THE FEEDING OF FERMENTED COLOSTRUM TO NEONATAL CALVES

by

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A THESIS

SUBMITTED TO THE FACULTY OF GRADUATE STUDIES AND RESEARCH IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE

DEGREE OF

DOCTOR OF PHILOSOPHY

CRAMPTON NUTRITION LABORATORY,
DEPARTMENT OF ANIMAL SCIENCE,
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P.Q., CANADA

SPRING, 1983

ABSTRACT

Doctor of Philosophy Animal Science (Nutrition)

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THE FEEDING OF FERMENTED COLOSTRUM TO NEONATAL CALVES

The possibility of improving the nutritive value of fermented colostrum for neonatal calves was investigated in 5 trials involving 164 Holstein calves and in 2 laboratory experiments.

The inoculation of colostrum with *S. lactis* culture ensured a highly palatable and more uniform product. Calves fed the inoculated product gained in weight quicker and consumed more starter than those fed naturally fermented colostrum.

The addition of sorbic acid to *S. lactis* inoculated colostrum controlled the mold contamination, particularly during hot summer weather. This lowered the degradation of lactose and protein and extended the storage life. The application of 1,000 ppm of sorbic acid during the first 4 days of storage is, therefore, recommended.

An undesirably narrow ratio of Digestible Energy/Digestible Protein in colostrum was widened through the use of a supplementary energy source. These treatments also improved the performance of calves.

RESUME

Ph.D. Nutrition

LUMIR A. DREVJANY

DISTRIBUTION DE COLOSTRUM FERMENTE A DES VEAUX EN PERIODE POST-NATALE

La possibilité d'améliorer la valeur nutritive du colostrum fermenté pour des veaux en période post-natale a été étudiée. Dans ce but, 2 expériences en laboratoires et 5 essais portant sur 164 veaux Holstein ont été effectués.

L'ensemencement du colostrum avec une culture de *S. lactis* a donné un produit plus homogène et ayant une excellente appétence. Les veaux ayant reçu du colostrum ensemencé ont eu une meilleure croissance et ont consommé plus d'aliment début que ceux auxquels on a donné du colostrum fermenté naturellement.

L'addition d'acide sorbique au colostrum ensemencé avec du *S. lactis* a permis de contrôler la contamination pour les moisissures, surtout pendant les chaleurs de l'été. Cette opération a aussi réduit la dégradation du lactose et des protéines et a prolongé la durée de conservation. De ce fait, il est recommandé d'ajouter 100 ppm d'acide sorbique durant les 4 premiers jours.

L'utilisation d'une source d'énergie supplémentaire a permis d'accroître le rapport Energie nette/Protéines digestibles, qui, autrement a une valeur faible et non souhaitable. Ces traitements ont aussi amélioré les performances des veaux.

CLAIMS TO ORIGINAL RESEARCH

1. The first demonstration, as far as the author is aware, that bovine colostrum and transitional milk are energy deficient when fed as the only dietary component to neonatal calves.

2. The first study, as far as the author is aware, which employs inoculation with homofermentative *S. lactis* in addition to mold and yeast inhibition by sorbic acid to increase the residual glucogenic energy pool in fermented colostrum, and direct energy supplementation to widen the DE:DP ratio in fermented colostrum to level required by neonatal calves.

3. The first report, as far as the author is aware, to assess the effect of sorbic acid application on residual content of nutrients, control of molds and yeasts and visual characteristics and odour of fermented colostrum. Similarly the effects of dose, time of application of sorbic acid and storage temperature on nutritional value of fermented colostrum during storage were determined.

4. The data obtained are the first, as far as the author is aware, to indicate that improved gains of calves, consuming colostrum treated with sorbic acid, do not reflect "growth promoting properties" of sorbic acid, but can be explained by increased residual level of lactose and protein in sorbic acid treated colostrum.

5. The first demonstration, as far as the author is aware, that supplementation of fermented colostrum with suitable energy source, particularly during the first 3 weeks of the calf's life, when colostrum is the main source of nutrients, will result in higher gains and improved feed conversion. While the digesti-

bility of nutrients in energy supplemented diets was somewhat depressed, the metabolism of protein and consequently its conversion into gain was improved with each increment of DE and widening of DE to DP ratio.

6. The first study, as far as the author is aware, confirming that the energy requirements of neonatal calves on fermented colostrum diets are in agreement with values established by other authors for neonatal calves raised on different milk-product based diets.

Suggested Short Title:

FEEDING FERMENTED COLOSTRUM TO CALVES

L.A. Drevjany

ACKNOWLEDGEMENTS

The author would like to express his sincere appreciation to Dr. E. Donefer, Department of Animal Science, Macdonald Campus of McGill University for his guidance throughout the entire study, and to Dr. L. Latrille, presently of the Department of Animal Science, Laval University, for his helpful advice and interest during all stages of my research.

The author would also like to acknowledge the considerable contribution of Dr. Fanous, Department of Agricultural Chemistry and Physics, Macdonald Campus of McGill University, for help in designing and statistical evaluation of data in the laboratory experiment with sorbic acid.

The author is most grateful for the financial support from the Ontario Ministry of Agriculture and Food.

The moral support and encouragement of members of the Kemptville College of Agricultural Technology Staff, particularly Mr. G.S. Hooper, are gratefully appreciated.

Appreciation is extended to Mr. O.R. Irvine for offering his helpful advice in the area of dairy science, Ms. E. Wheeler and Mr. D.R. McKnight for general assistance in many aspects.

For technical assistance and/or care of experimental animals, I am indebted to N. Woeldike, B. May, D. Thorpe, A. Whaley, G. Earl, D. Gowenlock, M. Seguin, and P. Putnam.

Finally I wish to thank my wife, Eva, for bearing with me.

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CHAPTER 1

GENERAL INTRODUCTION

The average dairy cow produces colostrum[1] and transitional milk[2] in excess of the requirements of the newborn calf. Usually, during the first 8 to 10 milkings postpartum, 50 to 60 kg of this surplus product is accumulated, averaging between 15-16% total solids. Freezing of colostrum, produced over the first two milkings, appears to be justified when preservation of its immunoglobulin content is required. However, when the total quantity of surplus colostrum is to be stored for future use in calf raising, a less expensive method of preservation should be used. Swanack (1971) stored colostrum at ambient temperature and fed it to calves as a fermented product. Natural fermentation, though convenient and inexpensive, leads, particularly during hot summer weather and/or prolonged storage, to a lack of uniformity and palatability of the product. Refusals by calves of naturally fermented colostrum during warm ambient temperatures have been reported by Kaiser (1976), Muller et al (1976), and Rindsig (1976). Initially, some calves fed naturally fermented colostrum required substantial encouragement for one or two feedings (Otterby et al, 1976). Rindsig and Bodoh (1977) observed that varying amounts of colostrum were refused by approximately one-half of a group of calves fed naturally fermented colostrum, and by two-thirds of a group of calves fed colostrum treated with 1% of propionic acid. Most refusals of the propionic acid treated colostrum occurred during the first 7 - 10 days. Similarly, refusals were greater by calves fed an acetic acid treated colostrum, than by calves fed naturally fermented colostrum or milk (Polzin et al, 1977).

[1]Milk secreted for a few days after parturition and characterized by high protein and immune body content.

[2]Transitional milk is the term chosen to characterize that collected during the first 4 days after calving.

During natural fermentation, microorganisms, which are often referred to as heterofermentative, degrade the nutrients in colostrum to a variety of products of questionable nutritive value, and their presence may result in production of toxins (Pelczar et al, 1977). Some of such products are lipogenic by nature (ethylalcohol, ketones, acetic and butyric acids) and do not contribute to meeting the requirements of newborn calves for glucose. These products often do not dissociate at all or as is the case of volatile fatty acids (VFA), they dissociate very little (pK 4.8). As pK is logarithmically expressed, the drop of pH by one unit (from pH 5 to pH 4) will result in a 10 fold drop in dissociation of acetic acid, the major VFA in naturally fermented colostrum.

This means that in order to reach an optimal pH for preservation of colostrum (pH 4-4.5) more glycogenic substrate must be degraded, resulting in a low level of residual lactose. Homofermentative microbes such as *S. lactis* on the other hand, are capable of converting the lactose to lactic acid, which dissociates very readily (pK 3.1) and lowers the pH without lowering unnecessarily the lactose level in colostrum. Inoculation of colostrum with *S. lactis* should quickly lower and then maintain the pH within 4 - 4.5. In such an environment, the contamination of colostrum by acid sensitive microbes will be lowered. Acid resistant molds and yeasts, however, will flourish. Methods to control fungal contamination, leading to an increased residual level of nutrients as well as higher palatability of colostrum, appear to be needed.

Sorbic acid, widely used as mold inhibiting agent in the food industry, seems to be an ideal mold inhibitor for use in fermented colostrum. It leaves no unacceptable residues, does not impair the palatability of colostrum or interfere with the vital reactions of microbes important for lactic acid producing fermentation. In the

fermentation of colostrum, the selectivity of the inhibitory effect of sorbic acid would be of high value. Emard and Vaughn (1951) found that catalase-negative bacteria (*S. lactis, S. thermophilus, L. bulgaricus*, etc.) grew without noticeable inhibition by sorbic acid.

Increased participation of homofermentative microbes (*S. lactis*) and application of sorbic acid should lead to a higher level of residual nutrients as well as increased production of lactic acid in fermented colostrum.

Lactic acid can be converted to glucose by gluconeogenesis and as such, is available to the calf as a source of energy and to spare other glucogenic substrates, particularly amino acids.

That increased availability of glucose results in improved efficiency of protein utilization was demonstrated by Hegsted et al (1976), who infused humans with hypertonic glucose and protein hydrolysates and observed that adequate energy permitted hydrolysates to be used for protein synthesis instead of glucogenesis.

Such a possibility does not exist however, when dietary fat is used as a source of supplementary energy. When fat was increased from 10 g to 300 g of dietary dry matter, the fat deposition in the carcass of the calf increased, but retention of nitrogen was unaffected (Roy, 1973).

Fermented or chemically preserved colostrum is commonly regarded as an ideal feed for the neonatal calf. Closer scrutiny will reveal, however, that due to a relatively high protein content and low energy value, its ratio of digestible energy (DE, kJ) to digestible protein (DP, g) is far from ideal. According to Cunningham et al (1958), a DE to DP ratio of less than 104.6 kJ (25 kcal) DE/g of digestible protein is growth limiting in neonatal calves.

Lister and Lodge (1973) concluded that 129.7 - 133.9 kJ (31-32 kcal) DE/g of DP would be required for optimal utilization of milk protein. In fermented colostrum, this ratio is between 63 and 84 kJ DE/g of DP.

Increasing the energy density of calf starter by adding steamed corn (15.6 kJ/g of starter) and reducing its protein content from the existing 18% to 10%, (DE/DP ratio of 158.9) did not bring about the necessary improvement (Drevjany, unpublished data). The intake of calf starter entering the non-functional reticulo-rumen is low and so is the digestibility of its components, particularly starch. In the absence of a significant number of starch digesting rumen microbes, the utilization of starch depends mainly on the presence and activity of pancreatic and intestinal enzymes produced by the neonatal calf. A study by Radostits and Bell (1968) of the digestibility of oat flour added to skim milk powder in diets for neonatal calves showed that the apparent digestibility of the energy in oat flour was zero at 6 days of age. Since the oat meal starch was apparently transported directly into the abomasum, the study demonstrated the absence of amylolytic activity of neonatal calf during the first week of its life, as well as absence of starch digesting microbes in the lower digestive tract, known to exist in functional ruminants. When the calves reached 24 days of age, the apparent digestibility had improved to only 26.0%. These authors concluded that polysaccharides like starch, and disaccharides like maltose are not well utilized until calves are about one month of age. Gelatinization of starch (Schoch and Elder, 1955) and/or addition of amylolytic enzymes (Morrill et al, 1970) may somewhat improve the digestibility of starch by the neonatal calf. The lack of sufficient capacity and qualitative development of the reticulo-rumen in the neonatal calf makes the attempts to change the DE/DP ratio in the diet of the young calf through calf starter rather questionable.

Incorporating components, which are highly digestible by the neonatal calf, into fermented colostrum, so that the mixture will be transported directly into the abomasum, appears to offer a practical alternative to change the DE/DP ratio.

RESEARCH OBJECTIVE

The study had two aims. The first was to develop a method of preserving the nutritive value of surplus colostrum. The second was to use this colostrum effectively in the diet of neonatal calves. Specific aims were:

- to improve the uniformity, palatability and storage life of fermented colostrum, particularly during hot summer weather.
- to lower the nutrient losses during fermentation and thus increase the content of nutrients subsequently available to the neonatal calf.
- to control the contamination of fermented colostrum by acid resistant molds and yeasts and establish conditions under which such control would be maximized.
- to widen the Digestible Energy: Digestible Protein ratio in fermented colostrum so that the requirements of the neonatal calf are appropriately satisfied and calf's performance improved.

RECOGNITION TO CO-AUTHORS

The author would like to recognize the helpful advice in the area of dairy science offered by his colleague, Mr. O.R. Irvine, co-author of articles 1 and 2 (Chapters 2 and 3). His preparation of pure cultures of lactic acid producing microbes was particularly appreciated. Recognition is extended to Mr. G.S. Hooper, head of Animal Science Section, Kemptville College of Agricultural Technology, second co-author of the above articles for his valuable suggestions in the area of data evaluation and their practical utilization.

In the course of preparation and evaluation of trials presented in Chapters 4 and 5, the author benefited greatly from critical comments and numerous consultations with co-authors of articles 3 and 4, Drs. E. Donefer, Department of Animal Science, Macdonald Campus of McGill University and L. Latrille, presently of the Department of Animal Science, Laval University.

Valuable contribution in statistical evaluation of the complex laboratory experiment was made by Dr. Fanous, Department of Agricultural Chemistry and Physics, Macdonald Campus of McGill University, co-author of article 3 (Chapter 4).

CHAPTER 2

PHASE 1 - THE EFFECT OF INOCULATION OF COLOSTRUM ON ITS STORAGE CHARACTERISTICS AND ON CALF PERFORMANCE[1]

A. INTRODUCTION

Fermented bovine colostrum is an ideal feed for young calves, but some of its unattractive properties have often deterred farmers from making greater use of it. During hot summer weather, naturally fermented colostrum becomes contaminated with undesirable microorganisms (Muller and Smallcomb 1977; Rindsig et al. 1977), nutrient degradation is excessive (Carlson and Muller 1977; Otterby et al. 1977; Polzin et al. 1977, Rindsig and Bodoh 1977, Rindsig et al. 1977), storage life is shortened (Muller and Syhre 1975), and acceptability by calves is lowered (Muller et al. 1976; Rindsig and Bodoh 1977). Colostrum that ferments spontaneously provides the same opportunity to all microorganisms in that environment where the colostrum was produced, manipulated and eventually stored, to participate in the fermentation process (Pelczar et al. 1977). The type of microbes participating in the fermentation and the final products to which lactose and other fermentable compounds are converted have a substantial effect on the nutrients available to the calf (Pelczar et al. 1977) as well as upon the palatability and uniformity of the fermented product.

In an effort to regulate the fermentation process in a desired direction, one promising alternative would be massive inoculation of colostrum cultures of homofermentative microbes. Such microbes, in the environment prevailing in colostrum, are capable of converting lactose with nearly 100% efficiency to lactic acid (Collins 1977; Keen 1972; Pelczar et al. 1977). It can be assumed that such inoculation

1 The contents of this Chapter have been published:

Drevjany, L.A., Irvine, O.R. and Hooper, G.S. 1980. The feeding of fermented colostrum to neonatal calves. 1. The effect of inoculation of colostrum on its storage characteristics and on calf performance. Can. J. Anim. Sci. 60: 885-897.

will simultaneously lower the participation of heterofermentative microbes producing variable proportions of lactic acid, volatile fatty acids, ethanol, higher alcohols, ketones, gases and other substances with questionable nutritive value (Pelczar et al. 1977; Pette 1964).

Limited attempts to inoculate fresh colostrum with homofermentative microbes have not shown consistent positive effects (Muller and Syhre 1975; Otterby et al. 1977). However, Najman and Hladik (1962) reported a significant increase in average daily gain ($P < 0.05$) by calves fed with 2% fermented milk, using *S. lactis* cultured buttermilk as the inoculum, compared to 2% fresh milk.

The purpose of this study was to determine if inoculation of colostrum with a specific homofermentative culture would lead to a more uniform, palatable and nutritious product with an acceptable storage life, particularly during hot summer weather. This study consisted of a laboratory trial (trial 1) and a calf growth experiment with fermented colostrum (trial 2).

B. MATERIALS AND METHODS

(a) Trial 1

Colostrum and transitional milk (transitional milk is the term chosen to characterize that collected during the first 4 days after calving) from one cow were thoroughly mixed and equally divided into five samples so that after eight milkings, 10.8 L were accumulated per sample. One hundred and fifty mL of a "specific lactic acid producing culture" (commercial lactic acid cultures for samples A, B and C were supplied by Horan-Lalley Company, Rexdale, Ontario) were added on the first collection day to samples A, B and C, while sample D was fermented naturally and E kept at 4°C (Table 1). *

*(See page 10).

