbent cow, then cow up and calf dropped. Cow very excited, blaring and licking very roughly. Occasionally pushing calf with nose.

19.28 Calf moved into sternal recumbency. Head shaking, snuffling.

19.31 Occasional bleat from calf.

19.36 Calf struggling to rise already; calf falls occasionally but often knocked over by cow's licking (mainly directed at calf's head).

19.45 Calf standing for a few seconds, then knocked over.

19.48 Cow now smelling contaminated straw.

19.55 Calf suddenly rose with very little difficulty. Cow, backing away from calf, and licking its head very roughly.

20.00 " " " " " " " " " " " "
Cow continually blaring. Calf determined to suckle.

20.10 Calf finally reached cow's underbelly. Pushing from axilla to udder and back again on left side. Then moving around cow.

20.17 Calf now spending most of time on cow's right side. Occasionally pushed above udder and suckled femoral fold.

20.25 Calf spending most of time at cow's right side, concentrating on pushing at its xiphoid and axilla. Bunting and tail wagging by calf. Cow standing quietly. For many minutes now, calf (very vigorously) pushing at cow's xiphoid, groin and flank; occasionally calf walks past cow and pushes at wall.

21.05 Calf tired, seemed to want to lie down, but again, making many unsuccessful attempts.

21.07 Slumped down and then rose.

21.11 Calf down. Cow eating contaminated straw. Occasionally calf makes head butting motions from the floor.

21.30 Cow eating hay.

APPENDIX 1 (continued)

BC10: 6.5.62

- 21.55 Calf up (suddenly). Teat seeking as above. Cow eating and standing well.
- 22.27 Calf (tired) down.
- 22.35 Cow down, later cuddling. Lying very close to calf.
- 23.42 Calf suddenly rose, wandering around box. Cow rose when calf neared the box door.
- 23.50 Calf teat seeking but still unsuccessful. Pushing too high and frequently anteriorly.
- 00.20 " " " " " " "
Calf still very keen to suckle. Cow still occasionally grunting and licking calf. Frequently licks calf's perineum as it teat seeks in udder area.
- 00.35 Calf wandering around box, occasionally pushing at wall.
- 00.53 Calf (very tired) down. Cow eating hay.
- 00.55 Calf took up sleeping position, but more curled up than other calves had been. Shivering.
- 01.03 Cow down, four feet from calf.
- 01.07 Cow in sleeping position.
- 01.40 Cow brightened up and started cuddling. Placenta now showing.
- 01.56 Cow suddenly rose and without straining, cleansed.
- 02.05 Cow then started to drink, but after a few moments started showing interest in the placenta, licking and nosing it.
- 02.10 Cow eating cleansing. Calf still has not moved and still shivering.
- 02.35 Cow finished almost all of placenta and a large amount of contaminated straw.
- 02.40 Cow completely finished placenta--down and cuddling.
- 03.26 Observation ended.

APPENDIX 1 (continued)

PH1: 12.2.67

- Dam: Large, white Ayrshire helper. Very good shape for suckling.
- Calf: Ayrshire-cross-Friesian helper. Small, dull. Obvious 3" diameter (approximately) umbilical hernia. Continual tachypnoea but no abnormalities detected on subsequent examination.
- 14.28 Calf dropped from standing position after being delivered to the hips by helper in sternal recumbency. Helper very attentive with gentle but persistent licking. Quietly grunting at calf.
- 14.30 Calf rose into sternal recumbency.
- 14.32 Calf struggling weakly. Helper still nuzzling calf but occasionally would break off when startled (seemed a little nervous).
- 14.33 Calf again struggling to rise. Helper not licking constantly but resumed whenever calf struggled.
- 14.32 Calf struggling, helper still licking.
- 14.40 Helper still licking calf (more frenziedly than at first). Licking calf's hindquarters.
- 14.43 Calf rose into kneeling position but knocked over by helper's licking. Soon up again, then fell.
- 14.47 Calf up and immediately shuffled towards helper. At once helper positioned herself to facilitate suckling by standing right over calf (although calf did not seem to be ready for determined teat seeking).
- 14.48 Helper licking calf's anal area.
- 14.49 Calf started butting helper's abdomen weakly. Pushing and nosing abdominal floor and then at the left side of the udder.
- 14.51 Calf fell after more teat seeking on helper's right side.
- 14.52 Calf's attempts to rise, frequently upset by helper's persistent licking.
- 15.02 Calf kneeling, then up. Immediately walked to the helper's left udder and nosed at teats. Nibbled teats for a few seconds, then moved away. Then repeating process on right side of udder. Helper at one point circled to get closer to calf. Seemed very keen to lick calf's perineal region.

APPENDIX 1 (continued)

DNI: 12.2.67

- 15.06 Calf sucking at right side of udder. Transiently suckled sides of teats.
- 15.10 Calf pushing at underbelly and then right axilla. Heifer still licking calf--especially perineal region.
- 15.15 Calf occasionally wandered off but quickly returned to heifer. Finally pushing right side of heifer's udder.
- 15.17 Heifer again moved hindleg to trap calf's nose in udder area but calf not very persistent and at one point, walked right under cow.
- 15.27 Calf biting side of R.A. teat but not suckling. Again heifer moved to allow calf easy access to udder--this time by abducting hindlegs. Heifer still licking calf.
- 15.36 Calf wandering for a while then pushing at right udder. Teats are not too large and udder seems just at optimum level for calf. However, for minutes on end this dull calf stands just with nose near teats, not really teat seeking.
- 15.39 Calf wandering again.
- 15.42 Calf pushing at right side of udder again. Even when teat seeking, there is no vigorous butting or tail wagging.
- 15.49 Calf wandering and being licked almost continuously by heifer. Heifer still grunting softly at calf.
- 15.5= Calf down fairly easily.
- 16.01 Heifer straining a little and grunting. Occasionally scrapes at bedding with forefoot.
- 16.04 Heifer down. Not relaxed.
- 16.08 Heifer up.
- 16.17 Calf up, nosing udder for a few minutes then wandered off.
- 16.25 Calf down, heifer eating a little clean straw--has never appeared to be interested yet in contaminated bedding.
- 16.29 Heifer down (calf two feet from heifer's shoulder). Licking calf and occasionally eating some clean bedding.
- 16.40 Heifer straining regularly.
- 16.41 Stopped straining and seemed more relaxed.

APPENDIX 1 (continued)

DHI: 12.2.67

- 16.49 Calf up; heifer licking it. Calf then wandered off; this appeared to worry the heifer--blaring.
- 16.51 Heifer up.
- 16.52 Calf teat seeking, finally at right of udder. Heifer licking calf's anus again.
- 16.55 Calf wandered away.
- 16.56 Calf nosing teats in a disinterested sort of way. Not suckling.
- 17.00 Heifer's placenta now reaching the ground.
- 17.06 Calf down. Heifer drinking then licking calf.
- 17.15 Heifer down two feet away from calf.
- 17.20 Heifer rolled into left lateral recumbency with head up, straining violently. Occasionally heifer bled loudly and whole episode suggested that a second calf was about to be born. Calf lying shivering violently.
- 17.39 Heifer stopped straining for a minute, then resumed for a further minute.
- 17.53 Heifer up, stretched and resumed licking calf, grunting occasionally.
- 17.55 Calf up, moved to the right of udder, but again there were no persistent attempts to suckle. Wandered off after a few minutes.
- 18.00 Calf attempted to run.
- 18.05 Calf down (this time with some difficulty). Due to heifer's licking calf almost immediately rose. Heifer (still positioning herself to allow suckling to take place) standing right over calf but to no avail.
- 18.08 Heifer eating contaminated bedding for the first time. Calf wandering around box.
- 18.11 Calf down, immediately followed by heifer (lying one foot away from calf).
- 18.26 Heifer up, licking calf and occasionally eating contaminated straw.
- 18.30 Heifer, still not completely cleansed, began eating end of placenta.
- 18.50 Heifer cleansed. Having great difficulty in eating placenta.

APPENDIX I (continued)

DHI: 12.2.67

- 19.16 Heifer finished eating placenta, then eating contaminated straw.
- 19.18 Calf up; immediately teat seeking in the left udder area. Several times now this has appeared to irritate heifer with the result that she has gently kicked out at calf. This immediately stopped calf's advances. Heifer later cuddling.
- 19.24 Heifer down, calf wandering slowly around heifer. This worried heifer--grunting and attempting to lick calf whenever it was near.
- 19.27 Heifer finally rose and finished last shreds of cleansing.
- 19.31 Calf (weakly) teat seeking again at udder region.
- 19.37 Calf down.
- 19.45 Heifer eating well at this point.
- 19.50 Heifer down, three feet from calf. Heifer at "sleepy" stage.
- 20.45 Heifer up, eating bedding. Occasionally would grunt at calf.
- 21.00 Heifer down, two feet from calf.
- 21.05 Heifer up.
- 21.10 Calf up. Teat seeking on right side of udder. Once or twice butted udder for the first time. This upset heifer and she moved away and kicked gently at calf. Immediately calf stopped its teat seeking.
- 21.16 Calf down, being licked by heifer. Calf then up and down several times.
- 21.19 Calf teat seeking but heifer unsettled and would not allow calf to nibble teats. Calf wandered away.
- 21.29 Calf down. Heifer eating clean bedding.
- 21.39 Heifer down, three feet from calf.
- 21.47 Heifer up, eating again.
- 21.55 Heifer down, right next to calf, sleeping. But soon uneasy and seemed to want to lie in lateral recumbency.
- 22.02 Heifer up.
- 22.10 Calf up, nosing right of udder very gently. Very weak attempts to suckle at this point.
- 22.18 Calf down, cow eating.

APPENDIX 1 (continued)

DM2: 12-2-67

Dam: Small Ayrshire heifer. Shape = poor, as fairly large abdomen, large udder and very short legs.

Calf: Ayrshire heifer, strong.

19.47 After bearing the calf to its hips while down, the heifer rose and the calf immediately was dropped from the standing position. Calf immediately started to bleat.

19.50 Heifer licking calf's head (calf still in lateral recumbency).

19.51 Calf rose into sternal recumbency. Heifer gently licking calf.

19.59 Cow at this point started breaking off to smell, lick and then eat contaminated bedding. Not so keen to lick calf.

20.03 Calf trying to rise, heifer gently licking it again.

20.17 Calf up after several powerful attempts. Calf pushing at heifer's right axilla.

20.20 Calf walking around box, occasionally running.

20.22 Calf fell and stayed down. Heifer still gently licking calf, occasionally still eating bedding.

20.40 Calf up. Heifer keen to lick the umbilicus and at one point, ran it through her mouth. Calf wandering around box. Occasionally pushing at the heifer's left axilla.

20.57 Heifer standing very well for suckling. Calf pushing udder occasionally, but teat sucking mainly concentrated on the axillary regions.

20.56 Calf suckling L.A. teat for $\frac{1}{2}$ minute, then bucking strongly.

20.57 Calf very strong and keen to suck but still down to the axillae.

21.01 Calf suckling L.A. teat for 10 seconds only.

21.04 Calf bleated whenever heifer licked perineum.

21.05 Calf sucked L.A. teat for another 10 second spell.

APPENDIX I (continued)

DH2: 12.2.67

- 21.10 Calf suckling strongly at L.A. and L.P. teats for 13 minutes, with frequent interruptions and much stamping, butting and tail wagging.
- 21.26 Calf stopped suckling and wandered away from helper, occasionally bucking. Helper still licked calf whenever it came close.
- 21.29 Calf fell but arose very quickly. Cow drinking.
- 21.43 Calf suckling L.A. and L.P. teats for five minutes continuously.
- 21.51 Calf fell and stayed down. Helper still licking calf.
- 21.53 Helper down, very gently, adjacent to calf.
- 21.57 Helper startled; rose and started licking calf.
- 22.02 Helper eating contaminated straw.
- 22.11 Helper down right next to calf.
- 22.14 Helper up, uneasy. Nosing at straw and eating a little.
- 22.46 Helper and calf up. Calf wandered off and started bucking and running--not keen to suckle.
- 23.01 Calf suckling L.A. and L.P. teats for 7 minutes, then wandered away from helper and lay down.
- 23.12 Helper down adjacent to calf.
- 23.15 Helper up and immediately cleansed. Started eating placenta.
- 23.28 Calf bleated loudly then arose, wandered around.
- 23.32 Calf down.
- 23.39 Calf up, stretched, then started circling helper. Calf frequently ran around, bucked and shook its head.
- 23.42 Calf down.
- 23.50 Helper finished eating placenta and soon lay down, one foot from calf. Helper soon sleeping.
- 00.03 Calf and helper both up. Helper eating hay and occasionally licking calf. Calf wandering around box.

APPENDIX I (continued)

DH2: 12.2.67

- 00.38 Cow drinking.
- 00.40 Calf down (there was never any difficulty experienced in lying down with this calf).
- 00.54 Heifer down, three feet from calf.
- 01.18 Calf up, pushing at heifer's neck and then running and bucking.
- 01.50 Calf down, sleeping.
- 02.03 Calf and heifer both up. Heifer eating, calf initially wandering but later stood still and strained persistently to pass small amount of meconium.
- 02.17 Heifer still eating hay, calf wandering.
- 02.18 Calf down.
- 02.42 Heifer down and cuddling quietly.
- 03.22 Heifer took up the sleeping position.
- 03.30 Calf up and wandering around (very bright). Heifer grunting quietly at calf.
- 03.36 Calf down.
- 03.37 Calf and heifer up.
- 03.47 Calf making teat seeking advances again on left side.
- Off watch.

APPENDIX 1 (continued)

DH3: 14.2.67

- Dam: Small, Ayrshire heifer, aged approximately two years. Nervous when approached but quiet in box. Shape good. Relatively large teats and udder but well tucked up.
- Calf: Ayrshire heifer, small.
- 04.33 Heifer calved from the recumbent position with the result that the calf was born into a large pool of uterine fluid in a twisted position. However calf's breathing appeared to be without difficulty.
- 04.38 Occasional movements from calf, but neither calf nor heifer making any real attempts to rise. Heifer seemed very dazed after parturition.
- 04.41 Calf moved into lateral recumbency and heifer arose and immediately started gently licking the calf (calf bleating).
- 04.42 Calf moved into sternal recumbency, started bleating and attempting to rise. Several attempts almost successful but calf kept slipping over.
- 04.48 Heifer up until this point was continually licking calf, but then started becoming interested in the contaminated bedding. Milk dripping from all four quarters. Calf still struggling up and slipping, and bleating quite noisily.
- 04.58 As above.
Calf would have stood early on if it had not been for the fact that it was born into a large pool of amniotic fluid.
- 05.30 Heifer still licking calf and calf still down. Calf frequently made suckling motions into the air, and occasionally sucked its own fetlock.
- 05.41 Calf again unsuccessfully attempted to rise several times.
- 05.48 Heifer eating contaminated bedding.
- 05.56 Calf up, then immediately fell down. It once again rose to the kneeling position but was knocked over by the heifer which kept making gentle feints at it with its head.
- 06.04 Heifer down, two feet from calf and continued to lick it.
- 06.12 Calf, usddenly and easily, up. Heifer up.
- 06.13 Calf knocked down by heifer. Heifer still licking calf.
- 06.21 Heifer down, again only two feet from calf.

APPENDIX I (continued)

DN3: 14.2.67

- 06.39 Heifer up; calf, after several attempts, up. Heifer still occasionally butts calf or makes a feint at butting calf but only very gently.
- 06.41 Calf down; half slipped, half knocked down by heifer again.
- 06.42 Calf (easily) up.
- 06.43 Heifer drinking. Calf wandering around box, looking very weak at this point.
- 06.48 Calf wandering around box. Heifer excited by this.
- 07.10 Over the next hour, calf wandered around box and only making occasional teat seeking advances to the heifer. The calf frequently stood for minutes on end, doing nothing. Heifer wandered around box too, dividing its time between licking the calf and eating bedding.
- 08.10 Heifer (with no obvious effort) cleansed and commenced eating placenta.
- 09.00 Placenta eaten. Heifer quieter.
- 09.33 Calf teat seeking again but not strongly, nibbled teat base on left side once.
- 09.35 Then suckled the side of L.A. teat for one minute. Then calf ran around the box, seemed quite strong again.
- 09.37 Calf again suckling at L.A. teat, butting udder, nibbling teat side. Heifer licking calf's perineal region and standing very well.
- 09.42 Calf suckling L.A. teat. Heifer cuddling for the first time.
- 09.47 Calf suckling L.P. teat. Calf suckling for a total of 14 minutes.
- 09.57 Calf now appeared tired but was not successful in lying down even after several attempts.
- 10.02 Calf again suckling L.A. and L.P. teats for total of $3\frac{1}{2}$ minutes.
- 10.08 Calf down, heifer quietly eating hay.
- 10.30 Heifer down, two feet from calf; both calf and heifer were moaning quietly.
- 10.38 Heifer up, licking herself and calf.