Storage temperatures during the first 4 days were adjusted to 23°C for samples A and D and 33°C for samples B and C. From 4 days up to the end of experiment at 28 days, all fermented samples (A-D) were kept at room temperature (20°C). To imitate farm conditions, each sample was thoroughly mixed twice daily. Samples were covered loosely with cheese cloth.

Titratable acidity and pH were analyzed six times per sample during the experiment. The pH values were obtained by a Fisher pH meter with glass electrode. The total titratable acid content was determined by titrating 9 mL of undiluted colostrum to a phenophthalein end point with 0.1 N NaOH. Fat, protein and lactose were analyzed at the end of the collection period (day 4), and residual lactose again at the end of the experiment (day 28). Fat was determined on a Mark II Foss Milko Tester and protein on a Foss Pro-Milk Tester, calibrated against semimicro-Kjeldahl analysis. Lactose was analyzed as reducing sugars using Association of Analytical Chemists (1970) polarimetric method 16-049.

(b) Trial 2

Thirty-two Holstein bull calves were purchased from Kemptville area farms. Within 12 h after birth, the farm was visited by a member of our technical staff who inspected the calf's general health (body temperature, signs of diarrhea) and assured early intake of

* To keep the text of the articles intact but to enable cross-referencing with Chapters 6 and 7, the numbering of the tables and figures appearing at the end of the thesis was changed. The Roman numeral refers to the phase of research, the Arabic number to the table or figure number appearing in the text.

fresh colostrum. Each calf received, intramuscularly, 1.8 cc of vitamin ADE solution (Poten ADE, Rogar STB, London, Ont.) and 1.2 cc of selenium and vitamin E solution (Dystosel, Rogar STB, London, Ont.).

Calves were fed 1.8 L of fresh colostrum twice daily for the first 4 days.

According to completely randomized design with four treatments, surplus colostrum from the first milking was randomly inoculated at the farm with 200 mL of the respective microbial culture as in treatments A, B and C in trial 1 (Table 1). Treatment D was allowed to ferment naturally and served as the control. Additional colostrum from the same source was added for the next seven milkings.

Farmers were encouraged to store the colostrum at temperatures considered desirable (20-25^{o}C for treatments A and D, 33-37^{o}C for treatments B and C).

At the end of day 4, the calves and colostrum were transported to the research facilities. The calves were housed in individual pens with wooden slatted floors in a forced-air ventilated room. Colostrum was stored in plastic containers with tight fitting lids at room temperature (22.5 $\pm$ 2.6^{o}C).

On arrival, each calf was checked for body temperature and signs of diarrhea and injected with 5 cc of penicillin. On the first day, all calves were offered 2.25 L of undiluted fermented colostrum twice a day. Starting on the second day, 2.25 L were offered once daily between 0800 and 0900 h, with simultaneous free choice offerings of water and calf starter containing 20% crude protein. Each calf was fed with colostrum produced by its dam. The consistency of feces and rectal temperature were recorded twice daily. Un-

consumed colostrum was recorded daily; starter weighbacks and body weight were recorded weekly. Calves were weaned abruptly from colostrum at 25 days of age.

At day 4 representative samples of colostrum were analyzed for total solids, protein, fat, pH and titratable acidity, using the same methods as in trial 1. Total solids in the colostrum were determined by vacuum oven drying at 100°C. Colostrum was analyzed for pH and titratable acidity at regular weekly intervals throughout the experiment.

Analyses of variance were according to methods described by Steel and Torrie (1960) for randomized design.

C. RESULTS AND DISCUSSION

(a) Total Titratable Acidity and pH

The effect of inoculation with various homofermentative lactic cultures on pH and titratable acidity development, during the storage period in trials 1 and 2, is presented in Figs. 1 and 2. The pH changes (solid line) and the variation among samples (striped area) for all treatments in the course of colostrum storage in trial 2 are compared with the curve from trial 1 (broken line) superimposed in the same figure.

Lactic acid production and lowered pH aid in extending the storage life of fermented colostrum as well as preserving nutritive value and palatability (Swanneck 1971).

Thompson and Marth (1974) demonstrated that when the pH of naturally fermented colostrum dropped

below 4.5, the reproduction of coliform bacteria, fecal *Streptococci* and gram-negative bacteria was inhibited to the point that their count started to decrease until the end of the storage period. This indicates that a sudden drop in pH below 4.5 will effectively suppress the reproduction of acid-sensitive contaminants in fermented colostrum. Acid-resistant lactic acid producing bacteria, yeasts and molds were unaffected. However, a further decrease in pH, particularly below the value of 4, may have, as noticed in trial 2, a negative effect on colostrum palatability. This is more obvious during the first 14 days of the calf's life. It appears that the optimal zone of pH within which the colostrum should be maintained is between 4 and 4.5.

In trial 1, the pH values of inoculated samples (A, B and C) had already dropped into the desired range below 4.5 at the start of the storage period (day 4) (Fig. 1). It should be noted that colostrum inoculated with *S. lactis* maintained pH within the optimal zone throughout storage and at the same time had the lowest variation in pH in the second half of the storage period. This positive effect of inoculation with *S. lactis* on uniformity of colostrum was also noticeable on visual appraisal of the sample (curd formation, separation of casein and whey fractions).

Colostrum fermented naturally, however, had a pH above 4.5 until after the 9th day, thus permitting the acid-sensitive microbes to compete for nutrients in colostrum. By day 20, the pH of this sample was below 4, which coincided with impaired palatability. In samples B and C, the pH had already dropped to 4.0 or lower by day 4 and remained there during the

entire storage period. Storage life of samples B and C was excellent, permitting an additional 3 mo. of storage. Both samples maintained their strong acidic odor and were free of surface mold contamination. However, colonies of molds were observed on the surface of samples A and D after approximately 10 days of storage. The pH curve of naturally fermented colostrum (D) was similar to those published by Otterby et al. (1977), Muller and Syhre (1975) and Rindsig et al. (1977). Variations may be partly explained by differences in initial contamination (Thompson and Marth 1974) as well as storage conditions, especially temperature (Muller and Syhre 1975).

The effect of temperature on pH changes was demonstrated by Muller and Syhre (1975). When colostrum inoculated with thermophilic *L. bulgaricus* and *S. thermophilus* was kept at 21°C, the pH curve was practically a replica of the untreated control sample, thus confirming that below 22°C, thermophilic lactobacilli do not grow well (Pette 1964). Morrill et al. (1974) cultured colostrum for 24 h with *L. acidophilus* and other lactobacilli species at 37°C and then kept it refrigerated until feeding. Optimal temperatures of 37°C should be provided for *S. thermophilus, L. bulgaricus* and *L. acidophilus* until the end of the collection period, usually the first 3 to 4 days (Pelczar et al. 1977). After that time, in agreement with common practices in the dairy industry, the product should be stored in a cool environment, thereby preventing excessive nutrient breakdown as well as impairment of palatability. Samples B and C show a relatively high pH variation in trial 2. This variation in pH is attributed to variation in temperature during the first 4 days of storage and the resultant effects on fermentation.

The titratable acidity changes are presented in Fig. 2, using the same technique as described for pH presentation. The titratable acidity values in trials 1 and 2 for treatment D and the second part of the storage period for treatment A were not consistent (Fig. 2). Variation was small in trial 2 and acidity did not exceed 1.15% (Fig. 2) in the course of 25 days of storage. Yu et al. (1976) presented similar data for naturally fermented colostrum. Conversely, Otterby et al. (1977) observed titratable acidity of over 4% for naturally fermented colostrum and 3.6% for *S. lactis*-treated product, both stored the same length of time. The positive effect of *S. lactis* inoculation on uniformity of titratable acidity values up to 15 days of storage was observed in both trials (Fig. 2). A higher build-up of titratable acidity for treatments B and C was obtained in both trials (Fig. 2).

(b) Lactose

Lactose is the only carbohydrate present in colostrum and, together with glucogenic amino acids, practically the only fermentable substrate. Due to a relative surplus of protein and insufficient energy content in bovine milk (Lister and Lodge 1973; Jacobson 1969), and colostrum in particular (Parish et al. 1950), it appears that a high residual level of lactose in sour colostrum would be of benefit to the calf. It would be of additional benefit to influence the fermentation so that the product would consist mainly of glucogenic lactic acid instead of lipogenic acetic acid, ethanol and other nutritionally inferior products. According to McCullough (1977), in homofermentation of silage, the recovery of dry matter would be 100% and energy recovery, 99.3%, whereas the dry matter recovery during heterofermentative

fermentation is 76% and energy recovery, 98.3%. Efficient conversion of lactose to glucogenic lactic acid together with a high level of residual lactose in *S. lactis*-fermented colostrum would ensure that substantial quantities of glucose would be available to meet the metabolic demands of the newborn calf. As can be calculated from Table 2, there was a 53.8-56.4% drop in lactose content during the first 4 days of fermentation for treatments C and B in trial 1, but only a 10.3-15.4% drop in treatments D and A. The residual level of lactose, after 28 days storage, was highest for treatment A (30.8%) followed by treatments B, C and D (22.8, 10.8 and 4.9, respectively). Yu et al. (1976) reported much higher residual level of lactose in naturally fermented colostrum after 35 days of storage.

(c) Protein, Fat and Total Solids

Analyses for protein and fat for trials 1 and 2 are shown in Tables 2 and 3 only after 4 days of storage. The differences between treatments were not substantial in trial 1, but in trial 2 (Table 3) the level of protein was significantly higher ($P < 0.05$) for treatments B and C, compared to A and D. Lactose level after 4 days, conversely, was significantly higher ($P < 0.05$) in treatments A and D. It is difficult to consider this pattern as a treatment effect because both the initial level of lactose and protein in transitional milk and the degree of their degradation in the first four days of fermentation are reflected in the mean value for each treatment. The same factors are involved in variations of total solids.

(d) Performance of Calves

Average daily gain and feed consumption for 53 days on test are presented in Table 4. Due to colostrum

intake refusal, no final data are available for treatments B and C.

There was a tendency toward better average daily gains for calves fed with colostrum inoculated with *S. lactis* (treatment A) up to weaning at 25 days. This trend was significant ($P<0.05$) for gains from 0 to 53 days. Muller et al. (1976), comparing naturally and *S. lactis*-fermented colostrum for newborn calves, did not find any differences in average daily gain. Daily gains in treatment D were higher than those reported by Otterby et al. (1976) (325 vs. 225 g), although the same quantity of colostrum was fed and free choice starter was available in both cases. Wide variations in performance obtained in numerous trials with fermented colostrum may be related to the quantity of colostrum fed as well as availability, quality and palatability of starter feed. Carlson and Muller (1977) observed no increase in body weight of calves up to 3 wk of age when colostrum was fed at 2.73 kg/calf/day and no dry feed was provided.

Muller et al. (1976) concluded that 1.82 kg of sour colostrum is inadequate for satisfactory growth and health when fed as the only feed source. Using Jacobson (1969) and Lodge and Lister (1973) data, it can be calculated that approximately 2.50 kg of colostrum just supplies the maintenance energy requirements for the newborn calf. Any gain must be supported by energy from starter feed.

Starter consumption up to weaning was slightly greater for treatment A than for treatment D (Table 4). To compare the readiness of calves for weaning, data on starter consumption during the weeks preceding

and following weaning are presented. Calves on treatment A consumed 19% more starter ($P<0.05$) before weaning but both A and D treatment were well above the 450 g/head/day generally considered for successful weaning. Quantity of starter consumed was approximately doubled during the week after weaning and was satisfactory for both treatments. Calves in treatment groups A and D consumed the colostrum willingly and without weighbacks. The majority of calves in groups B and C were reluctant to consume the full allotment of colostrum even at the beginning of the trial and had to be encouraged by finger feeding. No force feeding was used. Near the middle of the pre-weaning period, some calves in treatments B and C refused colostrum completely and all calves on these treatments were switched to milk replacer. The total quantity of refused colostrum by B and C treatments amounted to 58.6 and 16.2%, respectively, of that offered, which forced the premature termination of these treatments.

Low quality and palatability of colostrum in treatments B and C were reflected as well by the incidence of watery scours and increased body temperature (Table 5). Up to the premature treatment termination, feces in group B were classified as watery in 24 out of 103 calf days and 9 out of 104 calf days in treatment C. Increased rectal temperature was recorded in 48 and 17 calf days in treatments B and C, respectively. The incidence of watery scours and elevated rectal temperature were low on treatments A and D, with no appreciable differences between treatments. No respiratory disorders were identified.

In summary, it would appear that inoculation of colostrum with a *S. lactis* culture may ensure a

highly palatable and more uniform product, lower degradation of lactose and, when offered to calves, higher average daily gain and starter intake, compared to naturally fermented colostrum. No effect of inoculation with *S. lactis* on mold and yeast contamination was observed.

D. SUMMARY

Colostrum inoculated with *Streptococcus lactis* (treatment A), mixture of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* (treatment B), *Lactobacillus acidophilus* (treatment C), naturally fermented (treatment D) and kept fresh (treatment E) was used in laboratory storage trial and without treatment E in calf feeding trial. Both trials were aimed at better control of the fermentation process of colostrum and improving its acceptability and nutritive value as the main feed for calves up to weaning. The fermentation, as indicated by pH and titratable acidity changes, was most effectively controlled by an early inoculation with *S. lactis*, although it had no effect on mold and yeast contamination. The use of this culture produced better ($P<0.05$) overall daily gains (582.0 and 434.0 g for treatments A and D, respectively), higher ($P<0.05$) daily consumption of starter feed (1131 and 893 g for treatments A and D, respectively) and lowest incidence of watery diarrhea in the calf feeding trial. In the laboratory storage trial, it resulted in highest ($P<0.05$) levels of residual lactose in the fermented produce (3.41, 1.61, 1.63 and 3.15% for treatments A, B, C and D, respectively). Early development of high acidity (below pH 4) in colostrum treated by a mixed culture of *L. bulgaricus* and *S. thermophilus* or by *L. acidophilus* led to premature termination of both trial treatments due to total refusal of colostrum by calves. However, both products were free of mold and yeast contamination for the duration of 25-day storage. To assure high palatability of colostrum

and minimize acid sensitive contamination, it appears that fermentation should quickly lower and then maintain the pH within 4-4.5.

CHAPTER 3

PHASE II - THE EFFECT OF VARYING THE TIME AND SEASON OF APPLICATION OF SORBIC ACID TO FERMENTED COLOSTRUM ON CALF PERFORMANCE[1]

A. INTRODUCTION

Fermented colostrum represents a rich source of nutrients which, if properly preserved, can serve as the only source of milk protein until weaning of the dairy calf. Maintaining the quality, uniformity and palatability of colostrum often becomes difficult, particularly during hot weather. Efforts to control undesirable fermentation have included direct acidification with formic, acetic, propionic and lactic acids (Muller et al. 1976; Muller and Syhre 1975; Otterby and Dutton 1974; Polzin et al. 1975; Rindsig and Bodoh 1977; Rindsig et al. 1977), use of preservatives such as formaldehyde, sodium formate, sorbitol and gluconic acid lactone (Muller et al. 1976; Muller and Smallcomb 1977; Muller and Syhre 1975; Rindsig and Bodoh 1977; Rindsig et al. 1977; Lindahl 1974), and inoculation with lactic-acid-producing bacteria (Muller and Syhre 1975; Najman and Hladik 1969; Otterby et al. 1976; Drevjany et al. 1975). Propionic acid and formaldehyde are probably the most popular additives, but neither product controls mold and yeast growth in colostrum (Rindsig et al. 1977).

Acid-tolerant yeasts and fungi (Pelczar et al. 1977) contribute substantially to degradation of nutrients and formation of off-flavors, particularly during extended summer storage. Methods to control fungal contamination, leading to an increased residual level

1 The contents of this Chapter have been published:

Drevjany, L.A., Irvine, O.R. and Hooper, G.S. 1980. The feeding of fermented colostrum to neonatal calves. II. The effect of varying the time and season of application of sorbic acid to fermented colostrum on calf performance. Can. J. Anim. Sci. 60:899-905.

of nutrients, as well as higher palatability of colostrum, appear to be needed. An ideal fungal inhibitor should not leave unacceptable residues, impair the palatability of colostrum, or interfere with the vital reactions of microbes important for lactic fermentation.

Besides sodium benzoate and benzoic acid (Muller and Smallcomb 1977), sorbic acid seems to meet the requirements. Sorbic acid and its potassium or sodium salts are widely used as mold inhibiting agents in the food industry. According to Lück and Remmert (1974), the accepted levels vary between 1000 and 3000 ppm in most countries. In relation to fermentation of colostrum, the selectivity of its inhibitory effect is of high value. It is used in the fermentation of olives and pickles, where contaminating molds and yeasts are inhibited, but useful lactobacilli are not negatively influenced (Pfizer 1960). This claim is supported by Emard and Vaughn (1951), who found that catalase-negative bacteria (*S. lactis*, *Streptococcus thermophilus*, *Lactobacillus bulgaricus*, etc.) grew without noticeable inhibition by sorbic acid. Conversely, Hamdan et al. (1971) reported that both growth and acid production of *S. thermophilus* and *L. bulgaricus* were reduced by addition of sorbic acid at 500-1000 ppm.