APPENDIX 1 (continued)

DH3: 14.2.67

- 10.40 Heifer drinking and eating.
- 10.53 Heifer down, one foot from calf.
- 11.08 Heifer (startled) suddenly rose but later started cuddling.
- 11.28 Heifer eating and drinking.
- 11.36 Calf up and wandering around box. Later began running, jumping, bucking, etc.
- 11.42 Calf (easily) down. Heifer still standing and cuddling.
- 11.55 Heifer down, two feet from calf.
- 12.20 Heifer up.
- 12.21 Calf up. Again heifer took several gentle swings at calf with her head but never connected.
- 12.23 Calf down.

APPENDIX 1 (continued)

DH/s: 24.3.67

Dam: Small, Ayrshire heifer. Shape - good.

Calf: Ayrshire-cross-Friesian bull. Small, but very active.

23.43 Calf born from recumbent position. Heifer remained recumbent for three minutes and then rose but even while down began licking calf very roughly and blowing loudly.

23.47 Calf up into sternal recumbency.

00.05 Calf (easily) up and wandering around box while heifer licking trunk and umbilical area. Very soon, calf began teat seeking, started moving towards heifer and pushing at right side of udder.

00.08 Calf fell, heifer still licking roughly.

00.10 Calf kneeling for $\frac{1}{2}$ minute, then up again and pushing at wall.

00.12 Calf pushing at heifer's right underbelly, later at right shoulder. Heifer eating hay, seemed very hungry at this point and no longer really interested in licking calf.

00.13 Calf pushing at left side of udder. Occasionally wandered away but soon returned to the heifer and concentrated teat seeking mainly in the udder area.

00.20 Calf still pushing right, then left side of udder area. Calf very vigorous.

00.21 Calf fell but easily got up one minute later.

00.22 Pushing at left of udder again. Kicked (gently) by heifer but not put off. Still pushing mainly on right side, ranging from xiphoid to udder. Heifer not eating, licking calf. Calf suckling heifer's right stifle.

00.23 Calf stumbled to knees but up again.

00.26 Heifer again eating, calf pushing walls at corner of box. Then Heifer began licking calf again.

00.32 Calf suckling heifer's right thigh. Occasionally heifer kicks calf (gently). Calf then pushing at right side of udder, bunting, tail wagging. Continued as above.

00.43 Calf (easily) down. Heifer down, four feet from calf.

01.04 Heifer straining on placenta.

01.19 Calf up, heifer up and immediately grunting at calf and licking it. Calf stretching.

APPENDIX 1 (continued)

DHA: 24.3.67

- 01.20 Calf teat seeking again, helper standing well. Calf pushing left side mainly at udder, occasionally at xiphoid.
- 01.22 Calf suckling helpers' brisket and dewlap.
- 01.27 Calf suckling L.A. teat, then R.A. teat (from left side), then L.P. teat for a total of 10 3/4 minutes. When it emptied L.A. teat, after a total of 8 1/2 minutes suckling, it bunted the udder very hard until the R.A. teat was located.
- 01.41 Off teat, wandering around box. Helper eating contaminated bedding, but occasionally licking calf.
- 01.45 Calf pushing left of udder but not keen to suck.
- 01.46 Calf attempting to lie down, but finding it difficult.
- 01.48 Helper down.
- 01.56 Calf still trying to lie down.
- 02.00 Calf eventually down. Helper cuddling.
- 02.11 Helper up, grunting and licking calf. Straining a little on placenta. Almost immediately went down again, one foot from calf, cuddling.
- 03.37 Helper up, licking calf.
- 03.43 Helper down cuddling, calf up (roused by licking), stretched and then wandered around box.
- 03.56 Helper up, eating hay, licking calf.
- 04.02 Calf (easily) down.
- 04.22 Helper eating end of placenta.
- 04.23 Helper cleansed completely.
- 05.05 Finished eating placenta, then started on contaminated bedding. Calf wandering around.
- 05.34 Calf down.
- 05.35 Helper down, one foot from calf.
- 05.38 Helper up, licking calf, eating contaminated bedding.

APPENDIX 1 (continued)

DH4: 24.2.67

- 05.47 Calf up, suckling L.P. teat for one minute. Very powerful bunting action.
- 06.01 Calf down, heifer down and cuddling.
- 07.35 Heifer up, stretching then eating hay. Calf up, wandering.

APPENDIX 1 (continued)

DHS: 25.3.67

- Dam: Ayrshire heifer, quiet. Shape - not poor, but heifer very fat and very short legs. Therefore, as regards suckling, much depended on calf size.
- Calf: Small, Ayrshire-cross-Friesian heifer.
- 02.59 Calf born from recumbent position. Long labour period and heifer appeared to be very tired immediately post partum. Did not appear to be interested in calf.
- 03.00 Calf head shaking, sneezing, etc. Although heifer saw calf at this point, she did not seem interested in it.
- 03.03 Heifer still very subdued and down, with a mild head tremor.
- 03.05 Calf moved into sternal recumbency.
- 03.08 Heifer bled loudly just once but otherwise situation as before. Calf wet, miserable, shivering a lot. No attempts by either to rise.
- 03.22 Calf struggled a little. Heifer interested and grunting a little. Calf sneezing a lot.
- 03.30 Heifer looking brighter, licked herself several times and occasionally looked at calf. Calf very cold and miserable.
- 03.48 Heifer cuddling well, paying no attention to calf.
- 04.00 Calf attempting to rise again.
- 04.01 Calf stood for a moment, then fell. Heifer grunting excitedly. Calf attempting to rise again and its struggles eventually brought it up against the heifer. Heifer still down. Calf bleating.
- 04.08 Calf up, stood quietly by heifer's head. Ignored by heifer.
- 04.09 Heifer up, moved rapidly away from calf as if frightened of it. Calf fell but rose quickly. Heifer gently butting calf with head. Calf knocked down, up immediately.
- 04.11 Calf pushing underbelly and udder of heifer, but heifer not standing well and whenever calf passes cow while teat seeking, it moves on to push box walls and manger, etc. Calf butted and occasionally knocked over whenever near front of cow. Calf seemed very strong at this point.

APPENDIX 1 (continued)

DHS: 25.3.67

- 04.15 Calf very keen to suck but being butted hard by heifer. Finally knocked down.
- 04.17 Calf up again.
- 04.19 Calf still teat seeking, heifer circling. Calf knocked down again, but immediately up. Heifer kicking fully at calf whenever it neared hindquarters and butting whenever near front of calf. Heifer switching tail revously at this point.
- 04.25 Calf very persistent but still heifer would not let it suckle.
- 04.35 Heifer now charging calf and continually knocking it down.
- 04.40 For a few minutes, heifer stood fairly well for teat seeking advances but never at ease and eventually began butting calf again.
- 04.45 Calf down voluntarily after four or five attempts.
- 05.25 Heifer down, immediately started cuddling. Calf up and this immediately unsettled heifer. Heifer up. Calf teat seeking but heifer circling away.
- 05.28 Heifer charging calf. Calf occasionally knocked over but immediately climbed to feet. Very strong calf.
- 05.34 Calf down voluntarily and up again immediately. Attempted teat seeking again but once more attacked by heifer and knocked over.
- 05.47 Calf down, up two minutes later. As before, teat seeking advances met by aggression on part of heifer.
- 05.48 Calf up and down (voluntarily) several times.
- 05.52 Calf (very tired) down.
- 05.58 Heifer down, six feet from calf, cuddling.
- 06.20 Calf up, heifer up. Performance as before, except that heifer stood fairly well until calf actually made contact with nose, then calf attacked. Heifer kicking calf very hard.
- 06.55 Heifer cuddling quietly but still will not let calf near her udder.
- 07.00 Heifer cuddling.
- 07.10 Heifer down, cuddling. Calf wandering around box, occasionally pushing at heifer's body.

APPENDIX 1 (continued)

DUE: 25.3.67

- 07.12 Heifer up, cuddling. Did not seem so nervous, but still moved away from calf whenever it approached.
- 07.17 Heifer suddenly gave calf one or two experimental licks. Calf was standing near her head, no longer teat seeking.
- 07.20 Licking stronger and more regularly. Calf down and up again.
- 07.21 Calf down again. Heifer licking calf strongly. Calf suckling its own knee at this point.
- 07.26 Heifer down, six feet from calf, not cuddling. Both very quiet.
- 07.48 Heifer cleansed while down. No attempts made to smell or eat placenta.
- 08.00 Heifer (startled) up. Calf up. Calf pushing at left udder and not attacked by helper. Calf suckling L.A. teat, bunting and tail wagging a little. Heifer grunting at calf and licking calf's perineum. Strong bunting by calf did not seem to worry helper.
- 08.19 Heifer moved away from calf. Calf's abdomen very distended. Heifer then saw placenta and commenced eating it.
- 08.21 Calf still teat seeking on right side.
- 08.22 Calf suckling R.A. teat (3/4 minute).
- 08.25 Calf suckled for a total of 19 3/4 minutes.
- 08.30 Trying unsuccessfully to lie down.
- 08.40 Calf down. Heifer eating cleansing. Choking occasionally and breaking off to lick calf from time to time.
- 08.55 Heifer finished cleansing, then eating contaminated bedding.
- 09.01 Heifer down, one foot from calf. Licking calf.
- 09.04 Calf up (worried by licking). Heifer eating bedding.
- 09.09 Calf down next to helper.
- 09.50 Heifer cuddling.
- 10.16 Heifer (startled) up, calf up. Calf immediately moved to right side of cow and started suckling R.A. teat, then L.A. teat, then L.H. for total of 5 1/2 minutes. Heifer licked calf's perineum during this time and grunted softly.

APPENDIX I (continued)

DHS: 25.3.67

- 10.23 Calf running, bucking around box. Heifer very interested in this, grunting all the while at calf.
- 10.27 Heifer down, calf wandering. Heifer eating bedding. Calf frisking around box while heifer watches calf protectively.
- 10.59 Calf attempting to lie down.

APPENDIX 1 (continued)

DH6: 3.6.67

Dam: Ayrshire heifer, shape - good.

Calf: Ayrshire heifer, small but bright.

- 16.15 Calved. Calf born to hips while heifer recumbent, then heifer rose and dropped calf very easily. Whole of second stage labour took less than 15 minutes. Initially calf appeared dull and weak but brightened later.
- 16.16 Calf snuffling, sneezing and head shaking. Heifer not at all interested in calf. Stood motionless for several minutes following calving looking very subdued, then started eating clean straw and, very soon, hay. Ignored calf.
- 16.22 Calf rose into sternal recumbency.
- 16.25 Calf attempting to rise and rolling about floor, still being ignored by heifer (neither interested, nor frightened).
- 16.31 Calf shivering a lot, in tucked up position. Very miserable! Heifer alternately eating hay and standing motionless for long periods.
- 16.46 After several attempts, calf up.
- 16.48 Calf fell.
- 16.55 Calf up after second attempt. On rising, head accidentally came up under heifer's udder. Immediately kicked by heifer and trampled down. Stayed down, shivering. Heifer moved away, stood quietly with milk running from all four quarters.
- 17.21 Calf rose to knees easily, then up. Calf pushing very gently at wall.
- 17.23 Heifer at one point walked past the calf and took no notice of it.
- 17.24 Again calf pushing corner of box.
- 17.27 Calf fell. Heifer "fainted at calf" with head.
- 17.29 Calf up and snuffing around box.
- 17.31 Calf pushing corner walls again. Fell but immediately rose again.
- 17.38 Fell again, then up. Calf beginning to dry around eyes. Heifer ignoring calf.
- 17.41 By chance, calf pushed at heifer's elbow. Immediately heifer moved away and knocked calf down.

APPENDIX 1 (continued)

DHG: 3.6.67

- 17.43 Calf up easily, pushing corners. Calf drooling saliva and making sucking motions into air.
- 18.15 Heifer standing quietly. Calf gently pushing around walls.
- 18.19 Heifer cudding. Occasionally watching calf at this point.
- 18.32 Calf down in very hunched up position, depressed and shivering.
- 18.37 Heifer down.
- 19.14 Heifer up. Calf up (easily) pushing wall.
- 19.31 Calf ran a little, then fell. Immediately up again. Heifer (seemed very hungry) eating hay. Occasionally licking her own escutcheon and teats. Calf pushing wall.
- 19.33 Calf very gently pushed at heifer's right elbow, but heifer moved away and calf soon gave up. Whenever calf touched heifer, it visibly flinched, then moved away.
- 19.40 Due to calf's advances, heifer got very excited (tail switching) and kept circling around box, holding her head very low. Several times, calf tried to suckle from behind.
- 19.55 Several times calf pushing at udder but every time, heifer quickly moved away.
- 19.56 Calf fell and stayed down for a minute or so, then up and teat seeking.
- 20.04 Heifer knocked calf down again as it excitedly walked around box. Calf finally exhausted, lying in lateral recumbency, breathing hard.
- 20.10 Heifer down.
- 20.12 Calf up, teat seeking over heifer's left flank but heifer sleeping and taking no notice of calf.
- 20.50 Heifer in rising, knocked calf down. Calf stayed down. Heifer eating hay.
- 21.22 Heifer down, cudding.
- 21.22 Heifer no longer cudding. Calf very dull, lying hunched up and shivering.
- 22.11 Calf up, pushing heifer's sides and neck.

APPENDIX 1 (continued)

DI16: 3.6.67

- 22.27 Heifer up. Calf pushing heifer occasionally but mainly walls. Heifer sometimes almost running away from calf's advances but did not appear to be frightened by calf. Several times calf knocked down but arose easily.
- 23.07 Calf slumped down, shivering and very miserable. Heifer mostly standing quietly but occasionally took clean hay and straw. Not cleansed at this point, but not interested in contaminated bedding, nor worried by hanging placenta.
- 23.22 Heifer down. Occasionally cuddling.
- 00.15 Observation ended.

APPENDIX I (continued)

DH7: 27.6.67

- Dam: Small Ayrshire heifer. Aged about two years. Quiet. Shape - good. Udder large but well tucked up.
- Calf: Ayrshire bull. Small and initially dull.
- 15.00 Calved. Calf born from lateral recumbency after difficult and painful labour. Immediately on calving, helper rose, spun around, blared and started licking calf. Calf not moving at all.
- 15.07 Calf head shaking, snuffing. Breathing irregularly but approximately 10-12 times/minute.
- 15.10 Helper very rough and noisy. Kept circling calf while licking it, occasionally standing on its feet. Licking mainly concentrated on calf's underbelly.
- 15.18 Calf almost struggled into sternal recumbency. Helper so excited and wild that she trod on calf's chest.
- 15.22 Helper, still very wild and noisy, eating pieces of amnion, licking calf, eating contaminated bedding.
- 15.24 Calf rose into semi-sternal recumbency.
- 15.34 Calf head shaking, looking brighter. Muscular tremors of hindlegs. Helper occasionally shivering.
- 15.40 Helper still licking calf attentively but calf making no attempt to rise.
- 15.50 Calf rose suddenly into full sternal recumbency but in awkward position with chin on ground and forelegs splayed out. Helper still licking calf. Milk dripping from R.P. teat.
- 15.56 Calf struggling to rise and helper immediately blared loudly and renewed very rough licking.
- 16.04 Calf struggled into more comfortable position.
- 16.07 Calf kneeling, then knocked over by helper. Helper then eating contaminated bedding.
- 16.18 Calf again made several times to rise. Helper wildly excited.
- 16.19 Calf up and almost dry. Helper blaring loudly.
- 16.20 Calf fell but immediately (easily) up again. Helper circled away from it. Wandered weakly into corner and started pushing wall. Calf then wandering and teat seeking towards helper.

APPENDIX I (continued)

DH7: 27.6.67

- 16.25 Calf pushing anterior brisket of heifer momentarily then circling box followed by heifer.
- 16.28 Calf pushing heifer's axilla from the front.
- 16.31 Calf then pushed on weakly and nosed R.A. teat. Heifer stood momentarily.
- 16.33 Calf tail wagging and butting at R.A. teat. Then pushing at axilla. Heifer standing better and gradually quietening down.
- 16.38 Calf pushing at R.A. teat again, then sucking heifer's flank.
- 16.42 As above.
Heifer circled away gently. Grunting quietly at calf at this point--no longer blaring.
- 16.50 Calf suckling R.A. teat, then alternating between it and R.P. teat for total of 13 minutes. Heifer circled away finally.
- 17.04 Calf very distended, bucking a little.
- 17.09 Calf half fell then lay down.
- 17.11 Heifer eating contaminated straw.
- 17.20 Heifer down, two feet from calf.
- 17.25 Both (startled by dog barking) to feet. Heifer facing door of box.
- 17.28 Very soon calf started pushing right side of heifer again, then suckled R.A. for three minutes.
- 17.31 Calf wandering around box, apparently tired. Heifer not at ease, but standing quietly.
- 17.45 Calf, after several attempts, down. Heifer standing over calf, spending time licking calf, eating strands of amnion.
- 17.51 Heifer down, two feet from calf, licking it occasionally.
- 18.05 Heifer and calf up. Heifer straining.
- 18.08 Heifer cleansed.
- 18.09 Calf, without any prior teat seeking, started suckling R.A. teat, continued for five minutes (R.A. quarter must have been empty by this time).
- 18.14 Calf wandering, head butting into air, bucking, etc.

APPENDIX 1 (continued)

DH7: 27.6.67

- 18.18 Heifer eating clean bedding (just finished eating placenta).
Heifer down, calf still wandering.
- 18.19 Heifer up, moving around box.
- 18.31 Calf (easily) down.
- 18.36 Heifer down, right next to calf, occasionally licking it. Calf
sleeping. Heifer resting (not cuddling).
- 20.18 Heifer up.
- 20.21 Heifer down, touching calf, still not cuddling.
- 21.18 Calf up easily, stretching. Heifer grunting and licking calf, then
rose herself and continued licking.
- 21.25 Calf not apparently hungry.
- 21.30 Calf running around box, later just stood quietly. Heifer drinking.
- 21.37 Heifer eating clean straw keenly (seemed very hungry at this point
but only eating straw--hay available).
- 21.54 Calf teat seeking on right side again.
- 22.01 Calf suckling (for six minutes) R.A. and R.P. quarters. While doing
so, passed some meconium which was licked up by heifer as it left anus.
- 22.07 Heifer moved away and calf did not persist in suckling.
- 22.17 Heifer eating hay.
- 22.25 Calf down.
- 22.26 Heifer down, one foot from calf.
- 22.58 Heifer up, licking calf.
- 23.00 Observation ended.

APPENDIX 1 (continued)

DH8: 2.7.67

Dam: Ayrshire heifer. Very good shape for suckling. Quite nervous and, as calving approached, became slightly aggressive.

Calf: Ayrshire-cross-Friesian bull. Very strong calf.

05.43 After long labour, calf born easily. Delivered to hips while heifer recumbent, then dropped as heifer rose to feet. Heifer immediately turned and started licking calf, grunting quietly. Calf very bright, head shaking, snuffling, etc.

05.46 Heifer, at this point, started breaking off licking calf to take occasional quiet mouthfuls of contaminated bedding.

05.48 Calf rose into sternal recumbency. Head shaking, etc. This excited heifer.

06.07 Calf attempted to rise several times, but slipping on floor (heifer cleared bedding off floor while in labour).

06.09 Calf kneeling. Heifer very excited, grunting, licking.

06.13 Calf still kneeling, hind feet slipping.

06.14 Calf fell.

06.17 Heifer down, two feet from calf. Still licking calf.

06.22 Heifer much quieter at this point.

06.25 Calf made attempts to rise again, but slipping and fell right away from heifer. Heifer excited, up. Finally calf slumped down.

06.33 Heifer eating contaminated bedding.

06.36 Heifer drinking, then again licking calf.

07.09 Calf again made several attempts to rise but still could not get a grip on slippery floor. Heifer very excited.

07.28 Heifer down, two feet from calf, licking it. Calf very tired. Milk dripping from heifer's udder and she licked this up herself.

08.14 Calf again tried to rise but eventually fell exhausted into lateral recumbency.

08.30 Calf lying quietly. Heifer licking herself.

09.10 Heifer strained violently, rose and cleansed. Started eating placenta immediately.

APPENDIX 1 (continued)

DH8: 2.7.67

- 09.11 Calf rose into kneeling position, then fell.
- 10.04 Calf suddenly got up quite easily. Heifer licking it, grunting softly. Calf immediately made teat seeking advances to heifer, pushing at first dewlap and brisket, then underbelly.
- 10.06 Calf pushing left side of udder.
- 10.09 Calf fell but rose within $\frac{1}{2}$ minute with difficulty (seemed very tired).
- 10.11 Teat seeking, pushing at right of udder. Then wandering, giving occasional buck.
- 10.30 Calf fell down and stayed down. Soon took up sleeping position. Heifer standing quietly by.
- 10.42 Heifer down for a moment, then (startled) up and eating hay.
- 10.48 Heifer down, three feet from calf.
- 11.09 Heifer up, licking herself. Later eating hay.
- 11.13 As soon as heifer started licking calf it got up (easily) and started to teat seek very strongly at right of udder. Heifer licking calf's perineum. Calf sucking heifer's stifle and anterior aspect of tibia. Milk running out of heifer's R.P. quarter.
- 11.17 Calf's nibbling sides of teats caused heifer to gently kick it. Calf pushing left of udder. Calf suckled L.A. teat for an instant. Heifer cuddling.
- 11.21 Calf fell and stayed down.
- 11.22 Calf kneeling.
- 11.23 Calf up and pushing at first the underbelly, then brisket and axilla and finally at left side of udder. Calf making suckling motions in air.
- 11.28 Calf nibbling side of L.A. teat.
- 11.29 Calf suckling L.A. teat, then L.H. teat for total of 19 minutes. Occasionally, early on, heifer gently kicked calf's face and once or twice moved away for a second or two but calf very persistent. Some meconium that was passed while suckling was licked up by heifer. Towards end of suckling spell, heifer was cuddling.

APPENDIX 1 (continued)

DH8: 2.7.67

- 12.10 Calf down (easily). Heifer eating clean straw.
- 12.14 Calf in sleeping position.
- 12.46 Heifer down, immediately took up the sleeping pose.
- 13.07 Heifer up, cuddling. Calf stirred but then again took up sleeping pose.
- 1.43 Observation ended.

APPENDIX 1 (continued)

DH9: 7.7.67

Dam: Ayrshire heifer. Shape - good, large teats for heifer (about 3" long).

Calf Ayrshire heifer.

07.45 Calf born from recumbent position.

07.47 Heifer up, licking calf strongly and grunting. Initially looked stunned however. Calf still in lateral recumbency, struggling weakly. Head shaking, snuffing.

07.52 Heifer occasionally breaking off to lick and eat contaminated straw.

08.03 Calf in sternal recumbency.

08.05 Calf making first attempts to rise and heifer concentrating on licking calf again.

08.30 Calf up, swaying.

08.32 Calf took first shuffling steps towards heifer. Heifer licking calf's underbelly.

08.45 Calf still standing. Had not fallen but had not moved much either. Stood passively near heifer. Heifer still occasionally licking calf, grunting.

09.01 Heifer occasionally straining on placenta.

09.05 Calf fell and stayed down. Heifer then stood quietly over calf. Heifer still occasionally straining.

09.24 Calf suddenly and easily up, wandering. Heifer licking calf's perineum. This time calf walked towards heifer. Heifer still backing away from calf's advances, licking its head.

09.29 Heifer stood still finally. Calf almost immediately centred on the left side of the udder after pushing initially at the left underbelly.

09.31 Calf nuzzling left teats. Then pushed passed hindquarters of heifer, finally to push at manger.

09.36 Calf straining to pass meconium (before suckling).

09.37 Calf tried to go down unsuccessfully. Heifer drinking. Calf drooling saliva.

09.43 Calf with difficulty down. Heifer licking calf.

APPENDIX 1 (continued)

DH9: 7.7.67

- 10.02 Heifer drinking. Calf up, wandering, then pushing at wall.
- 10.04 Twice, on approaching heifer, calf was kicked hard. Calf pushing wall. Heifer licking calf's perineum from behind.
- 10.14 Calf, trying to run, fell. Immediately up and pushing wall.
- 10.22 Calf still pushing wall.
- 10.28 Calf pushing at right udder then suckling. Heifer standing well. At times, heifer moved away from calf and once she kicked it. Suckling for a total of 6½ minutes R.A. and (once) R.P. teats.
- 10.48 Heifer straining again, suddenly a large amount of placenta appeared, and heifer started circling to catch end of it. Calf wandering, tired.
- 11.00 Heifer finally caught end of placenta and started eating it.
- 11.07 Calf slumped down.
- 11.16 Heifer up and down several times over next few minutes. Clearly worried by placenta. Straining occasionally.
- 11.23 Calf up.
- 11.24 Heifer cleansed and began eating it. Calf down.
- 11.40 Heifer finished eating placenta.
- 11.50 Heifer down, resting. Not cuddling. Calf sleeping.
- 13.02 Heifer suddenly up. Calf up, too, but made no attempts to suckle. Just standing.
- 13.17 Calf down. Heifer eating remainder of contaminated straw.
- 13.21 Heifer down, straining occasionally.
- 13.30 Heifer took up sleeping pose.
- 14.42 Calf up (easily). Wandering slowly around box. Calf running and jumping a little, fell and stayed down.
- 14.47 Heifer (had been worried by calf moving about) up. Licking calf.
- 14.48 Calf up. Standing quietly. Heifer eating hay.

APPENDIX 1 (continued)

DH9: 7.7.67

15.11 Calf down. Still tired.
15.16 Heifer down, cuddling.
15.45 Observation ended.

APPENDIX 1 (continued)

DH10: 12.7.67

- Dam: Ayrshire helper, aged approximately two years. Shape - very good, quiet.
- Calf: Ayrshire-cross-Friesian bull. Large and very dull, slightly contracted fore and hindleg tendons but not severe enough to incapacitate calf.
- 23.40 Calved after relatively easy labour, considering calf quite large. Calf born to hips from helper in recumbent position, helper then rose and after several strong strains, passed calf.
- 23.45 Calf still lying in lateral recumbency, breathing regularly but not making any attempts to rise or head shaking, etc. Since calving, helper has been gently licking calf, mainly over trunk, softly grunting.
- 23.48 Helper beginning to take interest in contaminated bedding.
- 23.50 Calf stirring. Helper immediately began licking calf full time again, grunting. Helper occasionally straining.
- 23.52 Calf into sternal recumbency, snuffing, sneezing, head shaking. Helper licking calf and softly grunting, mainly licking head.
- 23.56 Calf still in same position. Helper again began eating small amounts of contaminated bedding, breaking off occasionally to lick calf.
- 24.00 As above. Calf very dull.
- 00.15 As above, except helper spending more time smelling and eating contaminated bedding.
- 00.19 Calf attempted to rise several times but weakly and soon gave up. Helper excited and renewed licking (more powerful than before). Calf only succeeded in just raising hindquarters off ground. Marked muscular tremors over calf's hindquarters.
- 00.24 After several more attempts, calf managed to assume kneeling position, but remained thus for several minutes, paddling with hind feet. Helper licking mainly calf's head, grunting.
- 00.27 In attempting to stand, calf fell and stayed down. Very dull. Helper eating clean bedding now.
- 00.36 Several more attempts by calf to rise, finally calf kneeling but then accidentally knocked over by helper's licking. Calf getting stronger.
- 00.38 Calf up but immediately sprawled. Calf in lateral recumbency.

APPENDIX 1 (continued)

DH10: 12.7.62

- 00.39 Calf into sternal recumbency.
- 00.44 Calf again kneeling.
- 00.50 Calf finally managed to stand after kneeling for six minutes. Very unsteady. Heifer licking calf's head.
- 00.51 Calf shuffling towards helper (helper very excited). Helper circling away from calf, still licking calf's head.
- 00.52 Helper standing and calf pushing helper's right side. Started at elbow and worked along to udder but then went on past helper's thigh and shuffled right to wall.
- 00.55 Calf still pushing wall and worked along to corner. Helper licking calf's back and hindquarters.
- 00.59 Calf fell but immediately rose and started advancing towards helper again. Pushing helper's underbelly then pushing on right side of udder. Helper standing wall, eating straw.
- 01.10 As above. At one point, calf started sucking helper's femoral fold, but helper moved away. Most of time, calf has been pushing at right of udder but missing teats altogether. Teat seeking advances were not too powerful anyway.
- 01.11 Helper moved away to take a drink then started cuddling.
- 01.12 Calf followed helper but did not renew teat seeking advances just stood by helper.
- 01.14 Helper down, cuddling but as soon as calf moved a few steps, rose grunting, and started licking calf.
- 01.18 Calf wandering around box. Helper standing quietly.
- 01.19 Calf attempting to lie down, having trouble with hindlegs.
- 01.20 Calf fell and stayed down, lying in hunched up pose; very dull.
- 01.23 Helper down, one foot from calf.
- 01.24 Helper up, licking calf initially then just standing quietly near calf.
- 01.29 Helper down, two feet from calf, cuddling.
- 01.30 Helper in sleeping pose.

APPENDIX I (continued)

PH10: 12.7.67

- 02.36 Heifer up, smelling bedding where her hindquarters were resting, then licking and eating bedding. Much of placenta now showing and this apparently upsetting heifer.
- 02.42 Heifer licking calf, grunting.
- 02.44 Calf up easily enough although spent at least $\frac{1}{2}$ minute on knees.
- 02.45 Calf weakly teat seeking. Centred on heifer's left udder quickly enough and pushed at right level but kept missing teats. Finally after several minutes just stood with nose by L.A. teat.
- 02.51 Calf nibbling L.A. teat from side. Heifer, which had been grunting softly and licking calf's perineum, moved away.
- 02.52 Calf followed heifer, pushing again on left side and showing occasional weak bunting action.
- 02.55 Calf again standing with nose among teats, not teat seeking.
- 02.56 Calf (still very dull) wandering around box. Gave slight buck and fell but rose immediately, only to stand quite still away from heifer.
- 02.57 Heifer eating hay, but not at ease. Still worried by placenta.
- 03.10 Heifer down. Calf wandering slowly around box for few minutes, then finally stood still.
- 03.45 Heifer up. Almost completely cleansed.
- 03.46 Heifer cleansed and began eating placenta. Did not strain; placenta apparently fell out.
- 03.47 Calf again pushing heifer's left side. Several times heifer slightly moved away from calf's advances and calf almost completely gave up.
- 03.59 Calf sucking heifer's stifle, again heifer moved away.
- 04.00 Calf wandering around box.
- 04.03 Calf down after several attempts, again in hunched up pose.
- 04.22 Heifer finished cleansing; starting on contaminated straw.
- 04.26 Heifer licking calf and grunting. Calf lifted head but did not rise. Very dull at this point.
- 04.33 Heifer down, two feet from calf.