The aim of these feeding trials was to measure calf response to the application of sorbic acid to fermented colostrum at different environmental temperatures and two dates of application.

B. MATERIALS AND METHODS

(a) Trial 1 - Summer

Thirty-six Holstein heifers were purchased at 4 days of age along with colostrum from their

dams. Other than the exceptions mentioned below, management, feeding, record keeping, analytical and statistical methods were as described previously (Drevjany et al. 1980).

The colostrum from the first milking of each dam was inoculated with 200 cc *S. lactis* culture (Horan-Lalley Company, Rexdale, Ont.). The transitional milk from eight milkings was added to ensure a total collection of 52 kg.

At 4 days of age, calves were transported to the research facilities and randomly allotted to two experimental treatments. For each batch of colostrum, 1000 ppm of sorbic acid (as K-sorbate) was applied to half. The rest remained untreated. Each calf obtained the colostrum from its dam. All calves were abruptly weaned after 21 days.

Representative samples (100 mL) of colostrum were taken from each container at 4 days, pooled according to treatment groups and analyzed for initial pH, titratable acidity, total solids, protein and fat. Ambient temperatures were recorded twice daily.

(b) Trial 2 - Winter

Forty-eight Holstein male calves, together with the dam's colostrum, were purchased at 4 days of age from local farms. Treatments similar to those in Trial 1 were studied.

(c) Trial 3 - Summer

Twenty-four Holstein male calves were purchased at 4 days of age with their dam's colostrum and

and allotted randomly to three treatments. Experimental procedures used were similar to those used in Trial 1.

Colostrum in group 2 was treated with 1000 ppm of sorbic acid at day 1, while in group 3 the same quantity was applied at day 4. Group 1 colostrum remained untreated.

The daily allotment of colostrum was 2L. All calves were abruptly weaned at 28 days of age.

C. RESULTS AND DISCUSSION

(a) Trials 1 and 2

Average composition of the colostrum samples is presented in Table 1. The content of total solids and fat were slightly lower than those published by Yu et al. (1976), but comparable to results reported by Rindsig and Bodoh (1977). The discrepancy can be explained by the fact that the seventh and eighth milkings were used on some farms in order to secure the required 52 kg of colostrum. The relatively low level of protein in all samples supports this assumption.

In both summer trials, the pH of composite samples dropped below the desired value of 4.5 by 4 days (Drevjany et al. 1980).

During Trial 1, the average daily gains (ADG) (Table 2) were significantly improved ($P<0.05$) by the inclusion of sorbic acid (300.6 vs. 199 g). This was despite the slightly higher initial content of total solids in the control colostrum.

ADG similar to ours were reported by Jenny et al. (1977) on lower (2.07 L) or higher (2.98 L) daily intake of colostrum. Otterby et al. (1976), feeding the same quantity of colostrum, reported ADG of 225 g. Other authors (Rindsig and Budoh 1977; Muller et al. 1976), feeding higher levels of propionic-acid - or formaldehyde-treated colostrum and free-choice starter, reported ADG from 140 to 227 g up to 3 wk of age.

During the winter period, the ADG were not significantly different between treatments (Table 2) but the growth rate of both groups compared favorably with the control group ADG in Trial 1.

The significant ($P < 0.05$) increase in ADG during the summer tests, as a result of sorbic acid application, is probably related to its mold inhibiting effect, thus lowering nutrient degradation in fermented colostrum. It was established (Drevjany, unpublished) that summer application of 1000 ppm of sorbic acid equivalent at day 4 will lower the initial mold and yeast contamination of 2×10^5 to zero by day 21, and simultaneously lower the protein degradation by 16.7% and lactose degradation by 6.8%. During winter, when the pH of fermented colostrum was higher, the same application of sorbic acid had a minimal effect on the inhibition of fungi (5×10^5 initially; 7×10^5 at 21 days).

Antimicrobial activity of sorbic acid is a function of three main factors: pH of the media, initial contamination and dose applied. Pfizer

(1960) suggested that the effective threshold of sorbic acid antimicrobial activity is a pH of approximately 6.5. Within pH 4-5, most frequently found in fermented colostrum, the portion of undissociated acid ranges between 37 and 86% (Wallhausser and Luck 1972).

Only undissociated sorbic acid exerts an inhibitory effect on microbes and, as such, is pH-dependent (FAO-WHO Report 1967). According to Wallhausser and Lück (1972), dissociated sorbic acid is about 20-40 times less effective against *Saccharomyces cerevisiae* and about 100 times less against *Aspergillus niger*, both potential contaminants of fermented colostrum. The activity against *Escherichia coli*, often the cause of scours in calves, is lowered 400 times.

The data indicate that sorbic acid applied at day 4 at 1000 ppm in the form of potassium sorbate to *S. lactis*-fermented colostrum, stored at temperatures 24.5 ± 2.5°C, will increase the ADG. The same application to colostrum stored at lower temperatures (10.5 ± 1.2°C) will have no effect. There were no refusals of colostrum in any of the trials.

The increased intake of 20% starter seems to be a contributory factor to higher ADG. It was calculated that approximately 2.5 L of fermented colostrum should supply the energy required to maintain a 45 kg calf.

Starter consumption up to 28 days was similar to (Jenny et al. 1977), or better than that reported by others (Polzin et al. 1977; Muller

et al. 1976; Otterby et al. 1976). Any gain, at this level of colostrum intake, must be supported by feeding calf starter.

Carlson and Muller (1977) did not expect gains when 2.73 kg of colostrum and no starter was fed. Feeding 2.25 L of colostrum will further increase the dependency of gain on starter consumption. Increased starter intake of 23.7% up to weaning and 22.5% during the week preceding weaning contributed not only to higher ADG, but aided in successfully weaning the calves. The starter intake during the winter period in Trial 2 was low in both groups. Higher intake of starter by calves fed colostrum, compared to whole milk, is often referred to as a compensatory effect for lower milk solids intake (Yu et al. 1976; Muller et al. 1975). However, some other experimental data show that even if the amounts of colostrum solids fed were approximately equalized to solid intake from milk replacer (Otterby et al. 1976) or from whole milk (Muller et al. 1975), the calf starter intake was still higher for groups fed colostrum. It seems that other factors, such as the ratio of digestible calories to grams of digestible protein in the liquid part of the diet, have an effect. In colostrum, such a ratio is 17-18 kcal DE/g of DCP, while for optimal utilization of milk protein, a ratio of 31-32 kcal DE/g of DCP is suggested (Lister and Lodge 1973). Lower protein degradation in sorbic-acid-treated colostrum maintains the already unfavorable ratio and may even, due to microbial degradation of energy during fermentation, narrow it further. It appears that it is the

need for additional energy, required for optimal utilization of available protein, which the calf attempts to balance by increasing its intake of starter.

No death losses occurred in either trial. The incidence of watery scours was higher in the control groups during both trials, although the average number of scour days was below 0.6 per calf, well below the values reported by Jenny et al. (1977) or Rindsig and Bodoh (1977). Incidence of increased body temperature requiring treatment was high in both groups during the summer trial 1, mainly due to respiratory problems (Table 3).

(b) Trial 3

The average content of total solids, protein and fat in colostrum, the pH, and titratable acidity are in Table 1.

Although the content of total solids was highest in the control group, a more rapid decline could be expected in this group during the storage (Carlson and Muller 1977). A relatively higher pH value at the beginning of the trial, compared to previous summer trials, is partly due to the dilution of colostrum with milk. This was necessary in order to bring the initial quantity of colostrum for each calf to 52 kg.

Palatability of colostrum was good, with no refusals throughout the trial. ADG, and starter consumption up to weaning at 28 days and during the pre-weaning week are in Table 4. When sorbic acid was applied at day 1, ADG were significantly increased ($P<0.05$) over control. Day 4

application, characterized by higher acidity and higher initial contamination of colostrum, showed a strong, but statistically nonsignificant ($P > 0.05$), tendency in the same direction. The mode of the antifungal action of sorbic acid described by Melnich et al. (1953 a,b) could help to explain why and how the dose of sorbic acid and the initial contamination affect its antimicrobial activity. The authors noted that the first step in normal metabolism of saturated fatty acids is dehydrogenation to yield the β unsaturated fatty acid. Adding the sorbic acid effectively constitutes an excess of the end-product of an essential enzymatic process, dehydrogenation. Feed-back inhibition of hydrogenase enzymes, which are basic in cell metabolism, is believed responsible for the fungistatic activity of sorbic acid (Smith and Rollin 1954; Melnich et al. 1953b).

Low concentration of sorbic acid or high contamination of molds leads to degradation of sorbic acid (Luck et al. 1972), to crotonic acid via β-oxidation (Melnich et al. 1953b), or to CO_2, H_2O and methyl ketones, when *A. niger* is involved (Kurogochi et al. 1974). For this reason, sorbic acid cannot be used to inhibit mold growth when the initial concentration of molds is already high (Lück 1973).

Starter consumption up to 28 days was similar to (Jenny et al. 1977), or better than that reported by others (Polzin et al. 1977; Muller et al. 1976; Otterby et al. 1976). The starter consumption during the week preceding weaning, although slightly lower than in the summer

period in trial 1, is, particularly for experimental treatments, close to the 500 g, considered important for successful abrupt early weaning of calves (Gorrill 1972).

Calf losses were nil for the trial. The number of calf days with watery scours was consistent with results from trials 1 and 2. Only one calf required antibiotic treatment due to elevated temperature, throughout the trial (Table 3).

The results appear to indicate that sorbic acid can be applied to *S. lactis*-fermented colostrum at any time up to 4 days of storage with similar effects on ADG.

D. SUMMARY

Three colostrum feeding trials involving 108 newborn Holstein calves were conducted in order to assess calf response to 1000 ppm of sorbic acid equivalent applied under various conditions. In the first two trials, the effect of colostrum storage temperature was tested. Calves were fed 2.25 L of colostrum inoculated with *Streptococcus lactis* once a day, together with 20% crude protein starter and water given free choice up to weaning (21 days). Potassium-sorbate-treated colostrum (1000 ppm of sorbic acid equivalent at day 4 of storage) resulted in better ($P < 0.05$) gains (300.6 vs. 199 g/day) and increased consumption of calf starter (502.9 vs. 410.6 g/day). Colostrum was stored at room temperature. Colostrum containing sorbic acid retained its palatability throughout the 21 days of the storage and was free of surface molds in contrast with the obvious mold growth on the surface of untreated

colostrum. No similar growth-promoting effect due to sorbic acid was observed during trial 2 when the ambient temperature was 10.5 ± 1.2°C and mold growth was inconsequential. As the antifungal effect of sorbic acid is a function of increasing pH and initial contamination, in trial 3 the effect of time of application on calf performance was tested. Calves were fed 2 L of colostrum cultured with *S. lactis* daily, free-choice water, and a calf starter containing 20% crude protein for 28 days. Similar growth response was obtained when potassium sorbate was applied at the beginning (day 1: high pH, low contamination) or end (day 4: low pH, higher contamination) of the collection period.

CHAPTER 4

PHASE III - LABORATORY EVALUATION OF MOLD-INHIBITING AND NUTRIENT-PRESERVING PROPERTIES OF SORBIC ACID APPLIED TO FERMENTED COLOSTRUM[1]

A. INTRODUCTION

Decreased palatability and calf refusals of fermented colostrum frequently occur after prolonged storage, particularly during hot summer weather (Muller et al. 1976). Thompson and Marth (1974), studying the microbial population in naturally fermented colostrum, demonstrated that in the initial stages of fermentation, when pH was lowered to 5 and titratable acidity reached 0.7%, the lactic-acid-producing bacterial count was matched by fecal Streptococci and surpassed by coliform bacteria. Colostrum was contaminated with yeasts and molds as well. These organisms, often referred to as heterofermentative, degrade nutrients in colostrum to a variety of products of questionable nutritive value or may result in the production of toxins (Pelczar et al. 1977). Presence of molds and yeasts in fermented colostrum should be of particular concern due to their tolerance to acidic environments. In the final stages of colostrum storage they often dominate the fermentation and cause undesirable breakdown of nutrients. Rindsig et al. (1977) reported that the mold and yeast count of naturally fermented colostrum increased from an initial value of 2.0×10^2 mL at

1 The contents of this chapter have been published:

Drevjany, L.A., Donefer, E., Latrille, L. and Fanous, M.A. 1982. The feeding of fermented colostrum to neonatal calves. III. Laboratory evaluation of mold-inhibiting and nutrient-preserving properties of sorbic acid applied to fermented colostrum. Can. J. Anim. Sci. 62: 191-205.

day 1 to 5.7×10^9 mL at day 28. Similar or even higher mold and yeasts counts were observed in colostrum treated with formaldehyde or propionic acid, documenting that direct acidification or the use of formaldehyde does not control mold and yeast contamination.

Sorbic acid, widely used as a mold and yeast inhibitor in the food industry, had a positive effect on calf's average daily gain (ADG) and preweaning consumption of calf starter, when added to fermented colostrum stored at summer ambient temperature (Drevjany et al. 1980b). In contrast to various organic acids used in preservation of colostrum, sorbic acid is applied as a neutral potassium salt and as such does not influence directly the H+ concentration in the medium. The efficacy of the mold inhibitory activity of sorbic acid depends on the dose applied, initial level of contamination and pH of the medium (Wallhauser and Lück 1972). While the efficacious dose of sorbic acid can be established rather easily, another dilemna remains. Sorbic acid can be applied early when the contamination of colostrum is low, but at that time the pH of the media is high. Alternatively, if the application of sorbic acid is delayed, the acidity of the colostrum is increased but it is also more contaminated. The purpose of this study was to investigate the effects of time and level of application of sorbic acid on nutritive value and degree of putrefaction in colostrum stored under hot (summer) or cold (winter) conditions.

B. MATERIALS AND METHODS

Colostrum, obtained from one cow during seven consecutive postpartum milkings, was instantly equally divided into 28, 2-L flasks, representing 14 treat-

ments replicated twice. Each sample was inoculated on the first day with 5 mL of a commercial lactic starter culture containing *S. lactis* and *Streptococcus cremoris* (Floran-Lalley Company, Rexdale, Ontario). Treatments 1 - 7 were stored for 28 days at ambient summer room temperature (S) with an average 0800 h temperature of 24°C and 1600 h temperature of 26.5°C. Treatments 8 - 14 were stored at simulated winter conditions (W) at a constant temperature of 10°C. Superimposed on both storage temperatures were four levels (L) of potassium sorbate (0 (control), 1000, 2000, and 3000 ppm of sorbic acid equivalent, respectively). Application of sorbic acid took place on the first day of the collection of colostrum (D1) or 3 days later, when the collection of colostrum was completed (D4). Flasks were loosely covered with paper towels and their contents were stirred twice a day.

Each sample was assessed daily for separation of whey, casein and fat, evidence of surface molds and formation of odor or off-flavor. Titratable acidity and pH of colostrum were determined in samples obtained at days 4, 7, 14, 21 and 28. Fat, protein (amido black dye binding and N x 6.38) and lactose were determined at days 4 and 28. Analytical methods were those described previously (Drevjany et al. 1980a,b). The amido black dye-binding procedure described by Weik et al. (1964) was used to assess the extent of protein degradation during fermentation. The mold and yeast counts were made at days 4, 21 and 28 using acidified (pH reduced to 3.5 with tartaric acid) potato dextrose agar and incubated at 22°C for 72 h. Colonies were counted and averaged for each treatment.

The experiment was designed as a split plot in time in a randomized complete block. The arrangement of treatments was a 2 (temperature) x 2 (day of application) x 4 (levels of application) x 2 - 5 (time of sampling of particular nutrient) factorial. Significance was tested by an F test and by Duncan's new multiple range test. For significant interactions only, the simple effects were examined (Steel and Torrie 1960). Analyses of mold and yeast counts were done on transformed data, but actual data are reported in the table of means.

C. RESULTS AND DISCUSSION

(a) pH and Titratable Acidity

The effect of storage temperature and level and day of application of sorbic acid on average pH and titratable acidity in colostrum fermented by *S. lactis* are presented in Table 1. The same table shows these values at specific sampling times.

Special attention was paid to treatment-related changes in pH, as the antimicrobial activity of sorbic acid and its salts depends on the presence of undissociated acid (Wallhausser and Luck 1972). Sorbic acid, with a dissociation constant of 1.73×10^{-5}, will remain 36 - 86% in an undissociated state within the pH range of 5 - 4. This is the pH range most frequently observed in fermented colostrum.

There were significant ($P < 0.05$) differences between the summer and simulated winter pH and titratable acidity values (Table 1). A drop in pH due to higher storage temperatures, by approximately 1 unit, was independent of superimposed treatments. Application of sorbic acid

at day 1, and to a lesser extent at day 4, resulted in substantial slowdown of the initial pH decline and in a slowdown of an increase in the titratable acidity (Table 2). A similar observation was made by Muller and Smallcomb (1977) for pH changes in colostrum treated with another pH-dependent antimicrobial agent, benzoic acid.

Summer application of sorbic acid at day 4 maintained the pH at a desirable level throughout the storage period, with minimal fluctuations, particularly when applied at higher levels (Table 2). A similar tendency was observed in titratable acidity. It can be estimated that at day 4 application (summer pH approximately 5) about 37% of sorbic acid remains in an undissociated state. This amount of undissociated sorbic acid in colostrum would be capable of effectively controlling contamination by molds and yeasts (Table 2) as well as suppressing the growth of desirable *S. lactis*, which is the main producer of lactic acid in fermented colostrum (Wallhauser and Lück 1972). Similar retardation of fermentative activity of *S. thermophilus* and *Lactobacillus bulgaricus* by sorbic acid was reported by Hamdan et al. (1971). Wallhauser and Lück (1972) observed that growth of *S. lactis* and *L. leishmanii* in a medium with pH 6, was depressed by approximately 50% when sorbic acid was applied at the 625 ppm level. Negligible growth of these organisms was observed at the 15,000 ppm level of sorbic acid. In the present work for the summer day 1 application (Table 2) in the first few days, the

environment (pH approximately 6.5) was outside the effective threshold of the antimicrobial activity of sorbic acid. However, even such a low level of undissociated sorbic acid (approximately 5.4%) was apparently capable of retarding activity of some lactic acid producers as indicated by slow buildup of titratable acidity and high initial pH (Table 2). It seems likely that the addition of acid (e.g. propionic or lactic) at day 1 in an amount that would instantly lower the pH to approximately 4.5, would provide the appropriate conditions for sorbic acid activity and thus enhance mold and yeast control in acid-treated colostrum.

Differences in the magnitude and/or direction of response in pH and titratable acidity due to the storage temperature, day of application, level of application and sampling day were the cause of significant interactions (Table 3).

During summer storage the effect of level of sorbic acid application on pH and titratable acidity was quadratic ($P < 0.05$) irrespective of the day of application. A high correlation between the dose of sorbic acid and antimicrobial activity exerted against *S. lactis* and *L. lieshmanii* was reported by Wallhauser and Lück (1972).

During simulated winter storage, application of sorbic acid at day 4 resulted in a significantly lower ($P < 0.05$) pH and a significantly higher ($P < 0.05$) titratable acidity than application at day 1, regardless of level of application (Table 1). The response in pH to increasing levels of sorbic acid was quadratic ($P < 0.05$) with day 1 application and linear ($P < 0.05$) with day 4

application. No change in pH was noted when level of sorbic acid was increased from 2000 to 3000 ppm at day 1 application (Table 2).

Summer storage of colostrum for 28 days resulted in a significant cubic decline in pH ($P < 0.05$) when sorbic acid was applied at day 1 and a quadratic decline ($P < 0.05$) when application occurred at day 4. A quadratic decline in pH of naturally fermented colostrum with an upward trend in pH at approximately 21 days of storage was reported by Wheeler et al. (1980). Decline in pH in the course of colostrum storage was reported by Carlson and Muller (1977) and Otterby et al. (1976). Changes in pH, leading to a cubic response, began with a steady decrease until about day 21, when pH began to increase, while titratable acidity increased at a slower rate (Table 2). It appears that this phenomenon was a result of increased contamination (Rindsig et al. 1977) with heterofermentative organisms, converting the diminishing fermentable nutrients from colostrum (in higher proportion) to volatile fatty acids (VFA). Also, the possibility of conversion of lactate to VFA by *S. lactis*, grown in colostrum with low level of lactose, cannot be excluded (Thomas 1979). An increase in the concentration of poorly ionized VFA (pK 4.7-4.8) is thus substituted for lactic acid (pK 3.1) and actually lowers the concentration of hydrogen ions (Pitts 1964).

(b) Lactose

Effect of sorbic acid application on the initial (day 4 of sampling) and residual (day 28 of

sampling) levels of lactose in fermented colostrum are present in Table 1 and Fig. 1. The decline in lactose content in the 28-day storage period was greater during summer than winter (Table 3).

The mid-trial (average values of days 4 and 28) content of lactose during summer storage was significantly ($P<0.05$) lower when compared to winter values. The decline in lactose content in the 28-day storage period was greater during summer than winter (Table 3). Day of application of sorbic acid did not significantly influence ($P>0.05$) the content of mid-trial lactose (Table 1). However, when sorbic acid was applied at day 4 during summer storage, the absence of an antimicrobial agent for the initial 4 days resulted in a significantly lower ($P<0.05$) initial average content of lactose in treated samples (1.52%), compared to day 1 application (2.11%). Conversely, the residual level of lactose after 28 days of storage was higher for the day 4 application when compared to the value for day 1 application (Fig. 1). The same tendency was observed in samples stored at winter temperature (Fig. 1) although residual lactose was considerably higher. It appears that the lower pH at day 4 application potentiated the antimicrobial activity of sorbic acid and thus retarded the microbial conversion of lactose to lactic acid and other fermentation products. Due to inherent losses of energy during microbial fermentation, a slowdown of fermentation should lead to energy richer colostrum for the calf. The losses of lactose between 4 and 28 days of storage over all levels of sorbic acid amounted

to 88.2% in summer and 32.5% during winter storage ($P<0.05$) (Table 1). Lower losses in summer (72.3%) were observed when sorbic acid was applied at day 4. Increased application of sorbic acid to 2000 ppm lowered the losses further to 59.6%. During winter storage the lactose losses in untreated samples amount to 60.12%, while no residual lactose was detected during summer storage.

Seidel and Shellenberger (1975) reported a 63% decrease in lactose content after storing colostrum fermented with *S. lactis* for 28 days. Only 22% reduction in lactose content was noticed by Yu et al. (1976) when naturally fermented colostrum was stored from day 8 to day 35. This relatively small decrease in lactose content, however, was accompanied by a very small increase in titratable acidity (11.2%) compared to the reported trial (146.9%). It may be assumed that minimal fermentation activity in the Yu et al. (1976) experiment led to minimal demands for fermentable substrate.

Out of the treatment combinations used, it appeared that day 4 application of 2000 ppm of sorbic acid resulted in the highest residual level of lactose after 28 days of summer storage. During winter storage the highest level of lactose was retained when sorbic acid, irrespective of the level, was applied at day 4.

(c) Protein

The degration of protein is indicated by the amount of dye-binding protein (DBP) (Table 1). A high DBP value indicates a greater amount of

undegraded protein. The extent of protein degradation during 24-day summer storage of fermented colostrum was significantly ($P < 0.05$) lowered only when sorbic acid was applied at the 2000 ppm level; However, the tendency towards lower degradation was strong with all doses of sorbic acid applied (Table 1). A similar trend toward less decrease in protein was reported by Muller and Smallcomb (1977) when colostrum was treated with sodium benzoate and benzoic acid. For winter conditions, the application of sorbic acid did not have a significant effect on protein degradation ($P > 0.05$) irrespective of the level applied. However, when sorbic acid was applied at day 4 of winter storage the mid-trial DBP level was significantly higher ($P < 0.05$) than when applied at day 1. There was no significant effect of storage temperature or day of application on DBP. Significantly higher DBP values at day 4 application than at day 1 application during winter storage, but no effect of day of application during summer storage, resulted in significant temperature by day of application interactions (Table 3). The summer storage of untreated samples resulted in increased ($P < 0.05$) protein degradation compared with winter storage. The protein degradation during summer storage amounted to 38.5% for untreated samples and approximately 25.0% for sorbic-acid-treated samples. During winter the corresponding values were 15.0% and approximately 10% (Fig. 2). The effect of length of storage on protein degradation was significant ($P < 0.05$) irrespective of the storage temperature. The decrease in protein nitrogen (N) in the course of the 24-day storage period amounted to 27.2% during summer and 10.9%

during winter (Table 1). Rindsig and Bodoh (1977) lowered the protein degradation during the 35-day summer storage period by application of 1% of propionic acid or 0.1% of formaldehyde. In propionic-acid-treated colostrum, 88.0% of the total N was in protein form compared with 86.0% for formaldehyde treatment and 79% for untreated colostrum. The same level of propionic acid failed to reduce protein degradation in another trial (Muller et al. 1976).

Total N was nearly identical in all treatments and did not differ after 24 days of storage ($P>0.05$) (Table 1). Similarly, no loss of N during storage was reported by Otterby et al. (1976, 1977) for naturally fermented colostrum or by Daniels et al. (1977) for lactic-acid-cultured or acetic-acid-treated colostrum. No change in N x 6.38 was detected from 4 to 35 days of storage for pooled, naturally fermented colostrum (Yu et al. 1976). Carlson and Muller (1977) observed a decrease in total N in colostrum over 20 days. The highest loss of N was noticed in naturally fermented, followed by formaldehyde and propionic-acid-treated colostrum. The authors explained the decrease in total N by the utilization of N by microflora during the fermentation. It could be argued that the transformation of colostral N into microbial N would not represent a net loss of N, since both forms of N remain within the fermentation broth. Furthermore, in an acidic environment which prevails throughout the storage of fermented colostrum, the minute quantities of ammonium formed during such transformation would be converted into ammonium salts of lactic and volatile fatty acids and remain relatively stable in colostrum.

(d) Fat

The day 4 summer application of sorbic acid reduced the breakdown of fat more effectively ($P<0.05$) than day 1 application (Table 1). During winter there was a similar, but nonsignificant ($P>0.05$) tendency (Fig. 3). Differences in the extent of fat breakdown between summer and winter application of sorbic acid were significant ($P<0.05$).

In the summer, the control as well as the highest level of sorbic acid (3000 ppm) had a significantly slower rate of fat decomposition ($P<0.05$) than samples with 1000 and 2000 ppm of sorbic acid. No explanation of the high residual fat content in the summer control treatment can be offered, though an analytical error seems to be a likely suspect. Explanation of the temperature by level of application interaction is meaningless if this is the case (Table 3). The average fat content during winter storage was not affected by the level of application of sorbic acid ($P>0.05$); however, there was a tendency towards reduced fat breakdown with increased level of sorbic acid (Table 1). In agreement with other (Foley and Otterby 1978), the average drop in fat percent between day 4 and 28 was nonsignificant ($P>0.05$). No explanation can be offered for the significant temperature by sampling day interactions (Table 3) as response to the treatment during higher temperature storage was opposite to what was expected (Table 1). The reports on the effect of fermentation or chemical treatment of colostrum on its content are controversial. Carlson and Muller (1977) noted less fat in naturally fermented and formaldehyde-treated colostrum than in colostrum treated with pro-

pionic acid. The opposite treatment effect was observed by Rindsig and Bodoh (1977). This seems to indicate that there are other factors involved in the fermentation process which are beyond the control of the treatment used. One of these factors may be the level of contamination of colostrum with acid-resistant molds and yeasts. Rindsig et al. (1977) reported that such contamination is not controlled by formaldehyde or propionic acid applied to the colostrum. The mold and yeast contamination may, therefore, influence, in addition to other nutrients, the content of fat of colostrum. Hydrolyses of milk fat by mold lipases was described by Pelczar et al. (1977).

(e) Mold and Yeast Count

Mold and yeast counts are presented in Table 1. When data were analyzed with the control included, the treatment effects were nonsignificant ($P > 0.05$) due to high variability and consequently high standard error. When the control data were excluded from the analysis, the day 1 application resulted in significantly lower ($P < 0.05$) average mold and yeast counts irrespective of storage temperatures. The effect of level of application was nonsignificant ($P > 0.01$). The summer application at day 1 lowered the mold count at day 4 from 10^5 (control) to 10^4 when applied at 1000 ppm, and further to 10^3 and 10^2 when applied at 2000 and 3000 ppm, respectively (Table 2). At day 21 the mold and yeast counts were zero. At day 28 there was a low, for all practical purposes, insignificant number of mold and yeasts detected (10^2). At that point the control mold count reached 10^7 (Table 2).

A similar increase in mold and yeast counts with time of storage in naturally fermented colostrum was reported by Muller and Smallcomb (1977), Palmer and Mudd (1972) and Thompson and Marth (1974). Application of propionic acid and formaldehyde (Rindsig et al. 1977) as well as sodium propionate, sodium acetate, gluconic acid lactone and sorbitol (Muller and Smallcomb 1977) did not reduce the mold and yeast contamination in colostrum. A significant reduction in yeast and mold count with the addition of sodium benzoate was reported by Muller and Smallcomb (1977). Because the antimicrobial activity of benzoic and sorbic acids depend on the presence of undissociated acid, high acidity of fermented colostrum would complement their efficacy. However, due to a higher dissociation constant (6.6×10^{-5}) only 13 - 60% of benzoic acid remains in an undissociated state within the pH range of 4 - 5 that prevails in fermented colostrum. In the case of sorbic acid the undissociated proportion would be between 37 and 86%. This may help to explain why benzoic acid (Muller and Smallcomb 1977), applied at a concentration five times higher than sorbic acid in the present trial, did not provide the same level of protection against molds and yeasts.

The day 4 summer application did not bring about a uniform drop in mold and yeast counts (i.e. only the dose of 1000 ppm was fully effective at day 21). However, at the end of the 29-day storage period, the mold and yeast counts were reduced at 10^3 or less (Table 2). Apparently the high initial contamination (10^5) lowered the overall efficacy of sorbic acid, compared

to day 1 application. However it should be noted that the total count of mold and yeast, after 28 days of storage, only amounted to approximately 0.08% of the control count (Table 2).

During simulated winter storage, application at day 1 led again to better control of yeasts and mold (Tables 1 and 2). However, counts at day 28 were considerably higher than for summer storage. With the increased level of sorbic acid application, the number of contaminating yeasts and molds decreased, irrespective of the day of application. Overall higher counts of yeasts and molds in control during winter storage, and to a lesser extent in treated samples, showed their tolerance to lower temperatures (Table 2). Compared to the control, however, the yeast and mold counts were lowered by more than 96% and 99% when treatments were applied at days 4 and 1, respectively (Table 2). As the pH of fermented colostrum during winter storage was significantly higher ($P<0.05$) compared to the summer one (Table 1), it appears that there was an interaction between environmental pH and antimicrobial activity of sorbic acid.

(f) Subjective Assessment of Stored Colostrum

Subjective assessment of colostrum for odor and separation of fractions is presented in Table 4. The odor of untreated samples increased with length of storage regardless of storage temperature. Changes had already occurred after 7 days of storage. Colonies of surface molds were first observed after 16 and 5 days of storage in summer and winter-stored

control samples, respectively. Furthermore, the number of mold colonies progressively increased throughout the storage, particularly in summer control samples. Odor of treated samples (Table 4) remained acceptable up to 28 days of storage, with summer storage generally giving a sweet odor and winter storage an acidic odor.

Separation of curd and whey was noticed at day 7 in winter control samples, with whey being sandwiched between two layers of curd. Separation also occurred in samples treated on winter day 4 (treatment 12-14); however, the curd was floating on a layer of whey. The remaining treated samples were practically unchanged during the first week of storage. At day 14, all samples stored at a summer temperature formed a uniform curd with a yellow layer floating on top. Separation of whey was complete at day 28, with the exception of treatment 7 (Table 4), which remained uniformly clotted.

All treated samples stored in the simulated winter environment showed separation of whey and curd at day 14 assessment and, at day 28, a layer of whey was sandwiched between two layers of curd. The separation of fat, curd and whey in fermented colostrum makes the thorough mixing of colostrum prior to feeding an absolute necessity.

D. SUMMARY

The effect of four levels of sorbic acid (0, 1000, 2000, 3000 ppm of sorbic acid equivalent) applied at day 1 or 4 of storage at ambient summer or simulated

winter temperatures, on nutritive value and degree of putrefaction of *Streptococcus lactis* fermented colostrum was examined. Application of sorbic acid, particularly at day 1, resulted in a slowdown of the initial pH decline and a slowdown of an increase in the titratable acidity. Day 4 application maintained the pH at a desirable level throughout the storage. The pH decreased quadratically during storage when sorbic acid was applied at day 1 and linearly when applied at day 4. The pH levels were positively related to the levels of sorbic acid applied. Storage losses of lactose were higher in summer (88.2%) than in winter (32.8%) conditions: however, day 4 summer application of 2000 ppm of sorbic acid reduced losses to approximately 60%. Protein degradation decreased when the dose of sorbic acid applied was increased. Degradation of protein was lower by 13% in summer and 5% in winter in sorbic-acid-treated samples than in control samples. No changes in total nitrogen were noticed during storage. All levels of sorbic acid applied at day 4 reduced the fat breakdown during summer storage more effectively than day 1 application. During summer storage, the day 1 application of sorbic acid led to complete elimination of molds and yeasts, while the control count grew to a level of approximately 10^6/mL. The same day of application brought about partial, but not complete, control of molds and yeasts during winter storage. Visual characteristics and odor of stored colostrum were greatly improved by the application of sorbic acid especially during summer storage.

CHAPTER 5

PHASE IV - EFFECT OF ENERGY SUPPLEMENTATION OF FERMENTED COLOSTRUM ON PROTEIN UTILIZATION[1]

A. INTRODUCTION

Whole milk is commonly regarded as the ideal food for the neonatal calf. However, Lister and Lodge (1973) and Lodge and Lister (1973) demonstrated, that cow's milk is actually deficient in digestible energy (DE) required for rapid growth. Cunningham et al. (1958) suggested that a ratio of less than 104.6 kJ (25 kcal) DE/g of digestible protein (DP) is growth limiting in neonatal calves. Lister and Lodge (1973) concluded that 129.7-133.9 kJ (31-32 kcal) DE/g of DP would be required for optimal utilization of milk protein. In whole milk the ratio is usually between 83.7 and 100.4 kJ DE per gram of DP.