APPENDIX 1 (continued)

DH10: 12.7.67

- 05.38 Heifer up, licking calf. Calf up (easily). Calf teat seeking on left side of heifer; quickly centred on udder but still not seeming to be able to get teats (not large) into mouth. Teat nibbling upset heifer (never kicked, just moved away).
- 05.43 Calf suckling L.A. teat for total of $4\frac{1}{2}$ minutes, in $\frac{1}{2}$ minute spells. Then heifer moved (really on changed position) and calf gave up. Heifer licked perineum for short while during suckling.
- 05.54 Calf wandering, very tired again. Heifer eating hay.
- 06.13 Calf down, sleeping.
- 06.18 Heifer down, two feet from calf, cuddling.
- 07.40 Observation ended.

APPENDIX 1 (continued)

DCI: 31.1.67

- Dam: Ayrshire cow; old; quiet. Shape - very poor.
- Calf: Ayrshire-cross-Friesian; bull- average size.
- 00.35 Calf born. Cow had been straining forcefully for about 4 hours and, at parturition, seemed very tired. Calf born from recumbent position and born almost completely before amnion ruptured. Result being that calf born into very large, deep pool of amniotic fluid.
- 00.37 Cow up, licking calf keenly. Calf coughing, mouth breathing, with a large respiratory rattle, much mucus.
- 00.38 Calf lying in sternal recumbency. Cow licking roughly and blaring loudly. Initially the cow concentrated on licking the underbelly (and umbilicus).
- 00.39 Calf's first attempts to rise, but slipping on amnion. Kept evading cow's attempts to lick head.
- 00.50 Cow still licking hard, gave an occasional yawn.
- 00.54 Cow showing interest in amnion and contaminated bedding. Went back to calf as soon as it again slipped on attempting to rise.
- 01.02 Cow (occasionally grunting) standing, doing nothing.
- 01.04 Calf (quite strong) still attempting to rise but slipping.
- 01.08 Cow eating contaminated straw quite avidly.
- 01.14 Calf still lying on dirty, wet straw-shivering.
- 01.19 Once again, calf attempted to rise and cow immediately rushed over and started grunting and licking it.
- 01.20 Calf up for a second, then slipped down again. Up and down several times.
- 01.27 Cow down. Very soon after, calf rose with little trouble. Cow excited but did not rise. Calf test seeking and pushing box wall.
- 01.33 Cow up and over to calf, grunting, licking.
- 01.38 Calf slipped down.
- 01.50 Cow down momentarily. Then calf up, still pushing wall. Being licked by cow.

APPENDIX 1 (continued)

DC1: 31.1.67

- 01.52 Calf down; bleating a little, especially when passing meconium. Cow no longer interested in amnion but still licking calf.
- 01.55 Calf pushing wall. Running a little then fell.
- 02.05 Calf up, pushing wall then fell. Cow standing quietly, occasionally grunting and giving calf a lick.
- 02.13 Cow down, seemed stiff. Slightly tachypnoeic and occasionally coughing. Calf three feet from cow's left shoulder, and it seems hunched up.
- 02.37 Suddenly calf up (easily), pushing wall and occasionally running. Cow grunting at calf, then up.
- 02.42 Calf fell but immediately arose and started pushing at cow.
- 02.48 Calf down, cow licking it occasionally and eating straw.
- 02.52 Cow up, running and jumping. Does not seem keen to teat seek at cow. Wandering.
- 03.00 Calf down; cow eating clean bedding, then cuddling.
- 03.08 Cow drinking for $\frac{1}{2}$ minutes.
- 03.28 Cow down, cuddling. Calf down four feet from cow's left shoulder.
- 03.58 Calf up; running, bucking and very alert.
- 03.59 Cow up. Occasionally calf bucking at cow. Still not interested in suckling.
- 04.15 Occasionally calf teat sought in a half hearted way, but spending most of time at cow's axilla. Cow standing well for suckling. When calf was pushing at front of cow, cow would occasionally push calf posteriorly with its head. Then calf sucking at cow's hock.
- 04.25 Cow eating straw.
- 04.31 Calf running, then fell and stayed down.
- 04.36 Calf up. Obviously hungry from this point. Circling cow very closely but pushing far too high up cow's side.
- 04.38 Cow down, straining occasionally on the placenta (just showing).
- 04.43 Cow cuddling; more and more placenta appearing now.

APPENDIX 1 (continued)

DC1: 31.1.67

- 04.46 Cow cleansed, still cuddling.
- 04.49 Calf down, three feet posterior to cow.
- 05.53 Calf, after two quick bleats, up. Cow blaring. Calf stretching.
- 06.00 Cow up, in response to calf's pushing and bleating. Calf obviously hungry, circling cow and continually pushing, but pushing too high and often anteriorly at brisket and axilla.
- 06.09 This cow did not seem interested in cleansing-rotting straw. Calf teat seeking and pushing, often at axilla.
- 06.12 Calf passing more meconium than teat seeking once more.
- 06.33 Calf down, cow licked it for a minute then resumed her eating.
- 06.48 Calf up, roused by cow smelling and licking. Calf sucked cleansing for a short time. Cow standing but "padding." It appeared that it was mainly standing for calf's advances and would really have preferred to lie down.
- 07.04 Calf sucking at placenta.
- 07.09 Cow down, cuddling. Calf exploring, occasionally sucking at placenta.
- 07.45 Cow sleeping.
- 07.45 Cow cuddling.
- 07.58 Calf chewing and sucking wet straw. Calf hungrily circling cow.
- 08.00 Calf down, then up. Seems very tired.
- 08.13 Calf sucking at blood, passed with placenta.
- 08.25 Observation ended.

APPENDIX 1 (continued)

DC2: 16.2.67

- Dam: Ayrshire, mature adult. Shape - poor, but not excessively so and teats on outside of udder.
- Calf: Ayrshire-cross-Friesian heifer; large and strong.
- 23.37 Calved. Calf born to hips while cow recumbent, then, when cow rose, calf was immediately dropped.
- 23.39 Calf weakly struggling. Cow eating amnion and licking and eating contaminated straw but not yet licking calf.
- 23.41 Calf sneezing, coughing. Much fluid ran from mouth and nose at this point (calf still in lateral recumbency).
- 23.42 Calf bleated. Cow very excited, blared loudly, and began licking calf.
- 23.44 Calf rose into sternal recumbency. Cow again more interested in amnion and contaminated bedding than in calf.
- 23.45 Cow again licking calf. Calf attempting to rise.
- 23.49 Calf almost up twice but slipped on amnion. Cow licking calf very strongly and grunting but still interested in amnion, etc.
- 23.56 Calf kneeling.
- 23.58 Calf up, but standing on anterior aspects of fore fetlocks. Fell after $\frac{1}{2}$ minute.
- 00.01 Calf kneeling.
- 00.02 Calf up again, then fell.
- 00.05 Calf up, fell again.
- 00.07 Calf making butting motions from floor. Cow still licking calf.
- 00.19 Cow eating hay.
- 00.20 Calf up (easily).
- 00.22 Calf making teat seeking advances to cow. Started pushing at cow's right axilla and then started pushing on right of udder. Strong bunting movements but unsteady on feet.
- 00.23 Calf pushing at left udder then fell under cow and stayed down. Cow, grunting softly, moved gently away from calf and started eating hay. Occasionally licked calf.

APPENDIX 1 (continued)

DC2: 16.2.67

- 00.26 calf up, pushing at walls.
- 00.28 Cow drinking. Calf pushing at left udder and nibbling teats.
- 00.31 Calf sucking side of L.A. teat for a few seconds. Cow licking calf's hindquarters.
- 00.35 Calf nibbling teats. Most of the time this calf concentrated on pushing at udder. However, whenever it did push anteriorly (at xiphoid) the dam walked forward so that calf was eventually forced to push at udder.
- 00.38 Calf pushing at right of udder and nibbling teats. Calf then wandered away from cow, then stood and passed meconium.
- 00.41 Calf teat seeking once more; nibbling L.P. teat. Seemed to be having trouble getting the end of the teat into its mouth.
- 00.48 Cow gently made "feint" at calf with head.
- 00.49 Cow drinking.
- 00.52 Calf straining and passed more meconium.
- 00.55 Calf bucking and running about box.
- 00.58 Once more calf was nibbling the left teats and sucking udder skin. Cow eating hay.
- 01.03 Calf down, then kneeling, then down again (some difficulty in doing so the second time). Cow licked calf, then cow down.
- 02.00 Cow and calf up.
- 02.05 Calf pushing right udder--cow standing well.
- 02.10 Calf suckling for total of 16 minutes on R.A., R.P. and L.A. teats.
- 02.40 Calf finally stopped suckling and ran about box.
- 02.42 Calf still running about box, coughing occasionally.
- 02.49 Calf down (easily). Cow eating hay and giving occasional lick to calf.
- 02.59 Cow drinking, then again licked calf.
- 03.02 Cow down, two feet from calf, resting.

APPENDIX 1 (continued)

DC2: 16.2.67

- 03.39 Calf up, bleating.
- 03.40 Cow up. Cow then showed "Flehman" while licking calf. Then calf wandered away, bucking occasionally.
- 03.47 Calf pushing at right udder but not keenly.
- 03.51 Calf down but very soon quickly got up and loudly bleated. Cow excited and blared for a few seconds.
- 04.09 Cow drinking.
- 04.10 Calf then suckled in small spells for a total of 12½ minutes from R.A., R.P. and L.P. teats. This was the first calf seen to suckle from behind its dam, (L.P. teat). During suckling, cow continually licked calf's perineum.
- 04.28 Calf stopped suckling.
- 04.30 Calf down.
- 04.52 Cow down, two feet from calf, cuddling.
- 05.22 Cow up, eating concentrates. A large amount of placenta appeared as cow rose. Cow then licking contaminated bedding.
- 05.30 Cow suddenly arched back and cleansed. Cow very excited grunted loudly and immediately began eating placenta.
- 05.31 Calf up and wandering around box.
- 05.44 Cow still eating placenta. Calf pushing at right udder for a moment, then wandering, running and bucking.
- 05.57 Cow left placenta for about two minutes to eat a little hay and concentrates, then returned to placenta.
- 06.00 Calf pushing right udder for ½ minute.
- 06.06 Calf down. Cow eating contaminated straw (finished placenta).
- 06.11 Cow drinking.
- 06.15 Calf up; both cow and calf grunting and bleating respectively. Calf wandering. Cow eating hay.
- 06.19 Calf down.

APPENDIX 1 (continued)

DC2: 16.2.67

- 06.22 Calf up into kneeling position, then up. Cow licking calf's hindquarters.
- 06.23 Calf down.
- 06.24 Cow down, right next to calf.
- 06.53 Cow up, eating contaminated straw. Calf sleeping.
- 07.37 Observation ended.

APPENDIX 1 (continued)

DC3: 23.2.67

Dam: Mature Ayrshire cow. Shape - poor, but not excessively so. Quiet.

Calf: Ayrshire bull; large and strong.

02.40 Calved, very quickly and easily. Calf born to hips while cow recumbent and then dropped as cow rose.

02.42 Calf into sternal recumbency (this calf was sneezing and head shaking immediately after birth).

02.50 Calf attempting to rise. Strong. Calf slipping though. Cow licking calf since calving. Gentle licking and all the time, softly grunting.

03.00 Calf lying quietly in sternal recumbency. Cow still licking calf, concentrating on head.

03.15 Cow occasionally breaking off to lick and eat contaminated bedding. Calf not attempting to rise, lying quietly, but strong and bright.

03.31 Calf again made attempts to rise and finally rose into kneeling position. Cow still licking calf.

03.34 Calf up, then fell. Already much of cow's placenta showing.

03.35 Calf up, but slipping badly on floor. Cow licking calf's perineum.

03.36 Calf down again. Already calf drooling saliva. Cow licking contaminated bedding again.

03.42 Calf kneeling, then walking around box on knees.

03.43 Calf down with little difficulty. Calf teat seeking over cow's body while down as cow stood over it. Cow licking calf.

03.44 Calf up easily, although slipping once on feed.

03.45 Calf down. Cow very gentle but attentive.

03.52 Calf up, approaching cow. Calf initially probing at cow's shoulders, then at underbelly.

03.55 Calf (pushing) approached cow's udder, then walked right past it to wall and started pushing at corner.

03.57 Calf again pushing at cow's brisket, then underbelly, then udder and once again continued to push right past cow's thigh. Several times over last few minutes, cow lifted a foreleg to allow calf to progress posteriorly from initial position at brisket.

APPENDIX I (continued)

DC3: 23.2.67

- 03.58 Calf pushing at right udder and nosing teats. Cow standing well, licking calf's perineum, softly grunting.
- 04.04 Calf fell but immediately stood up. Cow eating large amounts of contaminated bedding.
- 04.07 Calf pushing at cow's belly, then udder but pushing too high. Then fell.
- 04.09 Calf up, wandering. Once again, cow continuously licking calf.
- 04.16 Calf pushing cow's groin and flank. Then udder and finally at teat level. At one point, calf was nibbling the L.A. teat but seemed unable to get teat end into mouth.
- 04.18 Calf fell but stood up $\frac{1}{2}$ minute later. Cow eating clean straw. Calf then wandered about near cow, but not teat seeking.
- 04.30 Cow drinking, then licked calf, then started eating.
- 04.32 Cow drinking again.
- 04.45 Cow down near calf. Licking calf.
- 04.47 Calf down, three feet from cow.
- 05.03 Calf up, wandering.
- 05.08 Calf down, four feet from cow. While calf was wandering, cow seemed unsettled, especially when calf neared the box door (9 feet away).
- 05.09 Cow up, licking calf.
- 05.12 Cow eating contaminated bedding. Large part of placenta showing.
- 05.19 Calf up and immediately started pushing left udder. Cow grunting but moved away.
- 05.23 Calf running around box, bucking. Calf down.
- 05.24 Calf down, four feet from cow. Cow licking calf.
- 05.31 Calf up, wandering around cow.

APPENDIX 1 (continued)

DC3: 23.2.67

- 05.34 Cow cleansed with no apparent effort. Did not rise. Calf wandering around box, giving occasional weak butts into the air.
- 05.37 Calf down easily but immediately rose.
- 05.40 Calf down, four feet from cow. Cow sleeping.
- 06.37 Calf up, stretched. Wandering around cow.
- 06.41 As soon as calf reached the box door, the cow grunted and rose.
- 06.43 Cow started licking, then eating the placenta. Calf teat seeking again. Calf (drooling saliva) pushing first at left udder, then at the right axilla.
- 06.51 Calf again pushing at left udder (at teat level).
- 06.54 Calf down, under cow.
- 06.58 Calf (still looking very strong) up and vigorously pushing at manger. Tail wagging and bunting with head.
- 06.59 Calf running around box but then ended up sucking cow's left axilla, from the front. (It had done this several times by this time.)
- 07.02 Calf again sucking cow's left axilla, then brisket. Then wandered away from cow.
- 07.07 Calf down. Cow grunted occasionally at calf while eating placenta but otherwise showed no interest in it.
- 07.11 Cow finished placenta and started licking calf for few moments. Then eating contaminated bedding.
- 07.30 Calf teat seeking again. Cow frequently circled hindquarters towards calf and trapped calf's nose at the udder area. Calf nibbling side of teats, but still unable to get teat end into mouth.
- 07.37 Calf pushing and sucking axilla as often as udder skin, and occasionally wandering away from cow.
- 07.39 Calf down. Cow eating hay.
- 08.02 Cow down, cuddling.
- 08.13 Cow up, cuddling.

APPENDIX I (continued)

DC3: 23.2.67

- 08.16 Calf almost trod on by cow, startled, then stood up easily. Cow grunted occasionally to calf.
- 08.19 Calf pushing at cow's underbelly very powerfully (seemed very hungry). Finally calf pushing at axilla and brisket.
- 08.21 Cow down, grunting to calf, then cuddling.
- 08.25 Calf down.
- 09.11 Calf up, loudly bleating, very hungry. Cow up, stretching. Calf (bucking a little) pushing at cow's belly, then axilla.
- 09.15 Calf down. Cow licking it initially, then cuddling.
- 09.28 Once again, cow almost stood on calf and calf (startled) stood up. Cow grunting and cuddling. Calf then started pushing at cow's right axilla, brisket and foreleg.
- 09.41 Calf finally started suckling L.A. teat and continued to suckle this one teat for a continuous 18 minutes.
- 09.59 Stopped suckling and started passing meconium.
- 10.00 Calf again suckled from L.A. teat for a further 1½ minutes. Calf very bright and very distended.
- 10.08 Calf down. Cow licking calf, then eating hay and concentrates.
- 10.36 Calf up due to cow's licking and passed some more meconium. Then ran around box, bucking.
- 10.40 Observation ended.

APPENDIX I (continued)

DC4: 22.3.67

Dam: Mature Ayrshire. Quiet. Shape - very poor, although teats placed on outside of udder.

Calf: Ayrshire bull. Large and strong.

20.56 Calf born from cow in recumbent position throughout. Calf blinking, swallowing and moving head immediately but not breathing regularly until three minutes post partum.

21.00 Cow up, gently licking calf. More interested in eating amnion than in licking calf grunting. Calf rose into sternal recumbency (much fluid coming from calf's mouth and nose). Cow sucked up fluid rather than licked.

21.03 Cow licking roughly by this time and interested in the calf's umbilicus. Calf coughing. Cow getting excited and noisy.

21.08 Calf still fairly quiet.

21.10 Calf's first attempts to rise. Cow licking calf. Calf head shaking, sneezing, etc.

21.21 Calf again attempting to rise but slipping on wet straw.

21.33 Cow eating contaminated bedding.

21.36 Calf almost up, then slipped. Cow very excited, noisy, licking.

21.47 Calf standing for a few moments, then down.

21.50 Calf up, unsteady. Cow still excited.

21.51 Calf fell. Milk dripping from cow's back quarters.

21.52 Calf (easily) up.

21.55 Calf snuffling towards cow. Cow (very excited) kept backing away from calf as it advanced, licking its head.

21.58 Calf fell, struggled for a few moments, then lay quietly.

22.01 Calf up (easily). Cow still backing away from calf's advances, licking its head.

22.04 Calf pushed right udder for a few seconds, then fell.

22.07 Calf up.

22.15 Cow standing well and calf pushing at brisket and underbelly very vigorously. Cow moved away and calf ended up pushing manger.

APPENDIX 1 (continued)

DC4: 22.3.67

- 22.17 Calf fell and stayed down.
- 22.19 Cow drank for a moment, then started eating hay, grunting at calf.
- 22.34 Calf up. Cow licking calf then feinted gently at calf with head but accidentally knocked it over. Cow often backed away from calf's advances even at this point, while licking its head roughly.
- 22.36 Cow standing, grunting at calf. Calf pushing at left udder, then left axilla, then underbelly. Finally pushing into cow's left groin.
- 22.367 Calf again concentrating on pushing above udder, then axilla on left side. Then calf fell.
- 22.38 Calf up and again pushing at cow's underbelly.
- 23.00 Milk running from R.P. teat.
- 23.10 Calf vigorously pushing and bunting above udder again.
- 23.25 At this point calf held both right teats in mouth but was only nibbling them from the side.
- 23.29 Calf pushing above udder again, and salivating profusely (making loud sucking noises). Cow still occasionally moves away (does not like calf sucking its skin).
- 23.32 Calf pushing above udder (tired).
- 23.36 Calf standing quietly near cow. Cow (grunting) and smelling bedding.
- 23.37 Cow down.
- 23.39 Calf (tired) knelt down, then stood up again.
- 23.40 Calf down, three feet from cow.
- 00.30 Calf up easily. Cow up, grunting.
- 00.31 Calf immediately started pushing left udder. Cow standing well. Calf soon nibbling teats although was often still pushing too high; milk pouring out of teats.
- 00.33 Calf very vigorous again; butting and tail wagging; drooling saliva. Then ran around box, bucking.
- 00.57 Calf still teat seeking. Cow gently kicked it once or twice, where it got particularly rough while bunting or when it started sucking above the udder.

APPENDIX 1 (continued)

DE4: 22.3.67

- 01.14 Calf suckling R.P. teat, then L.A. and R.A. teats for a total of 7½ minutes (not all from right side). Cow eating hay.
- 01.29 Calf stopped suckling, wandered away from cow.
- 01.30 Cow down.
- 01.38 Cow up.
- 01.55 Cow eating hay; still grunting and occasionally licking calf. Calf very distended.
- 02.09 Calf down. Cow licked it initially then wandered around box, eating clean straw.
- 02.29 Cow down right next to calf, cuddling.
- 03.05 Cow and calf (startled) up. Cow eating and wandering around box. Calf just wandering too.
- 03.17 Calf urinated then passed meconium..
- 03.30 Cow cuddling.
- 03.54 Cow down, cuddling.
- 04.26 Cow up, wandering.
- 04.48 Calf knelt, then stood up again. Cow straining on cleansing.
- 04.56 Observation ended.

APPENDIX 1 (continued)

DC5: 24.3.67

- Dam: Mature Ayrshire. Shape - poor. Large udder with large teats.
- Calf: Ayrshire-cross-Friesian, bull, average size, strong.
- 22.15 Calf born from calving with cow in lateral recumbency. Long and difficult labour and cow did not immediately rise following the completion of calving. Calf head shaking and sneezing while still lying with hindfeet in cow's vagina.
- 22.18 Cow attempting to eat contaminated bedding while still down.
- 22.20 Calf rose into sternal recumbency.
- 22.22 Cow up and licking calf. Became interested in calf as soon as it started struggling.
- 22.25 Cow eating shreds of amnion, and seemed more interested in this than in calf.
- 22.30 Cow still mainly concentrating on amnion.
- 22.33 Calf struggling to rise and cow licking calf (finished licking amnion, but, anyway, much more interested in calf when it struggled).
- 22.35 Cow started eating contaminated bedding again.
- 22.40 Cow licking calf for a few moments, then back to contaminated bedding.
- 22.56 Cow down, four feet from calf, licking itself.
- 23.02 Cow up and licking calf. Calf again made a few attempts to rise. Later made suckling movements at the head and neck of the cow as it licked it.
- 23.10 Calf again attempted to rise and made kneeling position, but then fell. Lying quietly. Cow eating contaminated bedding.
- 23.20 Cow down, licking calf two feet away.
- 23.32 Calf up (easily) and immediately started pushing at recumbent cow. Cow still down but licking calf.
- 23.33 Calf then lay down.
- 23.34 Calf up, then fell but immediately rose and started wandering. Cow up.
- 23.40 Calf fell and stayed down.
- 23.43 Cow down.

APPENDIX I (continued)

DC5: 24.3.67

23.54 Calf up and wandering around box. Cow up.

00.01 Calf down and then up three minutes later. Walking towards cow this time.

00.08 Calf approaching cow and bumping cow's brisket and neck.

00.10 Calf passed cow, pushing wall and corners. Cow licking calf's perineal area from behind.

00.12 Calf pushing cow's underbelly, then at right udder but above teat level. Once more than passed on and started pushing at corner of box.

00.15 Calf began running around box.

00.16 Calf started pushing at right udder and started making suckling motions into the air. Then calf wandering.

00.19 Calf pushed cow's right thigh and cow immediately moved away. Calf then started pushing udder from behind cow.

00.20 Calf fell and stayed down. Cow eating contaminated straw.

00.23 Cow down, cuddling.

00.35 Cow (grunting) up and licking calf.

00.36 Calf (easily) up and wandering around box. Calf pushed at cow's brisket for a moment then started bucking and running around box.

00.42 Calf fell and stayed down. Cow licking calf.

00.55 Calf up, wandering around box.

01.00 Calf fell but stood up one minute later.

01.04 Cow drinking.

01.05 Calf down. Cow licking calf.

01.13 Cow down, licking calf for a while then cuddling.

01.43 Cow up, licking calf.

01.45 Calf up, weakly pushing cow, initially at brisket then worked back to right side of udder. Calf getting stronger latterly. Cow eating hay.

APPENDIX I (continued)

DC5: 24.3.67

- 01.53 Calf wandering around box. Cow licking calf.
- 01.54 Calf almost down but stirred up again by cow's licking.
- 02.00 Calf at one point, when pushing udder, almost suckled a teat but cow gently kicked it and then moved away.
- 02.01 Calf down.
- 02.04 Cow down, licking herself then cuddling.
- 02.42 Calf up, immediately followed by cow (grunting). Calf pushing right udder, finally nibbled teats, then wandered off.
- 02.51 Cow down, and then suddenly showed "Flehman." Calf fell but immediately up again.
- 03.18 After several weak strains, cow almost cleansed. Then cuddling.
- 03.23 Cow up, licking calf.
- 03.26 Cow then started eating contaminated bedding, then suddenly cleansed and started to eat placenta.
- 04.04 Calf up, stretching. Cow finished placenta, started on eating contaminated straw.
- 04.05 Calf, vigorously bunting and tail wagging, started pushing cow's underbelly and udder.
- 04.11 Calf pushing on right of udder and finally nibbling teats.
- 04.13 Calf suckling R.A. teat continuously for seven minutes. Cow initially drinking, later just standing well.
- 04.20 Calf stopped suckling but still pushing.
- 04.27 Calf down.
- 04.29 Calf up, running around box. Cow drinking.
- 04.35 Calf down and up several times. Cow eating clean straw. Calf finally down.
- 04.40 Cow down, cuddling.
- 05.29 Calf up, running around box.
- 05.30 Cow up.

APPENDIX 1 (continued)

DC5: 24.3.67

- 05.34 Calf down, then up one minute later.
- 05.39 Calf down; cow eating clean straw.
- 05.48 Cow down, licking herself one foot away from calf. Calf sleeping.
- 05.50 Cow resting.
- 06.15 Observation ended.

APPENDIX I (continued)

DC6: 30.4.67

Dam: Large Ayrshire cow. Quiet. Shape = poor, but cow tall.

Calf: Ayrshire bull; average size.

10.00 Calved. Calf born to hips while cow recumbent, then dropped as cow attempted to rise.

10.02 Calf (in twisted pose) struggling weakly. Cow seemed surprised. Quiet, giving only an occasional grunt. Gave calf an occasional lick but not really interested in it.

10.06 Calf shivering a great deal.

10.09 Calf struggled powerfully at this point but ended up lying on back with head and neck twisted below it.

10.10 Calf finally (due to own struggling) in lateral recumbency.

10.11 Calf rose into sternal recumbency. Shivering, snuffing, head shaking.

10.15 Calf moving much more and cow showing more interest in calf. Grunting a lot but only licking it occasionally.

10.27 A few attempts made by calf to rise.

10.45 At this point, nothing further had happened. Cow was just standing over calf and calf not really making any attempt to rise.

11.11 Calf attempted to rise.

11.14 Calf kneeling. Cow licking calf's perineum.

11.15 Calf up but immediately fell.

11.16 Calf up and cow moved forward to lick it (this was the first walking the cow had done since calving).

11.17 Calf down (slumped).

11.19 Calf up.

11.20 Calf fell and stayed down. Cow straining a little on placenta.

11.48 Calf up. Cow grunting to it.

11.50 Calf fell and stayed down; looking alert and lying with head up but shivering.

APPENDIX 1 (continued)

DC6: 30:4.67

- 12.01 Cow down, two feet from calf, resting.
- 12.15 Cow grunting and calf bleating for the first time. Then calf up (easily). Cow grunting and licking calf.
- 12.17 Calf pushing at cow's neck and then blundering about box (not too steady). Cow was much more interested in calf by this time but has never shown any interest in contaminated bedding, amnion, etc.
- 12.23 Cow up, grunting.
- 12.29 Calf down, cow licking it.
- 12.55 Calf up and wandering (much stronger).
- 12.58 Calf down but up straight away.
- 13.00 Calf pushed cow's brisket for a moment then wandered off.
- 13.01 Calf knelt down, then stood up again.
- 13.03 Calf pushing at right udder but too high for teats; cow standing well. Then calf made continual suckling movements into the air.
- 13.05 Calf suckling at cow's left axilla. Cow grunting, then drinking.
- 13.08 Calf pushing right udder, again too high. Then pushing right axilla. Then calf circled cow, vigorously pushing at her body, tail wagging and bunting.
- 13.14 Calf down. Cow eating hay.
- 13.16 Cow down.
- 13.28 Suddenly, calf up and pushing first box wall, then cow's neck. Cow grunting.
- 13.34 Calf knelt then stood up again.
- 13.40 Calf down, sleeping. Cow licking calf, then cuddling.
- 15.01 Calf (easily) up; circling still recumbent cow.
- 15.02 Cow up (grunting) and cleansed on rising. Smelt contaminated bedding but was not too interested. Calf pushing at cow's brisket again.

APPENDIX 1 (continued)

DCG: 30.4.67

- 15.03 Cow started eating placenta. Calf down (fell under cow), but soon up again. First started pushing cow's underbelly, then circled cow, pushing at cow's body, then passed by cow and started pushing wall.
- 15.15 Calf pushing determinedly at left of udder.
- 15.19 Calf down.
- 15.25 Cow stopped eating placenta after eating about 1/3 of it; eating hay.
- 15.45 Cow down, almost on top of calf. Calf (startled) up, circling cow. Cow licking calf, occasionally, but still eating hay.
- 15.54 Calf down next to cow. Calf's coat looks sleeked down because it was never really well licked.
- 16.47 Cow and calf (startled) up. Calf pushing at cow's left udder; this time at teat level. Nibbled both L.A. and L.P. teats but difficulty in getting teat end into mouth. Cow attempting to eat placenta again.
- 16.54 Calf finally suckling L.A. teat and appeared to be getting milk for first five minutes although quarter looked empty after about three minutes.
- 17.16 Cow left off eating placenta. Calf persisted in suckling L.A. teat but did not appear to be getting any milk.
- 17.27 Calf passing meconium.
- 17.28 Calf pushing cow's right axilla for a moment, then lay down (easily).
- 17.30 Cow eating hay.
- 17.37 Cow down, three feet from calf.
- 18.00 Observation ended.

APPENDIX 1 (continued)

DCZ: 28.5.67

- Dam: Mature Ayrshire cow. Shape - poor, but not excessively so (very large udder, but reasonably well tucked up).
- Calf: Small, strong Ayrshire bull.
- 02.20 Calved. Calf born to hips while cow down, then dropped as cow climbed to feet. Immediately calf began head shaking, sneezing and soon after, bleating. After calving cow immediately turned and began licking calf roughly; loudly grunting at it.
- 02.48 Calf rose into sternal recumbency. Cow still very attentive.
- 03.01 Calf (had been lying still, although did not appear to be weak) started attempting to stand.
- 03.21 After several attempts, calf up but very unsteady. Cow had not ceased licking calf at this point.
- 03.23 Calf still very unsteady and fell over backwards when attempted to walk. Cow very excited, licking.
- 03.27 Calf up, teat seeking but cow backing away from calf, still licking its head.
- 03.28 Calf fell. Cow eating contaminated bedding. Calf tired looking and shivering.
- 03.29 Calf making suckling movements into the air.
- 03.35 Calf up (easily), then fell. Cow licking calf and grunting.
- 03.37 Calf up.
- 03.42 Calf fell.
- 03.46 Calf up. Cow very excited again. Grunting at and licking calf. Calf making suckling movements into the air and wandering around the box. Then pushing at corner of box. Cow eating contaminated straw.
- 03.52 Calf pushing at left udder for a moment, then moved on to wall again. Later calf pushing to right of cow's udder. Cow drinking.
- 03.56 Calf pushing cow's left axilla.
- 04.01 Calf pushing cow's right axilla, then the underbelly from the left.
- 04.10 Calf pushing udder from behind cow, and suckling udder skin and point of cow's hock. Then pushing at left axilla.