In colostrum, due to its relatively high protein content, the ratio is even more unfavorable, often falling below 71.1 kJ DE/g DP. This could be further aggravated if fermentation is used as a preservation technique for surplus colostrum. During fermentation a substantial part of the glucogenic energy is converted to volatile fatty acids (VFA) and products of questionable nutritive value (alcohols), thus lowering the DE:DP ratio further. Successful

[1] The contents of this Chapter have been published:

Drevjany, L.A., Donefer, E. and Latrille, L. 1982. The feeding of fermented colostrum to neonatal calves. IV. Effect of energy supplementation of fermented colostrum on protein utilization. Can. J. Anim. Sci. 62: 439-448.

attempts have been made to reduce the energy losses by using preservatives (Rindsig and Bodoh 1977; Muller et al. 1976; Drevjany et al. 1977), direct acidification (Polzin et al. 1977; Otterby et al. 1977) and inoculation with lactic-acid-producing bacteria (Muller and Syhre 1975; Drevjany et al. 1975). However, the unfavorable DE:DP ratio was not changed substantially.

An average sample of colostrum from Holstein cows as reported by Parrish et al. (1950), contains 4.56% fat, 5.09% protein and 4.51% lactose. Using the data of McDonald et al. (1966) for gross energy and of Parrish et al. (1953) on digestibility of energy-containing compounds of colostrum, it can be estimated that the ratio of DE (kJ):DP (g) in fresh colostrum is about 74 instead of 140.6 kJ/g DP recommended for efficient protein utilization by Jacobson (1969).

This report provides information on the effect of energy addition, in the form of glucose and fat, to fermented colostrum and its effect on growth, nutrient digestibility, protein and energy utilization and calf health.

B. MATERIALS AND METHODS

Twenty-four Holstein male calves were purchased at 4 days of age along with 50 kg of colostrum per calf from local farms. Each lot of colostrum from the first milking was inoculated with 200 cc of a *Streptococcus lactis* cultured buttermilk and on day 4 colostrum was treated with 1000 ppm of sorbic acid equivalent in the form of potassium sorbate.

On arrival, each calf received intramuscularly 900,000 IU of vitamin A, 135,000 IU of vitamin D_3

and 90 IU of vitamin E (Poten ADE, Rogar STB, London, Ont.) and 3.6 mg of selenium in combination with 163 IU of vitamin E (Dystosel, Rogar STB, London, Ont.). Calves were housed in individual stalls with wooden slatted floors in a forced-air-ventilated barn. On the first day, each calf was offered 2.5 kg of electrolyte solution (IONAID, Diamond Laboratory, Calgary, Alta.). Water was available ad libitum throughout the trial. On the second day, calves were randomly allotted to four treatments (Table 1) and fed 4 kg of a mixture of 2.5 kg of fermented colostrum and 1.5 kg of hot water in two daily feedings, up to the end of the trial at 21 days. The daily allotment of colostrum was expected to provide energy at the maintenance level and a surplus of 93.2 g of DP above the maintenance requirements for a 45-kg calf. The differences in estimated levels of DP per calf per day among treatments reflect the actual crude protein content of colostrum as well as the varying contribution of protein, contained in the fat concentrate. The fat concentrate (Champlain Industries Ltd., Stanbridge Station, Que.) consisted of 58% of animal fat, 7% casein and sweet whey protein and 2% of lecithin used as an emulsifier. Prior to drying, the liquid mixture was physically homogenized. Supplementation of colostrum with a mixture of 46.4% cerelose (glucose monohydrate) and 53.6% of a fat supplement was calculated so that each calf in Treatment 4 would receive an estimated 8024.9 kJ of supplementary DE. This would change the ratio of kcal DE/g DP from 71.1 in Treatment 1 to 140.6 (Treatment 4) considered to be ideal by Jacobson (1969). The expected ratios of kJ DE/g DP were 74.1, 96.2 118.4 and 140.6 for Treatments 1, 2, 3 and 4, respectively. The supplement containing cerelose and fat was mixed with the colostrum prior to each feeding.

The consistency of feces (Larson et al. 1977) and rectal temperatures were recorded after each morning feeding. If body temperature exceeded 39.4°C the calf was injected intramuscularly with 1000 mg of chloramphenicol (AUSTICOL 500, Austin Laboratories, Guelph, Ont.). Calves that did not respond to this treatment within 24 h were treated with other anti-microbial agents. If diarrhea occurred, calves were treated, in agreement with manufacturer's instructions, with Diastat (Stevenson, Turner and Boyce, London, Ont.). Diastat contains Streptomycin, sulfathiazole and sulfamethazine as sources of anti-microbial activity and mixture of electrolytes.

Representative samples of each lot of colostrum were analyzed for pH and titratable acidity at days 1, 7, 14 and 21; pH was measured on a Fisher model pH meter with a glass electrode. Titratable acidity was found by titrating 9 mL of undiluted colostrum to phenol-phthalein end point with 0.1 N NaOH. Total solids, protein, nonprotein nitrogen (NPN), fat, lactose and ash were analyzed at days 1 and 21. Total solids and ash were analyzed according to methods in the Association of Official Agricultural Chemists (AOAC) (1970). Protein and NPN were determined on a Kjel-Foss tester using AOAC 7 B01 and B04 methods. To separate the protein for determination of NPN, 5 mL of colostrum were mixed with 2.5 mL of 15% (wt/wt) of trichloracetic acid and, after centrifugation, NPN was determined in the supernatant liquid. Lactose was analyzed as reducing sugars using AOAC volumetric method 31-051. During colostrum storage, some of the lactose is converted to lactic acid by *S. lactis* (Collins 1977), thus "lactose equivalent" was calculated by subtracting the protein, fat and ash contents from

the value of total solids. Gross energy values of the diets were calculated using the coefficients for fat, protein, lactose and cerelose given by McDonald et al. (1966). Parrish et al. (1953) data on digestibility of colostrum were used to calculate the digestible energy of the diets. A conversion coefficient of 4.184 was applied to transfer the DE values from calories to joules (J).

Blood samples were collected by jugular puncture at 4 h postfeeding on day 21 of the trial. Serum samples were analyzed for glucose (Peterson and Young 1968) and blood urea (Fawcett and Scott 1960).

Digestibilities of total solids, protein, fat, lactose equivalent and ash were determined by total fecal collection (Lodge and Lister 1973) in all calves from days 14 to 17. Analysis of fecal nutrients was conducted from frozen samples according to AOAC (1970) methods.

Data were analyzed statistically by analysis of variance. When the analysis of variance indicated significance ($P < 0.05$), the differences between individual means were tested by Duncan's new multiple range test (Steel and Torrie 1960).

C. RESULTS AND DISCUSSION

(a) Colostrum Characteristics

The basic characteristics of colostrum are presented in Table 2. There were no differences in the nutrient content, pH and titratable acidity values of fermented colostrum before supplementation.

Colostrum pH was uniform throughout the trial in contrast to results reported by Yu et al. (1976), possibly because of our inoculation with *S. lactis*. Low pH in all treatments might have contributed to feed refusal by some calves (Drevjany et al. 1975).

Titratable acidity values and their variation up to 21 days agree with data published by Muller and Syhre (1975) for colostrum inoculated with *S. lactis*. Relatively high initial acidity, compared with data published by Yu et al. (1976) and Otterby et al. (1977), reflects the high storage temperature (this trial was conducted during the summer months). Similar observations were made by Muller and Syhre (1975).

Total solids, protein and fat contents of colostrum (Table 3) are well within the ranges given in a review of Foley and Otterby (1978). Lactose levels are in agreement with those reported by Yu et al. (1976).

The breakdown of nutrients during storage amounted to 9.3% for total solids, 5.3% for protein, 2.4% for fat and 34.3% for lactose. NPN as a percent of total N amounted across the treatments to 6.6% at day 4 and 14.2% at day 21. Both values are similar to those reported by Muller et al. (1976), Polzin et al. (1977) and Rindsig (1976).

(b) Animal Performance

Although there were feed refusals, they were not related to treatment. Table 4 shows the estimated nutrient intake by calves and the ratios

of DE (kJ):DP (g) when calculated and corrected for the weighbacks. Gain increased significantly ($P<0.05$) with each increment of DE (Table 5). Based on net intake of estimated DE and using Jacobson (1969) requirements for the neonatal calf, lower gains should have been obtained in all treatments. However, when the requirements of Cunningham et al. (1958) were applied, the actual ADG (Table 5) were reasonably close to the theoretical calculations (-82.7, 58.4, 221.8 and 292.7 g for treatments 1, 2, 3 and 4, respectively).

To quantify the dependence of ADG on DE intake, a regression line was calculated (Fig. 1). The relationship between the daily intake of DE (kJ) and ADG (g) ($r=0.75$) at fixed protein intake can be described by the equation:

$$Y = 8326 + 12.26X$$

where Y = daily intake of DE (kJ) and X = ADG (g).

As the average midtrial liveweight of calves was 47.25 kg, the equation above can be transformed into a more general form:

$$Y = 176.2W + 12.26X$$

where W = liveweight in kg and X = ADG (g).

Based on the latter equation, it can be concluded that maintenance requirement of DE for a nonruminating calf fed fermented colostrum is 176 kJ/kg of liveweight and 12.3 kJ of DE required per gram of gain.

Such maintenance requirements of DE are in good agreement with the work of McGillard et al. cited

by Jacobson (1969) (171.5 kJ/kg) and Brisson et al. (1957) (187.0 kJ/kg). Substantially higher values were published by Jacobson (1969) (194.1 kJ/kg), Bryant et al. (1967) (201.7 kJ/kg), and Blaxter and Wood (1953) (219.2 kJ/kg).

Requirements of DE for production are slightly higher than data published by Brisson et al. (1957) (11.2 kJ/g of gain) and close to the value published by Blaxter and Wood (1953) (12.8 kJ/g). Bryant et al. (1967) and National Academy of Science-National Research Council (1978) published higher DE requirements per gram of gain by calves fed milk only.

The low DE requirements in this study could possibly be explained by the presence of small quantities of volatile fatty acids (VFA) in the *S. lactis* fermented colostrum (Collins 1977), for which no analyses were conducted. The VFA may have increased the total available DE slightly for calves and thus lowered the calculated DE requirements.

(c) Apparent Digestibility Study

An increase in energy supplementation resulted in a significant ($P < 0.05$) reduction in digestibility of total solids, lactose equivalent and ash (Table 6). The decrease in digestibility of fat and protein was not significant ($P > 0.05$). A reduction of protein digestibility was observed by Lodge and Lister (1973) when whole milk was supplemented with 8-10% cerelose. It seems that there is a high individual variation in a calf's ability to digest supplementary fat. While there was an average 10.25%

of fat in the fecal DM of control calves (Treatment 1), two calves in each of the supplemented treatments had over 40% fat in the fecal DM, decreasing their fat digestibility to as low as 52.4%.

The increase in fluidity of feces (Fig. 2) associated with increased energy supplementation may have been a contributing factor in lowering digestibility (Grimes and Gardner 1959).

Differences between expected and actual colostrum nutrient contents and their digestibilities altered the ratio of kJ DE:g DP in the rations offered very slightly. Actual ratios were 64.4, 82.8, 99.6 and 126.8 for Treatments 1, 2, 3 and 4, respectively (Table 4).

(d) Calf Blood Parameters

No significant differences ($P > 0.05$) among treatments for serum glucose (SG) were found (Table 7). There was a significant ($P < 0.05$) decrease in serum urea nitrogen (SUN) with increasing supplementary energy. Carlson and Muller (1977) found a positive correlation ($r=0.42$) between SUN and NPN in colostrum. Such a correlation did not exist in out trial ($r= 0.01$). It is postulated that it was the shortage of DE in Treatments 1, 2 and 3 that caused deamination of surplus DP (93.2 g in Treatment 1), resulting in formation of urea and utilization of the carbon skeletons as a source of energy. A negative correlation ($r= -0.67$) between DE intake and SUN and, a positive correlation ($r=0.94$) between surplus DP and SUN seem to support this theory.

(e) Health of Calves

With the increased supplementation of energy, the fluidity of feces was increased (Fig. 2). A similar tendency was observed by Lister and Lodge (1973). According to Blaxter and Wood (1953) a daily intake of more than 200 g of total carbohydrates per calf leads to diarrhea. This may explain the increased fluidity of feces from calves given Treatments 3 and 4, where daily net intake of lactose equivalent amounted to 201.4 and 271.5 g, respectively. No cases of Code 4 (watery) feces were recorded. The highest incidence of runny feces (Code 3) was observed during the second week.

The number of days with body temperatures above 39.4°C was similar among calves from Treatments 1, 2 and 3 (2, 3 and 2 calf days, respectively) but increased in Treatment 4 (9 calf days).

Feed refusals of diluted and/or supplemented colostrum were unrelated to treatment. Often, the feed refusal coincided with increased rectal temperature and/or higher fluidity of feces. In the course of the trial 2.56, 4.03, 7.60 and 6.13 kg of weighbacks were recorded per calf in Treatments 1, 2 3 and 4, respectively.

The supplementation of fermented colostrum with an energy source (46.4% of cerelose and 53.6% of fat supplement), resulted in an apparent improvement of colostrum protein utilization as demonstrated by better growth and decreased blood urea N. Fluidity of feces increased with increased supplementation, possibly indicating that the calves' ability to utilize carbohy-

drates and/or fat has been exceeded at the highest level of supplementation.

D. SUMMARY

Twenty-four Holstein calves were fed fermented colostrum (*Streptococcus lactis*) treated with sorbic acid, from 4 through 24 days of age to study the effects of increasing the ratio of calculated digestible energy (DE, kJ) to digestible protein (DP, g). Approximate ratios (kJ DE/g DP) studied were 64.4, 82.8, 99.6 and 126.8, which were obtained by adding increasing amounts of a mixture of cerelose (46.4%) and a fat concentrate (53.6%) to the basal colostrum diet. Recorded data included daily feed intake, body temperature, fecal characteristics, weekly body weights and analyses of colostral pH and titratable acidity. At the beginning and end of the experiment, colostrum was analyzed for protein, NPN, fat, lactose and ash. Blood serum samples taken at day 21 were analyzed for glucose and urea. A digestibility study was conducted between days 14 and 17. There was a significant ($P<0.05$) increase in gain with each increment of DE (15.9, 103.2, 174.6 and 321.4 g for DE/DP ratios of 64.4, 82.8, 99.6 and 126.9 respectively) and improvement of protein: gain (g/1000 g) ratio (9.095, 1.355, 849 and 444, respectively). Blood urea levels were being negatively correlated ($r= -0.67$) with DE intake and positively correlated ($r=0.94$) with surplus DP. There was a tendency for a reduction in digestibility of fat and protein with increasing supplementary energy. The reduction was significant ($P<0.05$) for total solids and lactose equivalent. The equation Y (DE) = 8326 + 12.3 x (ADG) was constructed to describe the relationship between the daily intake of DE (kJ) and ADG (g). The maintenance requirement of the nonruminating calf was estimated as being 176 kJ DE/kg of liveweight and 12.36 kJ of

DE required per gram of gain. Energy supplementation of fermented colostrum improved calf growth and decreased blood urea nitrogen but increased the incidence of loose feces.

CHAPTER 6

GENERAL SUMMARY

In a total of five trials, involving 164 neonatal Holstein calves and two laboratory experiments, the possibilities of improved palatability and preservation of nutrients in fermented colostrum were tested. Attempts were made to improve the protein utilization by calves by widening the digestible energy (DE) to digestible protein (DP) ratio in fermented colostrum through increased participation of homofermentative microbes, by prevention of unnecessary degradation of nutrients through yeasts and molds and eventually by direct supplementation of energy.

It was established (Chapter 2) that inoculation of colostrum with homofermentative *Streptococcus lactis* culture resulted in a more rapid drop in pH during the first four days of fermentation when compared to an uninoculated control. In addition, *S. lactis* cultured colostrum maintained the highest level of residual lactose after 28 days of storage when compared to natural fermentation or colostrum inoculated with a mixture of *Streptococcus thermophilus* and *Lactobacillus bulgaricus* or *Lactobacillus acidophilus*. Colostrum inoculated with *S. thermophilus* and *L. bulgaricus* or *L. acidophilus* was not acceptable to calves and feed refusals resulted in premature termination of the experiment for these treatment groups. There were no refusals by calves fed naturally fermented or *S. lactis* cultured colostrum. The use of *S. lactis* culture produced a better overall daily gain (582 g/day) than naturally fermented colostrum (434 g/day) and calf starter intake was significantly higher by calves fed colostrum inoculated with *S. lactis* than by those fed naturally fermented colostrum. The pH of *S. lactis* cultured colostrum was maintained

between 4.0 to 4.5 during the entire storage period. The drop in pH to 4.0 - 4.5 did not prevent acid resistant microbes, such as molds and yeasts, from further degrading the nutrients in stored colostrum.

Since acid-tolerant yeasts and fungi contribute substantially to degradation of nutrients and a formation of off-flavours, particularly during extended summer storage (Pelczar et al. 1977), control for such contamination is necessary.

Addition of sorbic acid (Chapter 3) applied at 1000 ppm during the first 4 days of summer storage resulted visually in effective control of yeast and mold contamination and improved odor. When *S. lactis* inoculated, sorbic acid-treated colostrum (1000 ppm) was fed to calves during the summer, the average daily gains and intake of calf starter were greater in comparison to naturally fermented colostrum.

Application of sorbic acid during winter storage brought only partial control of mold and yeast contamination and had no effect on calf performance. According to Wallhauser and Luck (1972), the mold inhibitory activity of sorbic acid depends on the dose applied, initial level of contamination, and the pH of the medium. In examining the effects of the above mentioned factors on efficacy of sorbic acid it was established (Table III-1) that even during winter storage, an increased dose of sorbic acid will lower the contamination of molds and yeasts. A negative effect of higher initial contamination of molds and yeasts on the efficiency of sorbic acid however, was observed in samples where application of sorbic acid was delayed to day 4 of storage (Table III-1).

Furthermore, the pH of colostrum fermented during winter was approximately 0.5 unit higher at the beginning of the

trial, and 1 unit higher at the mid-trial point, than in summer. (Table III-1). This difference in pH will substantially lower the undissociated - or effective - part of sorbic acid which is available to inhibit the molds and yeasts.

Storage temperature (Polzin et al. 1977) has an important effect on the type of microbes participating in fermentation. During winter storage, distillable acids of naturally fermented colostrum were only 0.02 mg/ml, while during summer storage were 0.19 mg/ml. Similarly the total titratable acidity, expressed as % of lactic acid was approximately 1.4% during winter and 3.06% during summer. Though the lactic acid concentration was similar irrespective of the storage temperature, distillable acidity was approximately 9.5 times greater in summer than in winter. It appears that low storage temperatures generally curtail the microbial activity in fermented colostrum. Microbes converting the fermentable substrates from colostrum in higher proportion to VFA and possibly other lipogenic substances are most affected. Controlling such microbial activity during hot summer weather is the main purpose of sorbic acid application to fermented colostrum; during winter, the same application is less justified.

A significant improvement in average daily gain (ADG) of calves fed *S. lactis* inoculated, sorbic acid-treated colostrum, stored during summer, could be explained by increased consumption of the calf starter as well as increased residual value of colostral nutrients. Small amounts of fermented colostrum were fed to calves to induce rapid initial intake of calf starter and stimulate high level of intake during the experiment.

The daily allotment of colostrum satisfied, approximately, the maintenance requirements of the new-born calf for

energy. Early intake of calf starter was required to support positive liveweight gains.

When *S. lactis* fermented and sorbic acid-treated colostrum is fed to neonatal calves, a specific factor may be involved in increased consumption of calf starter. Analyses of *S. lactis* fermented colostrum revealed that the initial level of riboflavin (0.14 mg/100 g) was increased during fermentation (0.21 mg/100 g), enough to satisfy the riboflavin requirements of the neonatal calf. No net synthesis of riboflavin was observed in naturally fermented colostrum (Drevjany et al. 1975). Riboflavin and some other vitamins of the B group are known to be synthetized by *Streptococcus lactis*, and are important co-factors in enzymatic systems regulating the energy and protein metabolism (Stryer, 1975). Sufficient amounts of vitamins of the B group may thus increase the turnover time of nutrient metabolism, and may indirectly induce higher intake of the starter feed.

The DE/DP ratio of calf starter was approximately 30% higher than the weighted average of all fermented colostrum (87.6 vs. 67.9 kJ/g). Closer examination of experiments 2 (Phase I) and 1 to 3 (Phase II) revealed that only in experiment 2 (Phase I) did higher consumption of calf starter result in an improved DE/DP ratio when compared to the control. In other trials (Phase II, trials 1 to 3), the differences in intake of starter between the control and experimental groups resulted in an improved DE/DP ratio, though the control ratios remained higher than the experimental ones. In trial 1 (Phase II) the colostral DE/DP ratio of 65.4 was increased by starter intake to 72.6 kJ/g, but remained still below the control total ration ratio of 76.0 kJ/g (Table 7-1). In the same experiment (Phase II-1) the DE/DP ratio of control

colostrum was 71.1, while in experimental colostrum, only 65.4 kJ/g. As a result, the DE/DP ratios were lower in all experimental groups in Phase II, when compared to the respective controls. The lower DE/DP ratios in experimental colostrum, were due to initial variance in nutrient composition of colostrum.

It appears that lower intake of colostral energy in all experimental treatment groups in trials 1 to 3 (Phase II), might have contributed, as pointed out by Muller et al. (1975), to the increased intake of calf starter. The ratio of DE/DP was increased by calf starter intake, but never exceeded 80 kJ/g. This ratio is approximately 40% below the optimal recommended by Lister and Lodge (1973).

Estimated intake of DE per calf per day varied from 8,077 kJ (control group in trial 3, Phase II) to 11,964 kJ (experimental group in trial 2, Phase I) (Table 7-1). Using the requirements for DE estimated in Phase IV, trial 1 (176 kJ/kg of liveweight; 12.3 kJ/g of gain), the extreme intakes of DE should have supported an average gain of 8 to 347 grams. The actual average daily gains for the same groups of calves, however, were 78 grams and 398 grams. That the daily intake of DE per calf seems to be closely correlated to the ADG is not surprising, when considering the surpluses of DP which existed in all diets. With the average mid-trial liveweight of 45.06 kg and ADG of 232.5 g, the requirements for DP would have been met by 64.2 g of DP (Jacobson, 1969). In reality, the average daily intake of DP varied between 105.1 g (control group in trial 3, Phase II) and 152.6 g (experimental group in trial 2, Phase I). Assuming that energy intake was not growth limiting, the digestible protein intake was thus sufficient to provide gains between 492 to 801 grams.

To clarify the mold inhibiting and nutrient preserving properties of sorbic acid, a laboratory experiment was conducted (Phase III). The aim of this evaluation was to observe the effect of four levels of sorbic acid applied at different times and held under two storage temperatures. During summer storage (Table III-2), the day 1 application of all levels of sorbic acid led to complete elimination of molds and yeasts, while the count in naturally fermented colostrum grew to a level of approximately 10^6 ml (Table III-2). Partial, but not complete, control of molds and yeasts was observed during winter storage at all levels of sorbic acid (Table III-2). Storage losses of lactose were higher in summer (88.2%) than in winter (32.8%) conditions. Day 4 application of 2000 or 3000 ppm of sorbic acid to colostrum stored under summer conditions, resulted in the highest residual level of lactose. Protein degradation decreased when the dose of sorbic acid applied was increased. Degradation of protein was lower by 13% in summer and 5% in winter in sorbic acid-treated samples than in control samples. No changes in total nitrogen were noticed during storage. Visual characteristics and odor of stored colostrum were greatly improved by the application of sorbic acid, especially during summer storage.

It appears that degradation of nutrients during summer storage and putrefaction of colostrum by acid-resistant molds and yeasts can be successfully lowered by application of sorbic acid during the first four days of storage. Application of 1000 ppm of sorbic acid in the form of potassium sorbate is recommended, as it controls successfully the mold and yeast contamination in fermented colostrum (Table III-2), and is well below the highest admissible level for preservation of cheese in Canada (Luck and Remmerk, 1974).

Use of homofermentative microbes leading to an increased proportion of lactic acid during fermentation, and application of sorbic acid resulting in higher residual levels of lactose in fermented colostrum, would help to widen the ratio of DE/DP to some extent, but not sufficiently to meet the requirements suggested by Lister and Lodge (1973).

Fermented colostrum was therefore supplemented with a mixture of cerelose and fat premix (with ratio of glucogenic and lipogenic energy similar to whole milk) so that its ratio of 64.4 kJ DE/g of DP (treatment 1) would be increased to 82.8, 99.6 and 126.8 for treatments 2, 3 and 4 respectively (Phase IV). Two and a half kilograms of *S. lactis* inoculated, sorbic acid-treated colostrum with (experimental groups) or without (control group) the supplement were offered per calf per day. The gains were significantly ($P<0.05$) increased with each increment of DE (15.9, 103.2, 174.6 and 321.4 g for treatments 1, 2, 3 and 4 respectively). Based on intake of DE and using Jacobson (1969) requirements for neonatal calf, lower gains should have been obtained in all treatments. However, when requirements of Cunningham et al. (1958) were applied, the actual ADG were reasonably close to the theoretical calculations (82.7, 58.4, 221.8 and 292.7 g for treatments 1, 2, 3 and 4 respectively). As a measure of efficiency of protein utilization, the protein/gain ratios were calculated. They followed a similar trend in response to increase in DE as was noted for ADG (9,095, 1,355, 849 and 444 g for treatments 1, 2, 3 and 4 respectively).

Improved protein utilization due to increased intake of DE was accompanied by significant drop ($P<0.05$) in blood urea levels (22.95, 14.57, 10.57 and 7.23 mg % for treatments 1, 2, 3 and 4 respectively). Blood urea levels

were negatively correlated (r = .94) with surplus DP. It appears that the shortage of DE in treatments 1, 2 and 3 caused deamination of surplus DP, resulting in formation of urea and utilization of the carbon skeletons as a source of energy.

There was a tendency for a reduction in digestibility of fat and protein with increasing supplementary energy. The digestibility of total solids and lactose equivalent were significantly lower as the DE/DP ratio increased. The DE for maintenance of the nonruminating calf was estimated as being 176 kJ/kg of liveweight and 12.4 kJ of DE/g of gain.

CHAPTER 7

CONCLUSIONS

Phase I

During the first phase of research, colostrum inoculated with *Streptococcus lactis*, a mixture of *S. thermophilus* and *Lactobacillus bulgaricus*, and *L. acidophilus*, was compared with naturally fermented or fresh colostrum in a laboratory storage trial and calf feeding trial.

The fermentation, as indicated by pH and titratable acidity changes and residual nutrient levels was most effectively controlled by an inoculation with *S. lactis*. The quick drop and then maintenance of pH within 4-4.5 assured high palatability and uniformity of colostrum and minimized acid-sensitive contamination. Inoculation with *S. lactis* had no effect on mold and yeast contamination.

Calves fed inoculated colostrum, exhibited higher average daily gain, higher calf starter intake and lower incidence of watery diarrhea than calves fed naturally fermented colostrum.

Phase II

Second phase of experimentation involved three colostrum feeding trials, designed to assess calf response to 1000 ppm of sorbic acid equivalent applied under various conditions.

Colostrum containing sorbic acid retained its palatability throughout the 21 days of the summer storage period, and was free of surface molds in contrast with the obvious

mold growth on the surface of untreated colostrum. Calves fed sorbic acid treated colostrum stored during summer had higher ($P<0.05$) daily gain, and increased consumption of calf starter than calves fed untreated colostrum.

No similar effects due to sorbic acid were observed during winter trial when the ambient temperature was $10.5 \pm 1.2^{o}C$, and mold growth was inconsequential. Time of application of sorbic acid did not affect the growth rate of calves, or intake of calf starter. Sorbic acid can be applied at any time up to 4 days of storage to *S. lactis* fermented colostrum.

Phase III

Laboratory experiment was conducted to clarify the mold inhibiting and nutrient preserving properties of sorbic acid. Sorbic acid was applied at day 1 or 4 of storage at four levels (0, 1000, 2000, or 3000 ppm sorbic acid equivalent), to *S. lactis* inoculated colostrum stored at ambient summer or simulated winter conditions.

Day 4 application maintained the pH at a desirable level throughout the storage. Nutrient degradation was greater in summer than in winter storage conditions. Application of 1000 ppm of sorbic acid at day 4 resulted in increased residual levels of lactose and protein and elimination of mold and yeast contamination, in comparison to untreated *S. lactis* fermented colostrum. Visual characteristics and odor of stored colostrum were greatly improved by the application of sorbic acid, especially during summer storage.

Phase IV

Final adjustment of an unfavourable digestible energy to digestible protein ratio in fermented colostrum, was accomplished by direct supplementation at the time of feeding with a mixture of emulsified fat and cerelose.

DE/DP ratios (kJ DE/g DP) were adjusted to 64.4, 82.8, 99.6 and 126.8. With each incremental increase in DE/DP ratio, increased gains and protein/gain ratios were observed in calves fed the supplemented *S. lactis*-sorbic acid treated colostrum. Blood urea levels were negatively correlated with surplus DP. A significant reduction in the digestibility of total solids and lactose equivalent and a tendency to reduction in the digestibility of fat and protein was observed in response to increased DE/DP ratios. The equation Y (DE) = 8,326 + 12.3 x (ADG) was constructed to describe the relationship between the daily intake of DE (kJ) and ADG.

Inoculation of colostrum with *S. lactis* culture, inhibition of molds with sorbic acid and supplementation with energy result in a more palatable, uniform and nutritious feed for neonatal calves.

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Table I - 1. Planned storage temperatures
(Trials 1 and 2)

Sample	Inoculant	Planned storage temperature (°C)	
		First 4 days	Day 5 - 28
A	S. lactis	23	20
B	L. bulgaricus and S. thermophilus	33*	20
C	L. acidophilus	33*	20
D	None (natural fermentation)	23	20
E	None	4	4

*For technical reasons, 33°C was used instead of recommended 37°C (Pelczar et al. 1977).

Table I - 2. Gross chemical composition of stored colostrum (Trial 1)

Item	Treatment and inoculant								
	A		B		C		D		E
	S. lactis		S. thermophilus L. bulgaricus		L. acidophilus		Naturally fermented		None
	4 days	28 days	4 days	28 days	4 days	28 days	4 days	28 days	4 days
Lactose (%)	3.30	1.20	1.70	0.87	1.80	0.17	3.50	0.42	3.90
Protein (%)	5.12	-	5.20	-	5.16	-	5.10	-	5.00
Fat (%)	4.36	-	4.36	-	4.36	-	4.36	-	4.36

Table I – 3. Gross chemical composition of fermented colostrum on day 4 of storage period (Trial 2)

Item	Treatment and inoculant			
	A	B	C	D
	S. lactis	S. thermophilus L. bulgaricus	L. acidophilus	Naturally fermented
Total solids (%)	14.35a ± 1.05	13.00a,b ± 1.01	12.50 b ± 2.06	14.01a,b ± 1.18
Protein (%)	5.15b ± .91	6.72a ± 1.10	6.19a ± .84	5.18b ± 1.09
Fat (%)	4.06 ± .42	4.14 ± .52	3.82 ± .40	3.86 ± .58
Lactose (%)	3.41a ± .37	1.61b ± .34	1.63b ± .40	3.15a ± .31

a,b Figures in rows having a different letter differ significantly ($P < 0.05$)

Table I - 4. Performance of Holstein calves as affected by microbial treatment of colostrum

Item	Treatment and inoculant A S. lactis	B S. thermophilus & L. bulgaricus	C L. acidophilus	D Naturally fermented	± SE
No. of calves	8	8	8	8	
Daily gains (g):					
to weaning (25 days)	398.0	*	*	325.0	45
to end of experiment (53 days)	582.0a	*	*	434.0b	41
Feed consumed (g):					
a) starter/calf/day up to 25 days	439.0	*	*	290.0	25
up to 53 days	1131.0a	*	*	893.0b	62
during week prior to weaning	571.0	*	*	478.0	44
during week after weaning	1063.0	*	*	1107.0	111
b) intake of colostrum up to weaning (litres)	49.5	12*	24.5*	49.5	
% refused	Ø	58.6	16.2	Ø	

*Experiment terminated prematurely.

a,b Figures in rows having a different letter differ significantly ($P < 0.05$)

Table I - 5. Incidence of watery scours and increased rectal temperature as affected by microbial treatment of colostrum

	Treatment							
	A		B		C		D	
	Days	%	Days	%	Days	%	Days	%
Incidence of watery scours (code 4) up to weaning*	5	2.98	24	23.3	9	8.65	8	4.76
Rectal temperature above 39.4°C**	8	4.76	48	46.6	17	16.35	8	4.76

*Physical appearance and consistency of feces characterized as watery scours agreed with the one described by Larson et al. (1977).

**Elevated rectal temperature above 39.4°C is considered cause for antibiotic treatment.

All figures appearing in the table refer to calf-days in absolute terms and as a percent of total. The total period from birth to weaning represents 168, 103, 104, and 168 calf-days for Groups A, B, C and D, respectively.