APPENDIX I (continued)

DC7: 28.5.67

- 04.13 Calf pushing on right side of cow. Calf pushing at right udder but too high for teats. Finally sucking cow's fibia.
- 04.16 Calf pushing right udder at teat level and very soon suckling R.A. teat. Cow licking calf's perineum. Calf then suckled R.P. teat, then L.P. from behind (on two occasions.). At one point calf gave up suckling to push at cow's left axilla. Suckling finally for a total of 17 minutes after which time calf was very distended.
- 04.45 Calf running around box, fell but up immediately.
- 04.46 Cow drinking.
- 04.50 Calf again appeared interested in suckling and finally suckled R.P. teat for $\frac{1}{2}$ minute. During this time, passed meconium which was licked up by the cow.
- 04.52 Passed meconium again and cow seemed very keen to lick this up.
- 05.00 Calf very tired but found it difficult on several occasions to lie down.
- 05.01 Cow down; calf wandering.
- 05.10 Many attempts by calf again to lie down. Spent much of time kneeling.
- 05.11 Calf down, two feet from cow.
- 05.39 Cow (grunting) up.
- 05.50 Cow down, two feet from calf.
- 06.20 Cow started cudding.
- 06.38 Calf up, then fell; seemed very tired.
- 06.42 Calf up again and wandering. Cow immediately up. Calf running, bucking, etc., then weakly pushing at some straw bales.
- 07.01 Cow drinking; calf wandering.
- 07.11 Cow down.
- 07.26 Cow up.
- 08.01 Calf down. Cow licking calf at first, then standing and cudding.

APPENDIX I (continued)

DC7: 28.5.67

- 09.20 Calf up, stretching. Cow licking calf's perineum but calf not making teat seeking advances, just standing quietly.
- 09.29 Calf down (easily). Cow cuddling.
- 09.53 Cow down, three feet from calf, cuddling.
- 10.20 Observation ended.

APPENDIX 1 (continued)

DE8: 30.5.67

- Cow: Ayrshire. Young adult (not heifer). Shape - very good for suckling but not a good one for dairy cow.
- Calf: Ayrshire-cross-Friesian, heifer, strong and small.
- 14.15 Calved (calf dropped in usual way from the standing position after being delivered to hips by recumbent cow).
- 14.18 Calf rose into sternal recumbency. Cow licking calf well and grunting occasionally.
- 14.27 Calf making several attempts to rise. Cow not licking calf at all at this point except when calf fell. Cow occasionally grunting to calf.
- 14.29 Cow eating contaminated bedding but went back to calf whenever it struggled. Calf head shaking and snuffling, bright and strong.
- 14.31 Calf attempting to rise for a few minutes, then resting.
- 14.50 Cow down, cuddling, four feet from calf. Calf making suckling motions in the air.
- 15.07 Cow up, licking calf. Calf up for a second, then accidentally knocked over by cow.
- 15.10 Calf up for a second, then accidentally knocked over by cow. This cow seemed quite interested in the calf's umbilicus. Several times, cow's licking has knocked calf over.
- 15.20 Calf standing; cow licking it.
- 15.23 Calf down, then immediately up. Then up and down several times, finally down.
- 15.25 Cow down, two feet from calf.
- 15.30 Calf (easily) up followed by cow. Pushing wall. Calf down and up again, then knocked over by cow.
- 15.33 Calf pushing above left udder. Cow standing well. Calf pushing left underbelly.
- 15.37 Calf pushing left udder at teat level.
- 15.40 Calf pushing right udder at teat level. Bunting and tail wagging. Calf pushing at left xiphoid area.

APPENDIX I (continued)

DCR: 30.5.67

- 15.49 Calf suckling L.A. teat for 5½ minutes. Cow licking calf's anus. Calf sucking cow's left hock. Calf suckling L.A. teat again for two minutes.
- 15.57 Suddenly cow excited, grunting, circled away from calf. Calf then wandered off.
- 16.00 Calf suckling L.A. teat on and off for a total of 10½ minutes.
- 16.14 Calf attempting to suckle on right side now. Calf then wandering, running and bucking.
- 16.19 Calf slumped down; cow down two feet from calf.
- 17.20 Calf up.
- 17.23 Cow up, strained once and cleansed. Eating placenta.
- 17.25 Calf suckling L.A. on and off for a total of eleven minutes. (Some of the time, the L.A. must have been empty though.)
- 17.50 Calf down.
- 18.09 Calf up and suckling for a few minutes at the L.A. teat (must be empty).
- 18.14 Calf off teat and slumped down. Coughing a little.
- 18.15 Calf up. Cow finished eating placenta. Calf (very distended) slightly dyspnoeic.
- 18.16 Cow eating contaminated straw. Calf running and jumping and then suckling L.A. teat for a few seconds.
- 18.19 Then calf up and down several times. Calf then up and pushing xiphoid and axilla for a second.
- 18.24 Cow down, calf initially wandering, then down.
- 18.29 Cow up and cuddling, straining a little. Calf up.
- 18.32 Suckling for a few minutes, again on L.A. teat (must have been empty). Calf wandered around to cow's right side but would not suckle on that side. Calf coughing again; cow eating straw.
- 19.09 Cow down; calf down.

APPENDIX I (continued)

DCR: 30.5.67

- 19.50 Cow up and eating hay.
- 20.03 Calf suddenly grunting loudly and licking calf. Calf immediately rose, stretching.
- 20.05 Calf suckling L.P. teat for total of 11 minutes. Cow licking away meconium from calf's perineum.
- 20.18 Calf wandered away, running, bucking, etc.
- 20.27 Calf down, cow eating.
- 20.36 Cow down and quickly into the sleeping position. Cow once or twice up for very short spell over next hour or so, but only to stand quietly, occasionally lick calf or sometimes to eat hay. Calf sleeping all the time.
- 22.15 Observation ended.

APPENDIX 1 (continued)

DC9: 3.6.67

Dam: Fairly old Ayrshire. Quiet. Shape - poor, as large abdomen and very pendulous udder.

Calf: Small Ayrshire-cross-Friesian bull. Strong.

19.31 Calved. Quick, easy labour. Calf born to hips while cow recumbent, then calf dropped while cow was getting up. Cow noisy, keen, rough licking immediately. Calf head shaking and snuffling from birth.

19.40 Calf rose into sternal recumbency. Cow started licking calf's head.

19.45 Calf attempting to rise.

19.47 Calf kneeling, then fell.

19.51 As above.

19.54 Calf up, but fell one minute later. Cow still licking calf.

20.04 Calf lying quietly. Cow (still grunting at calf) beginning to eat contaminated bedding.

20.07 Directly calf started trying to rise again, cow very excited, rushed over blaring and started to lick again.

20.13 Calf up (easily), walking straight away towards cow. Calf pushing cow's left axilla. Calf fell, up immediately.

20.15 Calf circling cow, pushing her underbelly, xiphoid, finally centring on the left udder. Calf pushing above teat level, pushing into groin. Later pushing at cow's brisket. Cow occasionally licking calf's perineum, but mainly interested in contaminated bedding.

20.17 Calf fell and got up several times over next few minutes.

20.21 Calf pushing underbelly of cow from either side until fell and stayed down.

20.32 Cow down, two feet from calf, occasionally grunting and licking calf's head.

20.42 Cow (grunting) up, licking calf.

20.49 Calf easily up, teat seeking. Calf circling cow and continuously pushing at cow's body. Cow grunting.

21.00 Although calf had L.A. teat in its mouth for a second, it almost always either pushed too high or anteriorally.

APPENDIX 1 (continued)

DC9: 3.6.67

- 21.01 Calf attempting to lie down.
- 21.02 Calf down.
- 21.05 Cow drinking.
- 21.13 Cow down, two feet from calf, occasionally licking it.
- 21.24 Cow (grunting) up, licking calf. Calf sleeping.
- 21.27 Calf up. Immediately started teat seeking again, but once more pushing too high or anteriorally.
- 21.28 Calf down.
- 21.34 Cow down, one foot from calf.
- 21.40 Cow (grunting) up, licking calf.
- 21.47 Calf up but keen to lie down again.
- 21.52 Calf down; cow eating clean straw.
- 21.57 Cow down, straining a little on placenta.
- 22.02 Cow up, worried by placenta, eating contaminated bedding.
- 22.33 Calf up. Cow very noisy and excited. Calf pushing cow's left side for a few moments, then wandering around box.
- 22.35 Cow drinking.
- 22.38 Calf pushing left axilla and above udder on left.
- 22.40 Calf (with difficulty) down. Cow eating straw.
- 22.42 Calf up, pushing cow's left side, then cow's right groin.
- 22.54 Calf down (easily).
- 23.11 Cow cleansed following two strains and immediately started eating placenta.
- 23.36 Calf up, pushing left udder, but still above teats, then nose into cow's groin. Cow finished eating placenta, eating contaminated straw.

APPENDIX 1 (continued)

DC9: 3.6.67

23.42 Calf down.

24.00 Cow down, one foot from calf, cuddling. Milk dripping from both hindquarters.

00.11 Calf up; cow grunting.

00.20 Calf down again.

00.35 Cow up for drink, then down again in same position.

00.50 Cow up.

00.54 Calf up, pushing at right udder, initially at correct level but later, as it became more excited, higher up flank.

01.00 Calf pushing xiphoid from left, then cow's udder from behind.

01.16 Cow down; calf wandering around box.

01.19 Calf down (tired).

01.20 Calf sleeping. Cow cuddling.

01.45 Cow resting now (no longer cuddling). Calf sleeping.

02.52 Calf up, quickly, followed by cow (grunting). Cow licking calf's perineum as calf wearily teat seeking again. Still pushing too high for teats or anteriorally. Cow standing well.

03.11 Calf wandering around box.

03.14 Calf down (tired). Cow drinking again.

03.20 Cow down two feet from calf.

03.31 Observation ended.

APPENDIX I (continued)

DE10: 27.6.67

- Dam: Ayrshire, mature adult, quiet. Shape - very poor, large belly and pendulous udder.
- Calf: Ayrshire bull; very large.
- 17.26 Calved; calf born to hips from recumbent position, then dropped as cow rose.
- 17.32 Calf still lying still in lateral recumbency. Dyspnoeic, with a loud respiratory rattle.
- 17.39 Calf rose into sternal recumbency.
- 17.42 Calf struggling to rise, soon shuffling on knees around the box. All this time, the cow had been gently licking calf and softly grunting to it.
- 18.00 Cow down two feet from calf. Calf still not up. Had shuffled on knees into the opposite corner of the box to where it had been born. Cow still occasionally licking calf.
- 18.28 Calf trying to rise again.
- 18.36 Calf lying still, appears tired already.
- 18.40 Calf up on second attempt. Teat seeking; cow licking and grunting.
- 18.55 Calf fell and stayed down. Cow eating contaminated bedding.
- 19.05 Calf up (easily). Teat seeking and probing cow's underbelly.
- 19.12 Calf still teat seeking, but spending plenty of time at cow's brisket and xiphoid. When it strays back to the cow's udder, it pushes too high.
- 19.20 Calf still teat seeking, but spending plenty of time at cow's brisket and xiphoid. When it strays back to the cow's udder, it pushes too high.
- 19.30 Calf still teat seeking, but spending plenty of time at cow's brisket and xiphoid. When it strays back to the cow's udder, it pushes too high. Cow still standing very well. Calf very tired.
- 19.40 Calf standing still and being licked down by cow. No longer teat seeking.
- 19.45 Very gently, calf pushing cow's right underbelly.

APPENDIX I (continued)

DC10: 27.6.67

- 19.50 Cow occasionally licking calf. Calf mostly standing by cow's shoulder doing nothing. Occasionally calf wanders around the box.
- 19.54 Cow eating contaminated straw.
- 20.03 Calf wandering. Cow drinking.
- 20.04 Calf wandering, then pushing at corner of box.
- 20.30 Calf occasionally making weak attempts to push at cow's udder, but is far too high up.
- 20.38 Cow straining on placenta.
- 20.48 Cow cleansed, then slowly turned and started eating it. Milk dripping from all four quarters of the cow's udder.
- 21.23 Cow having much difficulty in eating placenta. Calf just standing quietly nearby.
- 21.29 Calf down.
- 22.00 Cow finished cleansing, then stood quietly for some time, just giving the occasional strain.
- 22.20 Cow eating clean straw and hay.
- 22.36 Cow down, three feet from calf.
- 23.24 Calf (bleating) suddenly rose and stretched. Then stood for a second, then started wandering around the box. Whenever it neared the cow, she licked it.
- 23.33 Calf down.
- 00.26 Calf up, wandering, etc.
- 00.30 Cow up, calf pushing at her underbelly in a weak manner. Calf concentrating on anterior underbelly.
- 00.35 Calf pushing cow's brisket from the front.
- 00.36 Calf drooling saliva. Pushing udder but too high.
- 00.37 Calf wandering around box. Very dull, calf pushing at cow's xiphoid.
- 00.42 Calf still pushing at xiphoid. Cow eating hay. It sounded just as if calf was suckling, but calf was on sucking cow's skin.

APPENDIX 1 (continued)

DC10: 27.6.67

- 00.50 Calf wandering.
- 00.53 Calf sucking cow's dewlap then wandering again. This behaviour
 (i.e. wandering and weakly pushing especially around cow's
 forelegs) kept up until
- 01.25 Observation ended.

APPENDIX I

Table 1

Data Regarding the Ten Beef Cows Used in the Investigation (Part I, Sections I, II and III)
Into Natural Suckling in Cattle During the First Eight Hours Post Partum

Dam	Dam Shape	Time of Calving (hours)	Dam's Position at Calving	Time Taken to First Lick Calf	Duration of Initial Calf Licking (mins.)	Time Taken (after calf rose) to Stand to Calf's Advances (mins.)	Time Taken to Cleanse (mins.)
BC1	Good	00.25	Stood to complete act	Initiated immediately	45	3	315
BC2	Good	16.16	"	"	5	3	188
BC3	Good	11.01	Stood throughout	"	10	19	480 - 600
BC4	Poor	11.40	Stood to complete act	"	74	25	182
BC5	Good	12.14	"	"	26	12	172
BC6	Poor	19.45	"	"	56	40	140
BC7	Good	23.45	"	"	20	Immediate	241
BC8	Poor	16.30	Recumbent throughout	"	94	20	480 - 600
BC9	Poor	22.18	Stood to complete act	"	125	13	210
BC10	Poor	19.20	"	"	28	30	406

APPENDIX I

Table 2

Data Regarding the Ten Dairy Cows Used in the Investigation (Part I, Sections I, II and III)
 Into Natural Suckling in Cattle During the First Eight Hours Post Partum

Dam	Dam Shape	Time of Calving (hours)	Dam's Position at Calving	Time Taken to First Lick Calf	Duration of Initial Calf Licking (mins.)	Time Taken (after calf rose) to Stand to Calf's Advances (mins.)	Time Taken to Cleanse (mins.)
DC1	Poor	00.35	Recumbent Throughout	2	17	75	251
DC2	Poor	23.37	Stood to complete act	5	2	12	370
DC3	Poor	02.40	"	Initiated Immediately	35	23	174
DC4	Poor	20.56	Recumbent throughout	$\frac{1}{4}$	33	25	480 - 600
DC5	Poor	22.15	"	7	3	40	311
DC6	Poor	10.09	Stood to complete act	2	Calf licked only occasionally	107	302
DC7	Poor	02.20	"	Initiated Immediately	68	31	480 - 600
DC8	Good	14.15	"	3	9	13	188
DC9	Poor	19.31	"	Initiated Immediately	33	21	220
DC10	Poor	17.25	"	"	35	25	203

APPENDIX I

Table 3

Data Regarding the Ten Dairy Heifers Used in the Investigation (Part I, Sections I, II and III)
into Natural Suckling in Cattle During the First Eight Hours Post Partum

Dam	Dam Shape	Time of Calving (hours)	Dam's Position at Calving	Time Taken to First Lick Calf (mins.)	Duration of Initial Calf Licking (mins.)	Time Taken (after calf rose) to Stand to Calf's Advances (mins.)	Time Taken to Cleanse (mins.)
DH1	Good	14.28	Stood to complete act	Initiated immediately	5	Immediate	272
DH2	Poor	19.47	"	3	9	30	207
DH3	Good	04.33	Recumbent throughout	8	7	58	217
DH4	Good	23.43	"	Initiated immediately	29	7	244
DH5	Good	02.59	"	258	9	232	269
DH6	Good	16.15	Stood to complete act	Never initiated	-	Never stood to calf's advances	480 - 600
DH7	Good	15.00	Recumbent throughout	Initiated immediately	22	14	188
DH8	Good	05.43	Stood to complete act	"	3	Immediate	207
DH9	Good		Recumbent throughout	2	5	59	219
DH10	Good	23.40	Stood to complete act	Initiated immediately	8	2	246