Table II - 1. Average composition of fermented colostrum in trials 1, 2 and 3

Item	Trial 1 (summer)		Trial 2 (winter)		Trial 3 (summer) (time of application of sorbate)		
	Control	Sorbate	Control	Sorbate	Control	Day 1	Day 4
Total solids (%)	14.35	14.01	13.86	14.12	15.16	14.82	14.46
Total (N x 6.38) protein	5.18	5.15	5.32	5.58	5.72	5.66	5.56
Fat (%)	4.06	3.86	3.86	4.02	4.52	4.60	4.38
pH	4.30	4.54	4.98	5.02	4.85	4.86	4.56
Titratable acidity (1%)	.95	.94	.76	.66	.67	.56	.72

Table II - 2. Effect of sorbic acid on average daily gain and starter intake of calves in trials 1 and 2

	Number of animals	Starting weight (kg)	ADG up to weaning (g)	Average daily 20% starter consumption up to weaning (g)	Average daily 20% starter consumption during 3rd week (g)
Trial 1 - summer:					
Control	18	41.04	199.0b	239.8	410.6
Sorbate	18	37.44	300.6a	296.7	502.9
SE			20.8	20.4	38.2
Trial 2 - winter:					
Control	24	44.92	190.1	186.6	266.2
Sorbate	24	44.47	207.4	199.0	281.9
SE			14.4	9.9	17.0

a, b Means in columns with different letters are significantly different ($P<0.05$).

Table II - 3. Incidence of watery scours and increased rectal temperature as affected by presence and time of application of sorbic acid in trials 1, 2 and 3

Item	Trial 1 (summer)				Trial 2 (winter)				Trial 3 (summer) Time of application of sorbate					
	Control		Sorbate		Control		Sorbate		Control		Day 1		Day 2	
	Days	%	Days	%	Days	%	Days	%	Days	%	Days	%	Days	%
Incidence of watery scours (code 4) up to weaning*	11	2.91	2	.53	14	2.78	6	1.19	4	1.79	3	1.34	3	1.34
Rectal temperature above 39.4°C**	23	6.5	32	8.5	14	2.78	4	.79	-	-	3	1.34	-	-

* Physical appearance and consistency of feces characterized as watery scours were as described by Larson et al. (1977).

** Elevated rectal temperature above 39.4°C was considered cause for antibiotic treatment.

All figures appearing in the table refer to calf-days in absolute terms and as a percent of total. The total period from birth to weaning represents 378, 504 and 224 calf-days per treatment in experiments 1, 2 and 3 respectively.

Table II - 4. Effect of day 1 vs. day 4 application of sorbic acid on ADG and starter consumption in trial 3

	Number of animals	Starting weight (kg)	ADG up to weaning (28 days) (g)	Average daily 20% starter consumption up to weaning (g)	Average daily 20% starter consumption in last week prior to weaning (g)
Control	8	44.1	78a	193	353.6
Day 1 application	8	40.9	248b	260	442.7
Day 4 application	8	40.6	239a,b	238	444.5
SE			56	26	49.3

a, b Values with different letters in the same column are significantly different ($P < 0.05$).

Table III – 1. Mean effects of storage temperature, day of application, level of application and sampling time, on nutritive value of sorbic acid treated colostrum.

		pH		Titratable acidity		Lactose		Dye binding protein		N x 6.38		Fat		Mold and yeast count		Mold and yeast count (control excluded)	
		Summer	Winter	Summer %	Winter	Summer %	Winter	Summer %	Winter	Summer	Winter	Summer %	Winter	Summer	Winter	Summer	Winter
Day of Application	1	A,a 4.56	B,a 5.73	A 1.42	A,b .39	A 1.08	B 1.75	a 3.53	a 3.54	5.17	5.12	A,a 4.65	B 4.82	$3.79x10^3$	$1.33x10^4$		
	4	A,b 4.37	B,b 4.97	A 1.41	B,b .71	A .97	B 1.67	b 3.40	b 3.64	5.18	5.09	A,b 4.74	B 4.92	$1.16x10^5$	$6.27x10^5$		
	SE	.030	.032	.045	.012	.056	.094	.077	.048	.047	.076	.034	.076	$6.36x10^5$	$2.49x10^5$		
Level of application	0	A,a 3.98	B,a 4.84	A,a 2.16	B,a .75	A,a .79	B,a 1.17	A,a 3.21	B 3.50	5.08	5.06	A,a 4.80	B 4.48	$5.24x10^6$	$1.26x10^7$		
	1,000 ppm	A,b 4.14	B,b 5.19	A,b 1.88	B,b .59	A,a,b .91	B,b 1.58	a,b 3.39	3.57	5.17	5.08	b 4.64	4.79	$4.93x10^4$	$5.47x10^5$		
	2,000 ppm	A,c 4.47	B,c 5.41	A,c 1.37	B,c .53	A,b,c 1.04	B,b 1.79	b 3.59	3.58	5.16	5.15	b 4.66	4.84	$6.76x10^4$	$2.46x10^5$		
	3,000 ppm	A,d 4.77	B,c 5.45	A,d .99	B,c .53	A,c 1.13	B,b 1.75	B,a 3.44	a 3.62	5.19	5.09	a 4.79	4.98	$6.29x10^4$	$1.67x10^5$		
	SE	.015	.016	.023	.006	.028	.047	.039	.024	.024	.038	.017	.038	$3.18x10^5$	$1.24x10^5$		
Time of sampling day	4	A,a 5.40	B,a 5.91	A,a .61	B,a .45	A,b 1.78	B,A 1.94	a 3.97	a 3.78	5.12	5.12	4.66	4.88	$1.68x10^5$	$4.91x10^5$	$1.53x10^5$	$2.40x10^5$
	7	A,b 4.75	B,b 5.41	A,b .76	B,b .49												
	14	A,c 4.04	B,c 5.05	A,c 1.64	B,c .59												
	21	A,d 3.94	B,c 5.02	A,d 2.25	B,d .66									$7.12x10^5$	$2.59x10^6$	$2.69x10^4$	$2.67x10^5$
	28	A,e 3.85	B,c 5.01	A,e 2.35	B,e .71	A,b .21	B,b 1.31	A,b 2.89	B,b 3.37	5.21	5.08	4.76	4.74	$1.52x10^6$	$3.12x10^6$	$2.05x10^2$	$4.53x10^5$
	SE	.007	.006	.009	.003	.004	.106	.066	.049	.042	.080	.025	.080	$6.61x10^5$	$9.19x10^5$		

a,b,c,d,e, values within season with differing postscripts were significantly different at $P<0.05$.
A,B, values in rows for colostrum components were significantly different at $P<0.05$.

Table III - 2. Simple effects of storage temperature, day of application, level of application and sampling time on pH, Titratable Acidity and Mold Count of sorbic acid treated colostrum.

	TREATMENT		WINTER					WINTER				
	Day of applic.	Level Applied	Sampling Day 4	7	14	21	28	Sampling Day 4	7	14	21	28
pH	-	1	5.06	4.27	3.38	3.48	3.70	5.43	4.97	4.51	4.58	4.63
	1	2	5.56	4.91	3.82	3.45	3.62	6.08	5.60	5 29	5.27	5.32
	1	3	5.87	5.23	3.96	3.60	3,73	6.38	5.91	5.65	5.68	5.61
	1	4	5.99	5.31	4.86	4.31	4.25	6.36	5.93	5.59	5.64	5.72
	4	2	5.04	4.43	3.54	3.50	3.70	5.58	5.00	4.57	4.56	4.65
	4	3	5.09	4.49	4.30	4.24	4.23	5.70	5.19	4.69	4.67	4.68
	4	4	5.20	4.61	4.41	4.36	4.37	5.82	5.26	4.78	4.71	4.74
Titratable Acidity (%)	-	1	0.74	0.95	2.97	3.13	3.03	0.57	0.62	0.72	0.89	0.97
	1	2	0.56	0.65	1.64	2.97	3.09	0.36	0.39	0.47	0.53	0.60
	1	3	0.42	0.52	1.50	2.75	2.73	0.29	0.32	0.35	0.36	0.46
	1	4	0.39	0.52	0.78	1.27	1.49	0.28	0.30	0.37	0.38	0.46
	4	2	0.73	0.93	2.43	2.90	2.89	0.57	0.63	0.75	0.81	0.81
	4	3	0.75	0.88	1.14	1.43	1.65	0.54	0.61	0.76	0.82	0.84
	4	4	0.73	0.88	1.03	1.29	1.56	0.54	0.59	0.74	0.83	0.83
Mold Count (10^3)	-	1	265.2	-	-	4,825.0	10,625.0	1,995.0	-	-	16,550.0	19,150.0
	1	2	25.0	-	-	0.0	0.0	1.5	-	-	10.0	73.1
	1	3	8.1	-	-	0.0	0.0	1.0	-	-	3.8	26.3
	1	4	0.9	-	-	0.0	0.0	0.5	-	-	0.9	2.3
	4	2	270.6	-	-	0.0	0.4	543.0	-	-	669.0	1,983.0
	4	3	280.0	-	-	117.5	0.0	517.5	-	-	531.0	399.0
	4	4	331.6	-	-	44.0	0.8	379.0	-	-	387.0	235.0

Table III - 3. Significance of Treatment Interactions

Interactions*	pH		Titratable Acidity		Lactose		Dye Binding Protein		N x 6.38		Fat		Mold Count (10^6)	
	Signi-ficance	SE	Sig.	SE	Sig.	SE	Sig.	SE	Sig.	SE	Sig.	SE	Sig.	SE
2 way:														
TxD	*	.282	*	.160	NS	.013	*	.112	NS	.022	NS	.004	NS	7.925
TxL	*	.104	*	.238	NS	.039	NS	.042	NS	.027	*	.076	NS	1.703
TxS	*	.190	*	.861	*	.462	*	.281	NS	.052	*	.073	NS	5.370
DxL	*	.055	*	.113	NS	.035	NS	.072	NS	.033	NS	.059	NS	3.175
DxS	*	.013	*	.247	*	.422	NS	.046	NS	.000	NS	.046	NS	1.965
LxS	*	.062	*	.190	NS	.056	NS	.013	NS	.011	NS	.045	NS	6.972
3way:														
T*D*L	*	.123	*	.197	NS	.129	NS	.022	NS	.067	NS	.094	NS	8.911
T*D*S	*	.294	*	.432	NS	.088	*	.193	NS	.051	*	.111	NS	7.203
T*L*S	*	.094	*	.615	NS	.049	NS	.035	NS	.039	NS	.068	NS	9.328
D*L*S	*	.077	*	.384	NS	.031	NS	.056	NS	.023	NS	.051	NS	11.423
4 way:														
T*D*L*S	*	.140	*	.587	NS	.200	*	.075	NS	.081	NS	.080	NS	14.747

P<0.05

*T=Storage Temperature; D=Day of application; L=Level applied; S=Sampling time.

Table III – 4. Subjective assessment of the stored colostrum

Treatment				Observed on storage day											
				7				14				28			
					Appearance				Appearance				Appearance		
No.	Season	Day of appli.	Level of appli. (ppm)	Odour	T	M	B	Odour	T	M	B	Odour	T	M	B
1	S	–	0	moldy, yeasty	1/6 F*	–	5/6 C	moldy, yeasty	1/6 F	–	5/6 C	moldy, yeasty. off	1/3 C	1/3 W	1/3 C
2	S	1	1,000	pleasant	1/6 F	–	5/6 U	slightly acidic	1/6 F	–	5/6 C	pleasant, sweet	1/6 F	2/6 W	3/6 C
3	S	1	2,000	pleasant	1/6 F	–	5/6 U	slightly acidic	1/6 F	–	5/6 C	distinct, cheese	1/6 F	1/6 W	4/6 C
4	S	1	3,000	pleasant	1/6 F	–	5/6 U	slightly acidic	1/6 F	–	5/6 C	distinct, cheese	1/6 F	1/6 W	4/6 C
5	S	4	1,000	sweet, distinct	1/6 F	–	5/6 U	sweet, pleasant	1/6 F	–	5/6 C	sweet	1/6 F	2/6 W	3/6 C
6	S	4	2,000	sweet, distinct	1/6 F	–	5/6 U	sweet, pleasant	1/6 F	–	5/6 C	sweet	1/6 F	2/6 W	3/6 C
7	S	4	3,000	sweet, distinct	1/6 F	–	5/6 U	sweet, pleasant	1/6 F	–	5/6 C	sweet	1/6 F	–	5/6 C
8	W	–	0	yeasty, moldy	1/6 C	2/6 W	3/6 C	yeasty, moldy	1/6 C	3/6 W	2/6 C	yeasty, moldy, off	2/6 C	3/6 W	1/6 C
9	W	1	1,000	pleasant	1/6 F	–	5/6 U	slightly acidic	1/6 C	3/6 W	2/6 C	slightly acidic	1/6 C	3/6 W	2/6 C
10	W	1	2,000	pleasant	1/6 F	–	5/6 U	slightly acidic	3/6 C	–	3/6 W	slightly acidic	1/6 C	2/6 W	3/6 C
11	W	1	3,000	pleasant	1/6 F	–	5/6 U	slightly acidic	3/6 C	–	3/6 W	musty	1/6 C	2/6 W	3/6 C
12	W	4	1,000	acidic	3/6 C	–	3/6 W	slightly acidic, off	1/6 C	3/6 W	2/6 C	slightly acidic	1/6 C	2/6 W	3/6 C
13	W	4	2,000	slightly acidic	3/6 C	–	3/6 W	slightly acidic	3/6 C	–	3/6 W	slightly acidic	1/6 C	2/6 W	3/6 C
14	W	4	3,000	slightly acidic	3/6 C	–	3/6 W	slightly acidic	2/6 C	2/6 W	2/6 C	slightly acidic	1/6 C	2/6 W	3/6 C

Legend:

F – yellow (fat) layer

C – curd

W – whey

U – unchanged

T – top

M – middle

B – bottom

* fraction before the letter pertains to the proportion of the storage container.

Table IV - 1. Level of energy supplementation of fermented colostrum diets

Treatments	Colostrum/ calf/day	Supplement		Total estimated DE intake/ calf/day	Total estimated DP intake/ calf/day	Estimated DE/DP ratio:
		Cerelose/ calf/day	Fat concentrate*/ calf/day			
1.	(kg)	(g)	(g)	(kJ)	(g)	(kJ/g)
Colostrum	2.5	-	-	8,635.8	134.3	64.3
2. Colostrum and 1/3 supplement	2.5	58	67	9,786.8	118.1	82.9
3. Colostrum and 2/3 supplement	2.5	116	134	12,426.5	127.5	97.5
4. Colostrum fully supplemented	2.5	174	201	13,610.6	107.2	127.0

* Contains 60% fat (of which 2% is lecithin) and 7% milk protein.

Table IV - 2. Colostrum fermentation characteristics

Item	Day	Treatments 1.	2.	3.	4.	SE of means
pH	1	4.36	4.47	4.55	4.55	.08
	7	4.22	4.30	4.14	4.28	.08
	14	4.06	3.91	3.84	3.96	.02
	21	3.78	3.73	3.67	3.85	.10
Titratable acidity (%)	1	1.09	.96	1.01	.98	.14
	7	1.35	1.08	1.52	1.14	.16
	14	1.60	1.81	2.25	1.76	.27
	21	2.13	2.29	2.70	2.03	.23

Table IV - 3. Chemical composition of fermented colostrum prior to supplementation

Analyses	Treatments								SE of means	
	1.		2.		3.		4.			
	Day 1	Day 21	Day 1	Day 21	Day 1	Day 21	Day 1	Day 21	Day 1	Day 21
Total solids (%)	15.7	14.4	14.8	13.5	15.6	14.0	15.1	13.7	.44	.53
Protein(Nx6.38)(%)	6.8	6.1	5.8	5.7	6.5	6.1	5.7	5.5	.38	.41
Fat (%)	4.7	4.6	4.3	4.3	4.6	4.5	4.4	4.8	.24	.23
Lactose (%)	3.2	2.5	3.3	2.1	3.1	1.8	3.3	2.1	.14	.30
Ash (%)	.90	.92	.90	.88	.91	.93	.93	.93	.02	.02
NPN (%)	.37	.82	.42	.78	.48	1.02	.36	.72	.02	.01

Table IV - 4. Average daily intake of digestible energy and digestible protein by calves

Treatments	Average daily nutrient intake per calf up to weaning				DE/DP	
	Estimated DE (kJ)		Estimated DP (g)			
	offered	consumed	offered	consumed	offered	consumed
					(kJ/g)	(kJ/g)
1	8,636	8,021	134	123	64.4	65.2
2	9,786	9,276	118	111	82.9	83.6
3	12,426	11,155	128	111	97.1	100.5
4	13,606	12,389	107	97	127.2	127.7
SE	515	707	9.5	10.3		

Table IV - 5. Rate and efficiency of gains by calves

Treatment	Mid-trial ave. live-weight of calves	Maintenance* Requirements		Potential ADC** based on available		Actual ADG	CP/gain
		DE	DP	DE	DP		
	(kg)	(kJ)	(g)	(g)	(g)	(g)	(g/1000 g)
1	47.9	9,410	28.7	-100.5	592.2	15.9[a]	9,095[a]
2	46.1	9,067	27.7	15.3	520.4	103.2[b]	1,355[b]
3	46.4	9,117	27.8	147.8	522.8	174.6[c]	849[c]
4	48.7	9,577	29.2	203.8	424.2	321.4[d]	444[d]
SE	2.5					15.7	144

*Maintenance requirements used were 196.6kJ (47kcal) of DE and .6 g of DP per kg of liveweight.

** Production requirements 13.8kJ (3.3 kcal) of DE and .16 g of DP per gram of gain (Jacobson 1969).

a,b,c,d Means within columns with different letters were significantly ($P<0.05$) different.