APPENDIX I

Table 4a

Data Regarding the Ten Beef Cows' Calves Used in the Investigation (Part I, Sections I, II and III)
 Into Natural Suckling in Cattle During the First Eight Hours Post Partum

Calf No.	Time Taken to Rise into Sternal Recumbency (mins.)	Time Taken to Make Initial Coordinated Attempts to Rise (mins.)	Time Taken to Stand (mins.)	Test-Seeking Time Prior to First Suckling (mins.)	Test-Seeking Time (Total over 8 Hours) (mins.)
BC1	6	11	23	13	24
BC2	8	15	45	10	11
BC3	1	4	16	12	17
BC4	5	15	26	Calf did not suckle	122
BC5	2	6	26	6	6
BC6	1	4	20	40	46
BC7	32	45	55	15	15
BC8	22	50	65	32	46
BC9	2	3	37	16	26
BC10	8	16	35	Calf did not suckle	100

APPENDIX I

Table 4b

Data Regarding the Ten Beef Cows' Calves Used in the Investigation (Part I, Sections I, II and III)
into Natural Suckling in Cattle During the First Eight Hours Post Partum

Calf No.	Time to First Suckling (mins.)	No. of Suckling Spells over First 8 Hours Post Partum	Total Suckling Time Over First 8 Hours Post Partum (mins.)	Teat First Suckled Successfully	48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
BC1	45	3	28.0	L.A.	24
BC2	58	3	15.0	R.A.	14
BC3	35	4	18.5	L.A.	38
BC4	Calf did not suckle	0	0	-	19
BC5	39	2	21.5	L.A.	7
BC6	180	2	3.5	R.A.	29
BC7	70	2	15.5	L.A.	32
BC8	158	3	7.5	L.A.	24
BC9	66	2	9.0	B.P.	33
BC10	Calf did not suckle	0	0	-	1

APPENDIX I

Table 5a

Data Regarding the Ten Dairy Heifers' Calves Used in the Investigation (Part I, Sections I, II and III) into Natural Suckling in Cattle During the First Eight Hours Post Parturition

Calf No.	Time Taken to Make Initial Coordinated Attempts to Rise (mins.)	Time Taken to Sternal Recumbency (mins.)	Time Taken to Stand (mins.)	Teat-Seeking Time Prior to First Suckling (mins.)	Teat-Seeking Time (Total Over 8 Hours) (mins.)
DH1	2	4	21	Calf did not suckle	97
DH2	4	16	30	16	16
DH3	9	9	99	9	9
DH4	14	22	22	31	32
DH5	6	61	69		
DH6	7	10	31	Calf did not suckle	
DH7	24	56	79	22	29
DH8	5	24	261	21	21
DH9	18	20	45	3	3
DH10	12	39	70	44	44

APPENDIX I

Table 5b

Data Regarding the Ten Dairy Heifers' Calves Used in the Investigation (Part I, Sections I, II and III) Into Natural Suckling in Cattle During the First Eight Hours Post Partum

Calf No.	Time to First Suckling (mins.)	No. of Suckling Spells over First 8 Hours Post Partum	Total Suckling Time Over First 8 Hours Post Partum (mins.)	Teat First Suckled Successfully	48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
DH1	Calf did not suckle	0	0	-	1
DH2	60	3	26.0	L.A.	23
DH3	299	2	17.5	L.A.	20
DH4	104	2	12.0	L.A.	50
DH5	301	2	25.5	L.A.	35
DH6	Calf did not suckle	0	0	-	1
DH7	110	4	27.0	R.A.	29
DH8	346	1	19.0	L.A.	22
DH9	163	1	6.5	R.A.	25
DH10	363	1	4.5	L.A.	4

APPENDIX I

Table 6a

Data Regarding the Ten Dairy Cows' Calves Used in the Investigation (Part I, Sections I, II and III)
into Natural Suckling in Cattle During the First Eight Hours Post Partum

Calf No.	Time Taken to Rise into Sternal Recumbency (mins.)	Time Taken to Make Initial Coordinated Attempts to Rise (mins.)	Time Taken to Stand (mins.)	Teat-Seeking Time Prior to First Suckling (mins.)	Teat-Seeking Time (Total Over 8 Hours) (mins.)
DC1	3	4	52	Calf did not suckle	57
DC2	7	8	43	31	35
DC3	2	10	55	64	64
DC4	4	14	54	105	105
DC5	5	18	77	37	37
DC6	11	27	76	35	35
DC7	8	41	61	20	20
DC8	3	12	65	16	16
DC9	9	14	23	Calf did not suckle	81
DC10	14	17	75	Calf did not suckle	107

APPENDIX I

Table 6b

Data Regarding the Ten Dairy Cows' Calves Used in the Investigation (Part I, Sections I, II and III)
 into Natural Suckling in Cattle During the First Eight Hours Post Partum

Calf No.	Time to First Suckling (mins.)	No. of Suckling Spells Over First 8 Hours Post Partum	Total Suckling Time Over First 8 Hours Post Partum (mins.)	Teat First Suckled Successfully	48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
DC1	Calf did not suckle	0	0	-	3
DC2	170	2	28.5	R.A.	30
DC3	421	1	19.5	L.A.	30
DC4	258	1	7.5	R.P.	36
DC5	358	1	7.0	R.A.	39
DC6	414	1	5.0	L.A.	11
DC7	116	2	17.5	R.A.	49
DC8	91	3	32.5	L.A.	46
DC9	Calf did not suckle	0	0	-	6
DC10	Calf did not suckle	0	0	-	5

APPENDIX 2

APPENDIX 2

Table 1

Data Regarding the Ten Non-Mothered Calves Which Were Allowed to Suckle
Their Own Dams at Fixed Times Post Partum (i.e. at 6 Hours and Again at 12 Hours)

Calf No.	Sex	Breeds	Date of Birth	Birthweight (lbs)	Colostrum Intake (lbs)			48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
					At 6 Hours Post Partum	At 12 Hours Post Partum	Total	
D1	F	A	28-6-67	63.5	5.5	0.5	6.0	6
D2	F	AXF	29-6-67	81.25	4.75	1.0	5.75	8
D3	M	A	6-7-67	57.5	4.0	4.0	8.0	15
D4	F	A	6-7-67	73.0	4.75	4.5	9.25	20
D5	M	A	6-7-67	70.75	5.75	1.0	6.75	24
D6	M	AXF	10-7-67	100.25	4.5	2.25	6.75	13
D7	M	A	10-7-67	90.75	3.25	4.0	7.25	19
D8	F	AXF	12-7-67	89.75	6.75	1.75	8.5	19
D9	M	AXF	15-7-67	62.5	3.0	6.25	9.25	29
D10	F	AXF	16-7-67	100.0	5.5	1.25	6.75	30

* A = Ayrshire

AXF = Ayrshire-cross-Friesian

APPENDIX 2

Table 2

Data Regarding the Ten Mothered Calves Which Were Allowed to Suckle Their Own Dams at Fixed Times Post Partum (i.e. at 6 hours and Again at 12 Hours)

Calf No.	Sex	Breed:	Date of Birth	Birthweight (lbs)	Colostrum Intake (lbs)			48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
					At 6 Hours Post Partum	At 12 Hours Post Partum	Total	
A1	M	AXF	5-2-68	80.5	7.0	3.0	10.0	39
A2	F	AXF	7-2-68	78.0	7.5	3.0	10.5	40
A3	M	AXF	9-2-68	68.25	6.5	1.75	8.25	19
A4	F	AXF	10-2-68	84.0	7.0	3.5	10.5	23
A5	F	A	11-2-68	75.0	6.25	3.0	9.25	36
A6	F	AXF	13-2-68	79.0	3.25	3.5	6.75	27
A7	M	AXF	15-2-68	90.75	3.25	2.25	5.5	36
A8	M	AXF	25-2-68	58.25	5.5	3.0	8.5	28
A10	F	A	9-3-68	70.0	4.75	2.0	6.75	31
A11	F	A	11-3-68	42.5	4.0	3.0	7.0	33

* A = Ayrshire

AXF = Ayrshire-cross-Friesian

APPENDIX 2

Table 3

Data Regarding the Ten Mothered Calves Which Were Allowed to Suckle Their Own Dams at Six Hours Post Partum Only

Calf No.	Sex	Breeds	Date of Birth	Birthweight (lbs)	Colostrum Intake (lbs)		Time Spent Suckling (minutes)	48 Hour Serum Immune Globulin Concentration (Z.S.T. Units)
					At Six Hours	Post Partum (Total)		
A9	M	A	1-3-68	83.0	5.5	5.5	20.0	23
A12	M	AXF	11-3-68	81.25	6.75	6.75	18.0	24
A13	F	AXF	22-3-68	75.75	5.0	5.0	17.5	26
A14	M	A	24-3-68	66.25	5.25	5.25	29.0	32
A15	F	AXF	17-4-68	65.5	2.5	2.5	22.0	15
A16	F	A	23-6-68	66.25	5.0	5.0	13.5	26
A17	M	A	19-7-68	66.25	5.75	5.75	13.0	16
A19	M	A	9-8-68	59.25	7.5	7.5	26.0	30
A20	F	A	9-8-68	57.25	7.5	7.5	27.0	40
A21	M	AXF	12-8-68	64.0	6.75	6.75	11.0	17

* A = Ayrshire
AXF = Ayrshire-cross-Friesian

APPENDIX 3

APPENDIX 3

Table 1

The Total Protein and Immune Lactoglobulin Concentrations of Whey
Samples Prepared from Random Samples Obtained from Colostrum Pool No. 1

<u>Colostrum Whey Sample No.</u>	<u>Total Protein Concentration (gm/100 ml whey)</u>	<u>Immune Lactoglobulin Concentration (gm/100 ml whey)</u>
ST5	10.6	7.49
ST8	10.6	7.27
ST10	10.8	8.12
ST12	11.2	8.05
ST15	10.4	7.60
ST16	10.4	7.60
ST17	10.6	7.48
ST18	10.6	7.48
ST23	10.8	8.18
ST25	9.8	7.14

APPENDIX 3

Table 2

The Total Protein and Immune Lactoglobulin Concentrations of Whey
Samples Prepared from Random Samples Obtained from Colostrum Pool No. 2

Colostrum Whey Sample No.	Total Protein Concentration (gm/100 ml whey)	Immune Lactoglobulin Concentration (gm/100 ml whey)
S4	9.4	6.57
S5	9.4	6.08
S6	9.6	6.64
S9	9.7	6.79
M1	10.0	7.48
M4	10.0	6.50
M5	10.6	7.41
M7	10.2	6.96
M12	10.2	6.93
X5	9.4	6.54
X10	9.6	6.85
C2	9.8	6.56
C3	9.8	6.64
C4	9.6	6.66
C9	9.0	6.21
C10	8.8	6.76

APPENDIX 3

Table 3

The Total Protein and Immune Lactoglobulin Concentrations of Whey
Samples Prepared from Random Samples Obtained from Colostrum Pool No. 3

<u>Colostrum Whey Sample No.</u>	<u>Total Protein Concentration (gm/100 ml whey)</u>	<u>Immune Lactoglobulin Concentration (gm/100 ml whey)</u>
F2	11.0	8.09
F5	12.4	9.40
F10	11.4	8.31
F16	11.1	8.45
F18	11.3	7.83
F19	10.6	8.22
F21	10.9	8.86
F28	11.6	8.94
F30	12.5	9.74
F34	12.0	9.03
F39	12.2	9.17
F40	10.6	7.85

APPENDIX 3

Table 4

Data Regarding the Ten Ayrshire Calves Used in the Initial Investigation (Part 3, Section 11)
into the Possibility of Producing Uniform Immune Lactoglobulin Absorption

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed *	Ambient Temperature (°C)		Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Minimum	Maximum	Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
ST1	F	5-10-67	59.0	1475	17.0	21.5	1	9	8
ST3	M	10-10-67	70.25	1756	17.0	23.5	0	10	10
ST11	F	19-10-67	75.5	1888	16.8	22.8	1	13	12
ST14	F	21-10-67	75.5	1888	16.5	21.0	0	19	11
ST15	M	25-10-67	86.5	2163	17.5	21.0	0	10	10
ST17	M	29-10-67	78.25	1956	19.0	22.5	1	14	13
ST20	M	31-10-67	88.5	2213	19.0	22.0	1	12	11
ST21	M	3-11-67	64.5	1613	19.0	23.5	2	15	13
ST23	F	13-11-67	65.75	1644	16.8	22.5	0	13	13
ST24	F	6-11-67	65.0	1625	17.5	21.3	1	10	9

* Colostrum Pool No. 1

APPENDIX 3

Table 5

Data Regarding the Ten Ayrshire-cross-Friesian Calves Used in the Initial Investigation
(Part 3, Section II) into the Possibility of Producing Uniform Immune Lactoglobulin Absorption

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed (ml)	Ambient Temperature (°C)		Serum immune Globulin Concentration (Z.S.T. Units)	Postcolostral (48 hours post partum)	48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Minimum	Maximum			
ST2	F	8-10-67	63.25	1581	16.0	21.5	1	17	16
ST4	F	10-10-67	65.75	1644	18.0	22.5	1	15	14
ST5	M	10-10-67	80.5	2013	17.8	21.5	1	16	15
ST6	F	11-10-67	78.5	1963	16.0	20.7	2	16	14
ST7	M	14-10-67	75.25	1881	16.5	22.0	1	15	14
ST8	M	17-10-67	71.0	1775	19.0	23.0	1	14	13
ST16	F	25-10-67	72.25	1806	18.0	22.0	1	19	18
ST18	M	30-10-67	71.25	1781	19.0	22.0	2	12	10
ST19	M	30-10-67	76.5	1913	19.0	21.5	1	23	22
ST22		3-11-67	73.75	1969	17.0	21.5	1	14	13

*: Colostrum Pool No. 1

APPENDIX 3

Table 6

Data Regarding the Ten Ayrshire Calves (Controls) Used in the Investigations (Part 3, Sections II and III) into the Effects of Low Ambient Temperature, Nourishing and Corticosteroid Therapy on Immune Lactoglobulin Absorption

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed *	Ambient Temperature (°C)		Serum Immune Globulin Concentration (Z.S.T. Units)	Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Minimum	Maximum				
S1	F	15-11-67	70.5	1763	17.0	20.4	1	11	10	
S2	F	16-11-67	56.0	1409	17.5	20.2	1	12	11	
S3	F	17-11-67	65.25	1631	17.9	20.2	0	14	14	
S4	M	18-11-67	59.25	1481	18.5	20.3	1	13	12	
S5	F	18-11-67	86.5	2163	18.5	20.3	1	8	7	
S6	F	20-11-67	61.0	1525	18.0	20.0	0	8	8	
S7	M	21-11-67	71.25	1781	17.0	18.7	1	13	12	
S8	M	23-11-67	77.5	1938	18.2	20.4	1	12	11	
S9	F	29-2-68	57.5	1438	20.0	21.2	1	7	6	
S12	M	5-3-68	80.25	2006	18.5	20.3	2	14	12	

* Colostrum Pool No. 2

APPENDIX 3

Table 7

Date Regarding the Ten Ayrshire Calves Used in the Investigation (Part 3, Section II) into the Effect of Low Ambient Temperature on Immune Lactoglobulin Absorption

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed (ml)	Ambient Temperature (°C)		Serum Immune Globulin Concentration (Z.S.T. Units)	Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Minimum	Maximum				
C1	M	31-1-68	83.75	2094	+1.5	+4.75	2	15	13	13
C2	M	4-2-68	47.5	1188	-2.0	+5.0	1	14	13	13
C3	M	4-2-68	54.0	1350	-2.0	+5.0	1	10	9	9
C4	F	16-2-68	71.5	1788	+2.0	+5.5	1	11	10	10
C6	M	10-11-68	89.25	2231	-9.0	+2.0	1	10	9	9
C7	F	13-11-68	62.5	1563	+0.5	+1.0	1	11	10	10
C8	M	14-11-68	68.0	1700	+0.5	+2.5	1	12	11	11
C9	M	14-11-68	67.25	1681	+0.5	+2.5	0	12	12	12
C10	F	17-11-68	73.25	1831	-5.7	+4.7	1	10	9	9
C11	F	19-11-68	75.5	1888	0.00	+4.5	1	12	11	11

* Colostrum Pool No. 2

APPENDIX 2

Table 8

Data Regarding the Ten Ayrshire Calves Used in the Investigation (Part 2, Section III)
into the Effect of Nourishing upon Immune Lactoglobulin Absorption

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
M1	M	23-2-68	82.5	2063	2	18	16
M3	M	7-3-68	74.25	1856	2	27	25
M4	M	9-3-68	58.75	1469	1	21	20
M6	M	15-3-68	61.5	1528	2	21	19
M7	F	15-3-68	79.25	1981	3	19	16
M8	F	21-3-68	84.5	2113	1	15	14
M9	F	22-3-68	59.75	1494	1	15	14
M10	F	24-3-68	67.5	1688	1	19	18
M11	F	24-3-68	72.25	1806	1	20	19
M12	M	26-3-68	70.0	1750	1	17	16

* Colostrum Pool No. 2

APPENDIX 3

Table 2

Data Regarding the Ten Ayrshire Calves Used in the Investigation (Part 3, Section III)
into the Effect of Corticosteroid Administration on the Absorption of Immune Lactoglobulin

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
X1	F	1-2-68	76.25	1906	3	18	15
X2	F	7-2-68	74.5	1863	4	16	12
X3	F	15-2-68	49.75	1244	5	17	12
X4	F	15-2-68	53.0	1325	4	19	15
X5	M	19-3-68	68.5	1713	2	18	16
X6	F	20-3-68	72.0	1800	1	17	16
X7	F	20-3-68	79.75	1994	1	17	16
X8	F	20-3-68	65.75	1644	1	18	17
X9	M	28-3-68	71.25	1781	2	14	12
X10	M	29-3-68	60.25	1506	2	19	17

* Colostrum Pool No. 2

APPENDIX 3

Table 10

Data Regarding the Twenty Avyrshire Calves Used in the Investigation (Part 3, Section IV) into the Relationship Between the Immune Lactoglobulin Concentration of a Dam's Colostrum and the Resulting Serum Concentrations of Absorbed Immune Lactoglobulin of a Calf Fed that Colostrum

(Details of the colostrum fed to the calves in this experiment are presented in Appendix 4, Table 1)

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed :- (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
Q1	F	16-4-68	82.75	2069	2	25	23
Q2	M	19-4-68	69.0	1725	2	4	2
Q3	M	23-4-68	69.5	1738	1	9	8
Q4	F	23-4-68	78.75	1969	1	12	11
Q5	M	25-4-68	71.5	1788	1	11	10
Q11	M	8-5-68	65.0	1625	1	11	10
Q12	M	10-5-68	67.75	1694	1	17	16
Q13	M	10-5-68	68.25	1706	1	9	8
Q14	F	10-5-68	76.25	1906	1	31	30
Q15	F	11-5-68	54.75	1369	1	28	27

APPENDIX 3 - Table 10 (continued)

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.I. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.I. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
Q16	M	14-5-68	79.75	1994	1	35	34
Q17	M	17-5-68	84.75	2119	1	11	10
Q18	M	24-5-68	70.75	1769	1	18	17
Q19	M	26-5-68	71.0	1775	2	27	25
Q20	M	26-5-68	78.5	1963	2	28	26
Q21	F	27-5-68	65.0	1625	2	17	15
Q22	M	29-5-68	78.25	1956	1	23	22
Q23	M	31-5-68	75.0	1875	6	38	32
Q24	F	4-6-68	72.5	1213	0	9	9
Q25	F	5-6-68	39.25	981	6	17	11

* All of these calves were fed their own dam's colostrum at the standard rate (as in the other experiments) of 25 ml/lb birthweight.

APPENDIX 3

Table 11

Data Regarding the Ten Ayrshire Calves Used in the Investigation (Part 3, Section V) into the Effect of Time of First Colostrum Feed on the Absorption of Immune Lactoglobulin (Colostrum feeding carried out at one hour post partum)

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
F2	M	27-5-68	63.5	1588	1	20	19
F6	F	11-6-68	45.0	1125	1	28	27
F8	M	13-6-68	71.75	1794	3	29	26
F10	F	14-6-68	67.5	1688	3	24	21
F13	M	15-7-68	81.5	2031	1	18	17
F18	M	9-8-68	77.5	1938	1	23	22
F21	M	14-8-68	76.75	1919	1	41	40
F27	M	23-8-68	72.25	1806	1	17	16
F29	M	26-8-68	70.0	1750	2	23	21
F33	F	30-8-68	64.25	1606	1	22	21

* Colostrum Pool No. 3

APPENDIX 3

Table 12

Data Regarding the Ten Ayrshire Calves Used in the Investigation (Part 3, Section V)
 Into the Effect of Time of First Colostrum Feed on the Absorption of Immune Lactoglobulin
 (Colostrum feeding carried out at five hours post partum)

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
F1	F	25-5-68	83.75	2094	1	17	16
F4	M	7-6-68	84.5	2113	2	21	19
F5	F	10-6-68	71.5	1788	2	21	19
F7	M	12-6-68	106.0	2650	1	23	22
F12	M	24-6-68	70.25	1756	1	24	23
F19	M	14-8-68	63.25	1581	0	10	10
F20	F	14-8-68	50.75	1269	1	23	22
F25	M	20-8-68	74.5	1863	1	15	14
F26	F	22-8-68	74.0	1850	1	20	19
F28	M	25-8-68	66.5	1663	0	13	13

* Colostrum Pool No. 3

APPENDIX 3

Table 13

Data Regarding the Ten Avrshire Calves Used in the Investigation (Part 3, Section V)
 into the Effect of Time of First Colostrum Feed on the Absorption of Immune Lactoglobulin
 (Colostrum feeding carried out at nine hours post partum)

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed * (ml)	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum	Postcolostral (48 hours post partum	
F9	M	13-6-68	79.5	1993	5	19	14
F11	M	23-6-68	71.5	1788	1	12	11
F14	M	6-8-68	80.5	2013	2	14	12
F15	F	8-8-68	61.25	1530	3	17	14
F16	F	8-8-68	69.0	1725	5	18	13
F22	F	18-8-68	71.75	1734	0	10	10
F23	F	20-8-68	72.0	1800	1	14	13
F24	M	24-8-68	90.25	2256	3	11	8
F30	F	26-8-68	76.5	1913	3	17	14
F32	M	29-8-68	68.5	1713	4	20	16

* Colostrum Pool No. 3

APPENDIX 3

Table 14

Data Regarding the Ten Ayrshire Calves Used in the Investigation (Part 3, Section VI)
into the Effect of Repeated Feeding of Colostrum on the Absorption of Immune Lactoglobulin

Calf No.	Sex	Date of Birth	Birthweight (lbs)	Total Volume of Colostrum Fed *	Serum Immune Globulin Concentration (Z.S.T. Units)		48 Hour Serum Concentration of Absorbed Immune Lactoglobulin (Z.S.T. Units)
					Precolostral (15 mins. post partum)	Postcolostral (48 hours post partum)	
F34	F	1-9-68	53.5	1588	2	22	20
F35	F	2-9-68	57.0	1425	0	20	20
F36	F	2-9-68	58.25	1456	1	20	19
F37	F	4-9-68	76.0	1900	2	24	22
F38	F	5-9-68	71.5	1788	2	17	15
F39	F	5-9-68	66.5	1663	2	24	22
F40	M	5-9-68	61.0	1525	0	18	18
F41	M	6-9-68	58.5	1463	2	25	23
F42	M	6-9-68	75.75	1894	0	16	16
F43	M	6-9-68	68.25	1706	1	19	18

* Colostrum Pool No. 3

APPENDIX 3

Table 15

Changes in the Serum Concentrations of Absorbed
Immune Lactoglobulin During the First 48 Hours of
Life of Ten Non-Mothered Ayrshire Calves Maintained
Under Standard Conditions of Colostrum Feeding and
Management

Calf No.	Serum Immune Globulin Concentrations (Z.S.T. Units)			
	at the Following Times Post Partum (hours)			
	5	9	24	48
S1	3	6	13	10
S2	2	5	7	6
S3	2	8	13	11
S4	7	12	16	14
S5	6	11	17	12
S6	3	7	9	7
S7	4	6	9	8
S8	1	6	9	12
S9	5	8	14	11
S12	4	8	9	12
Mean	3.7	7.7	11.6	10.3
S.D.	± 2.5	± 2.1	± 3.3	± 2.4
S.E.	± 0.8	± 0.7	± 1.0	± 0.8

APPENDIX 3

Table 16

Changes in the Serum Concentrations of Absorbed
Immune Lactoglobulin During the First 48 Hours of
Life of Ten Mothered Ayrshire Calves Maintained
Under Standard Conditions of Colostrum Feeding and
Management

<u>Calf No.</u>	<u>Serum Immune Globulin Concentrations</u> <u>(Z.S.T. Units)</u> <u>at the Following Times Post Partum (hours)</u>			
	<u>5</u>	<u>9</u>	<u>24</u>	<u>48</u>
N1	5	17	20	16
M3	4	15	18	25
M4	4	6	16	20
M6	5	12	18	19
M7	6	12	19	16
M8	5	9	17	14
M9	6	11	14	14
M10	5	14	17	18
M11	4	12	18	19
M12	7	12	13	16
Mean	5.1	12.0	17.0	17.7
S.D.	± 0.9	± 2.9	± 2.1	± 3.1
S.E.	± 0.3	± 0.9	± 0.7	± 1.0

APPENDIX 4

APPENDIX 4

Table 1

The Volume of Colostrum Obtainable at the First Milking
Post Partum of Twenty Non-Premilked Cows and Heifers and
The Total Protein and Immune Lactoglobulin Content of Whey
Samples Prepared from These Colostra

Colostrum Sample No.	Volume Obtained at First Milking (litres)	Colostrum Whey Total Protein Concentration (grm/100 ml)	Colostrum Whey Immune Lactoglobulin	
			Percentage (%)	Concentration (grm/100 ml)
Q1	3.5	15.4	71.6	11.02
Q2	4.5	4.3	52.3	2.25
Q3	16.0	5.7	57.4	3.27
Q4	14.5	5.3	65.1	3.45
Q5	4.25	6.7	57.0	3.82
Q11	13.25	6.7	67.5	4.52
Q12	6.5	9.4	65.6	6.17
Q13	8.0	8.6	60.8	5.23
Q14	5.25	14.6	77.1	11.25
Q15	3.0	13.8	75.7	10.45
Q16	16.0	16.4	75.2	12.34
Q17	3.5	8.0	74.3	5.94
Q18	3.5	12.1	70.7	8.55
Q19	4.5	19.2	68.3	13.12
Q20	6.0	11.3	74.7	8.44
Q21	6.0	10.8	63.2	6.83
Q22	5.0	16.0	74.3	11.89
Q23	4.75	13.4	73.3	9.82
Q24	6.0	6.2	68.2	4.23
Q25	6.5	12.0	66.8	8.02
Mean	7.0	10.8	67.9	7.53
S.D.	± 4.6	± 4.2	± 6.8	± 3.33
S.E.	± 1.0	± 0.9	± 1.5	± 0.7

APPENDIX 4

Table 2

The Carotene, Vitamin A and Immune Lactoglobulin Concentrations of 100
First-Milking Colostrum Samples from Non-Premilked Cows Calving Between
October, 1967 and September, 1968 (Part 4, Sections II and III)

Colostrum Sample No.	Month of Calving	Carotene Concentration (International Units/100 ml Colostrum)	Vitamin A Concentration (International Units/100 ml Colostrum)	Immune Lactoglobulin Concentration (gm/100 ml Colostrum)
1	February	<75	42	7.53
2	"	88.1	100.5	8.33
3	"	303.0	1131	7.54
4	"	<75	14.3	6.58
5	"	<75	72	9.54
6	"	131.3	222.8	7.75
7	"	82.5	75	5.42
8	"	<75	4.5	3.7
9	March	195	119.3	5.15
10	"	232.5	412.5	2.44
11	"	<75	75.0	9.97
12	"	163.5	369	7.69
13	"	193.5	215.3	6.90
14	"	91.5	1405.5	6.29
15	"	100.5	856.5	5.97
16	"	<75	43.5	7.09
17	"	<75	32.3	9.09
18	"	75	1.5	12.15
19	"	82.5	127.5	11.59
20	"	108.8	420.8	9.52
21	"	<75	34.5	7.26
22	"	91.5	85.5	10.05
23	April	<75	66.0	2.14
24	"	160.0	143.5	3.31
25	"	<75	33.0	3.56
26	"	<75	117.0	3.75
27	"	315	2892	9.75
28	May	131.3	1400.3	16.60
29	"	82.5	160.0	4.22
30	"	148.5	132.8	5.07
31	"	131.3	76.5	4.43
32	"	<75	104.3	9.82
33	"	307.5	276	8.13
34	"	375	331.5	8.13
35	"	<75	70.5	10.79
36	"	<75	24.0	6.28
37	"	1072.5	1450.5	6.46
38	"	146.3	142.5	9.88
39	"	862.5	178.5	6.11
40	"	1125	1713	6.70

APPENDIX 4 - Table 2 (continued)

Colostrum Sample No.	Month of Calving	Carotene Concentration (International Units/100 ml Colostrum)	Vitamin A Concentration (International Units/100 ml Colostrum)	Immune Lactoglobulin Concentration (gm/100 ml Colostrum)
41	May	922.5	2788.5	7.30
42	"	1200	3984	5.41
43	"	435	108.8	8.16
44	"	180	1635.0	9.20
45	"	547.5	1590.0	8.90
46	"	258.8	589.5	5.64
47	"	<75	18.0	5.89
48	"	292.5	195.0	9.06
49	"	<75	64.5	5.87
50	"	697.5	1458	6.85
51	"	450	1226.3	7.42
52	June	525	1622.3	8.17
53	"	408.8	436.5	2.99
54	"	960	3054	6.03
55	"	615	1056	9.41
56	"	585	1419.8	8.55
57	"	103.5	33.8	6.02
58	"	133.5	372.8	5.26
59	"	810	1857	6.16
60	August	225	86.3	6.53
61	"	225	86.3	4.44
62	"	110.3	52.5	4.87
63	"	825	2005.5	5.46
64	"	435	980.5	6.01
65	"	840	3279	6.23
66	"	195	447	6.44
67	"	90	20.3	4.29
68	"	202.5	82.5	3.07
69	"	690	1032	9.55
70	"	307.5	763.5	7.70
71	"	150	430.5	9.29
72	September	810	2532	5.40
73	"	367.5	1611	7.97
74	"	85.5	33.8	5.09
75	October	1125	2613	5.48
76	"	862.5	763.5	5.34
77	"	1050	2577.8	5.84
78	"	862.5	1926	6.28
79	"	712.5	571.5	5.87
80	"	1125	1293	10.03
81	"	1012.5	1383	5.87
82	"	1185	1512	6.80
83	"	465	519	3.60
84	"	1162.5	4888.5	8.74
85	"	251.3	238.5	4.22

APPENDIX 4 - Table 2 (continued)

Colostrum Sample No.	Month of Calving	Carotene Concentration (International Units/100 ml Colostrum)	Vitamin A Concentration (International Units/100 ml Colostrum)	Immune Lactoglobulin Concentration (gm/100 ml Colostrum)
86	October	397.5	171.8	9.63
87	"	547.5	255	6.41
88	"	322.5	698.3	7.67
89	November	300	382.5	6.16
90	"	1050	2877.7	4.40
91	"	510	595.5	8.83
92	"	108.8	117.8	5.47
93	"	585	1097.3	6.64
94	"	990	1834.5	6.01
95	"	622.5	0	5.57
96	"	1087.5	1785.7	6.40
97	"	465	1149	9.22
98	"	315	672.7	6.43
99	"	765	719.2	9.32
100	"	110.6	105	5.11