Table IV - 6. Apparent digestibilities (%) of dry matter, protein, fat, lactose equivalent* and ash

Digestibility coefficients	Treatments				SE of means
	Colostrum only	Colostrum and 1/3 supplement	Colostrum and 2/3 supplement	Colostrum fully supplemented	
Dry matter	89.9[a]	85.1[a,b]	83.5[a,b]	77.7[b]	2.86
Protein	85.9	78.5	74.8	67.6	4.59
Fat	96.7	84.4	85.3	79.2	5.05
Lactose equivalent	90.8[a]	94.4[a]	91.5[a]	83.4[b]	2.07
Ash	82.1[a]	75.0[a,b]	68.1[b]	67.5[b]	3.72

a,b Means within rows with different letters were significantly ($P<0.05$) different.

* Lactose equivalent represents the difference between total solids and the sum of protein, fat and ash in colostrum. It is assumed that it includes lactose and lactic acid.

Table IV – 7. Calf blood parameters at day 21

Item	Treatments				
	1 Colostrum only	2 Colostrum and 1/3 supplement	3 Colostrum and 2/3 supplement	4 Colostrum fully supplemented	SE of means
	(mg/100 ml)	(mg/100 ml)	(mg/100 ml)	(mg/100 ml)	
Serum glucose	70.8	83.7	88.0	81.7	4.42
Serum urea	22.95[a]	14.57[b]	10.57[c]	7.23[c]	1.34

[a,b,c] Means within rows with different letters were significantly ($P<0.05$) different.

TABLE 7-1

SUMMARY OF THE MAIN PERFORMANCE PARAMETERS IN TRIALS I-2, II-1, II-2 AND II-3[1]

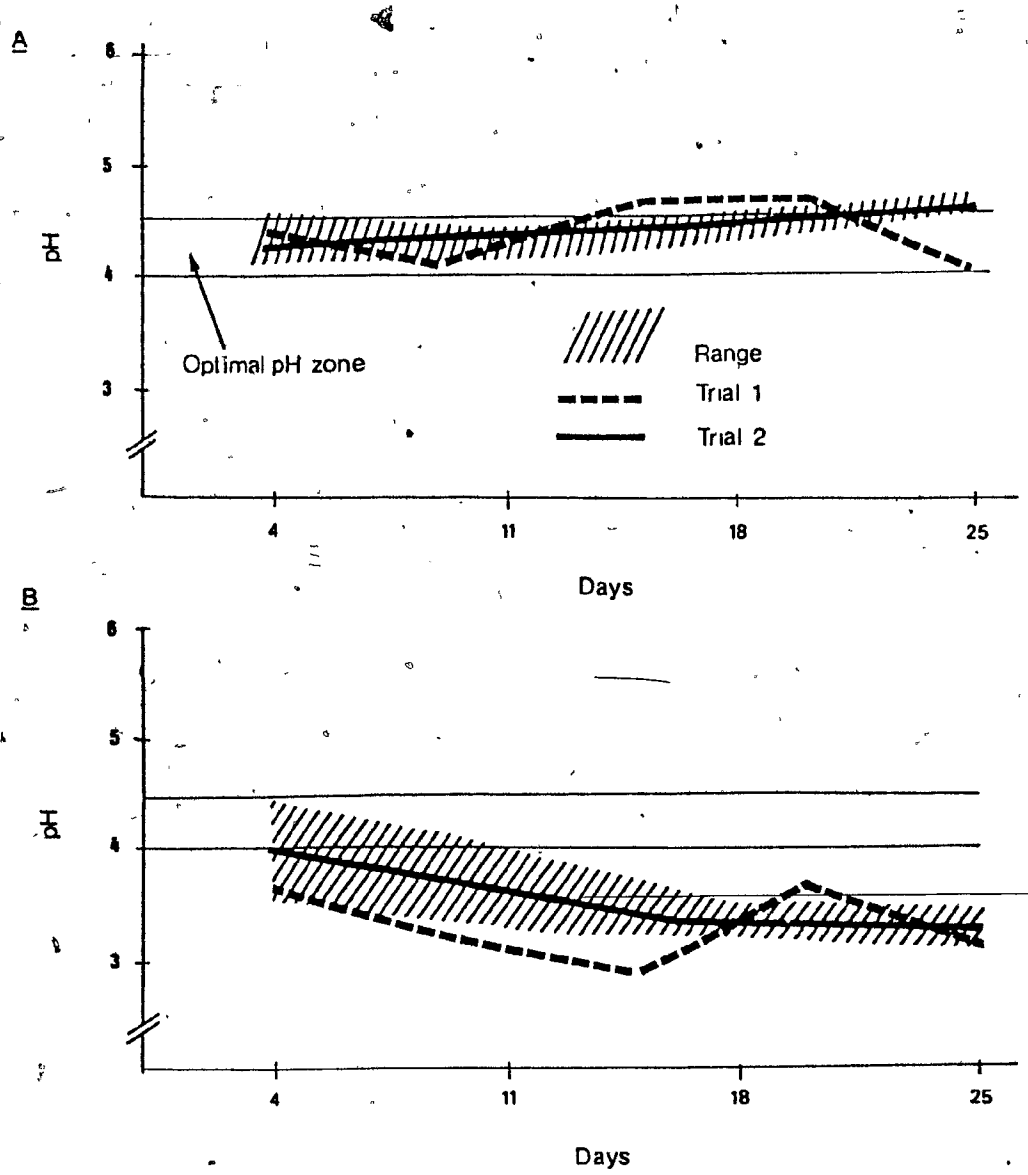
PHASE	TRIAL	TREATMENT: STORAGE TEMP.	TREATMENT: TYPE OF FERMEN.	TREATMENT: DAY OF APPLIC. OF SORB.AC.	ADG (g)	CONSUMED PER calf/day: DIGEST. PROTEIN (g)	CONSUMED PER calf/day: DIGEST. ENERGY (MJ)	CONSUMED PER kg of gain: DIGEST. PROTEIN (g)	CONSUMED PER kg of gain: DIGEST. ENERGY (MJ)	RATIO OF DE (kJ): DP (g) IN THE COLOSTR.	RATIO OF DE (kJ): DP (g) IN COMP. RATION	RATIO OF DE cons. STARTER: COLOSTR.
I	2	S*	N**	--	325	131	9.8	402	30.0	68.3	74.7	1:1.5
I	2	S	S.ℓ.	--	398	153	12.0	383	30.1	71.4	78.4	1:1.07
II	1	S	S.ℓ.	--	199	123	9.4	619	47.0	71.1	76.0	1:2.14
II	1	S	S.ℓ.	4	301	139	10.1	460	33.4	65.4	72.6	1:1.58
II	2	W	S.ℓ.	--	190	118	8.4	618	44.2	66.3	71.4	1:2.41
II	2	W	S.ℓ.	4	207	132	8.9	636	43.0	61.8	67.6	1:2.41
II	3	S	S.ℓ.	--	78	105	8.1	1,347	103.6	72.7	76.9	1:2.18
II	3	S	S.ℓ.	1	248	123	9.3	496	37.4	69.9	75.5	1:1.72
II	3	S	S.ℓ.	4	239	124	8.7	516	36.5	63.8	70.7	1:1.79

1 Roman numeral pertains to the phase of research, Arabic number to the particular trial.

* S = Summer, W = Winter.

** N = Natural, S.ℓ. = Streptococcus lactis

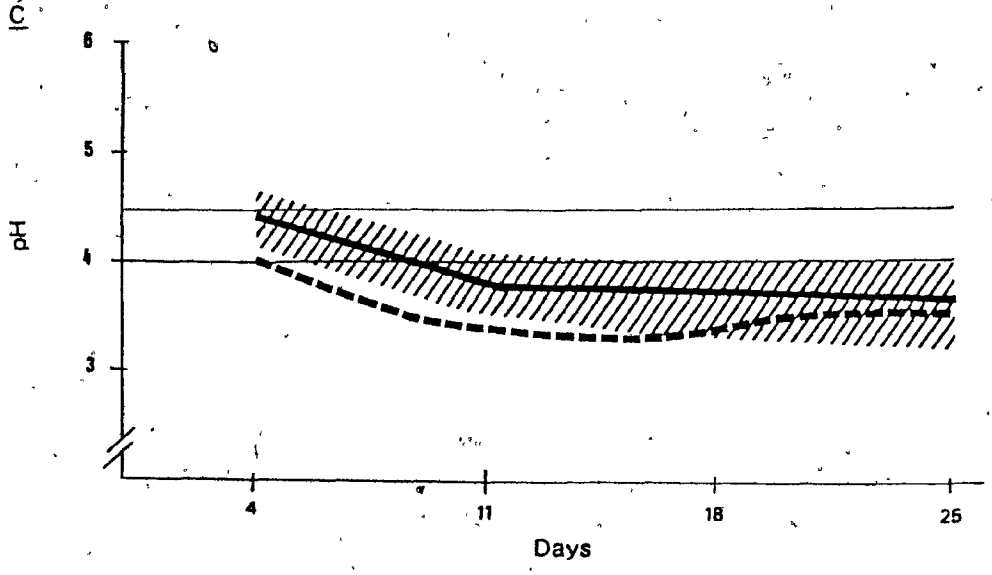
FIG 4-1.



pH changes in fermented colostrum as affected by microbial treatment with (A) - Streptococcus lactis, (B) - Lactobacillus bulgaricus and Streptococcus thermophilus, (C) Lactobacillus acidophilus, or (D) - naturally fermented.

FIG. 1 - 1.

C



D

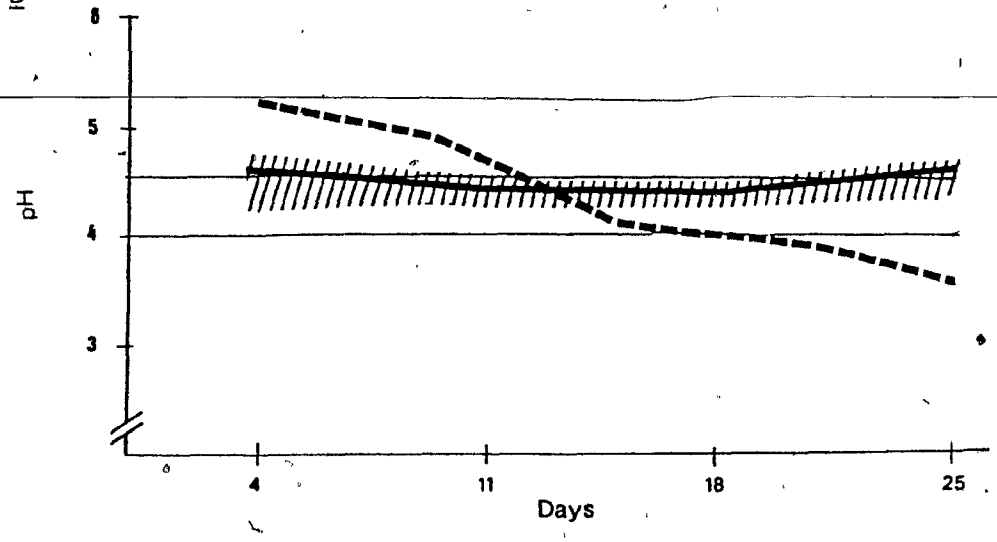
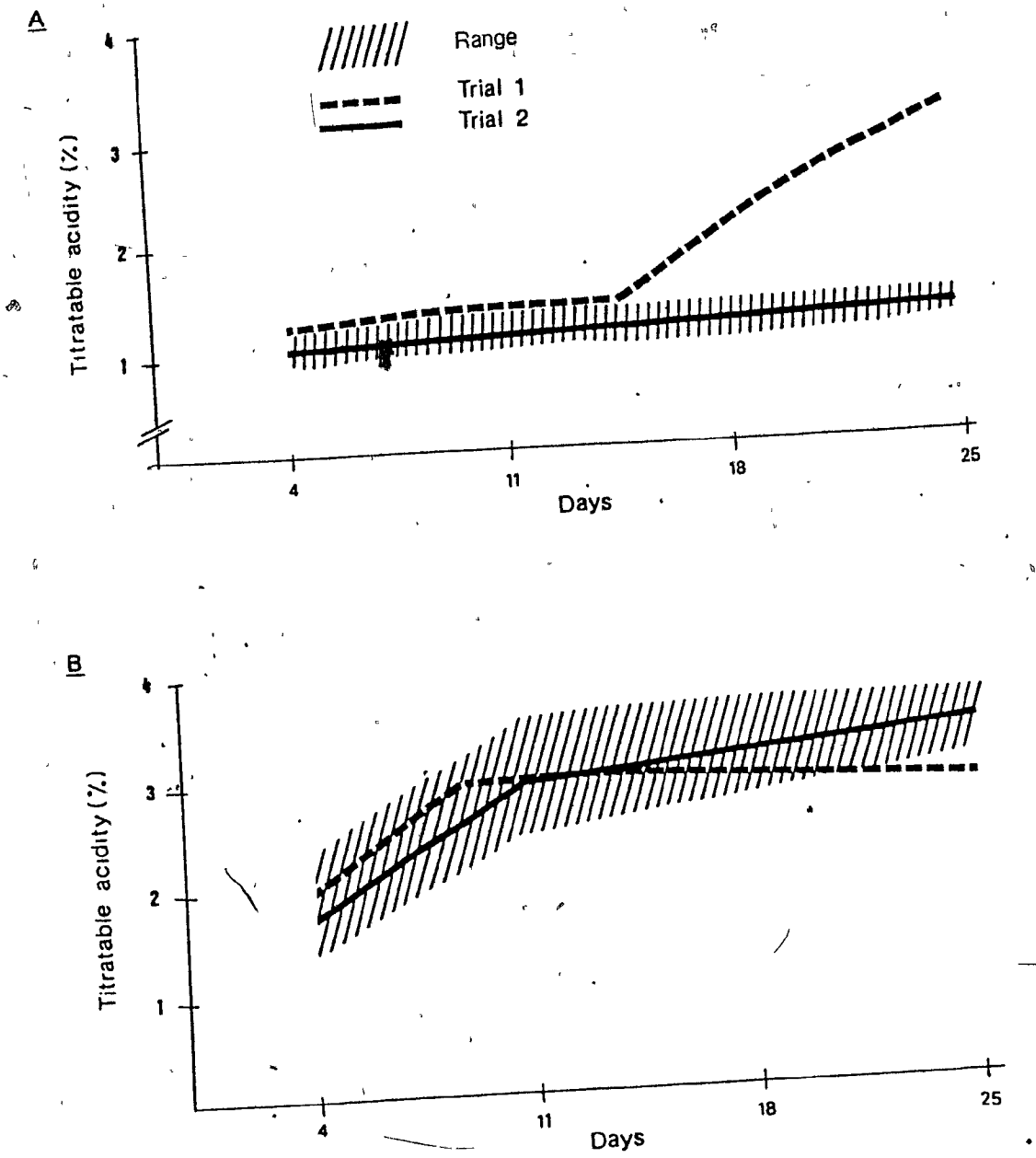


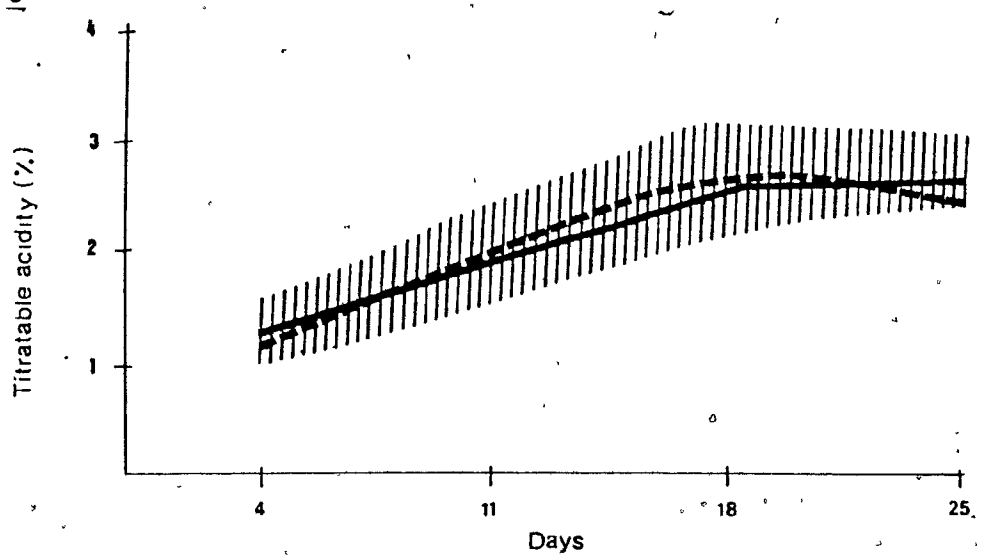
FIG 1-2



Titratable acidity changes in fermented colostrum as affected by microbial treatment with (A)- Streptococcus lactis, (B)- Lactobacillus bulgaricus and Streptococcus thermophilus; (C)- Lactobacillus acidophilus or (D)- naturally fermented.

FIG 1 - 2

C


Titratable acidity (%)
4
3
2
1
4
11
18
25
Days

D

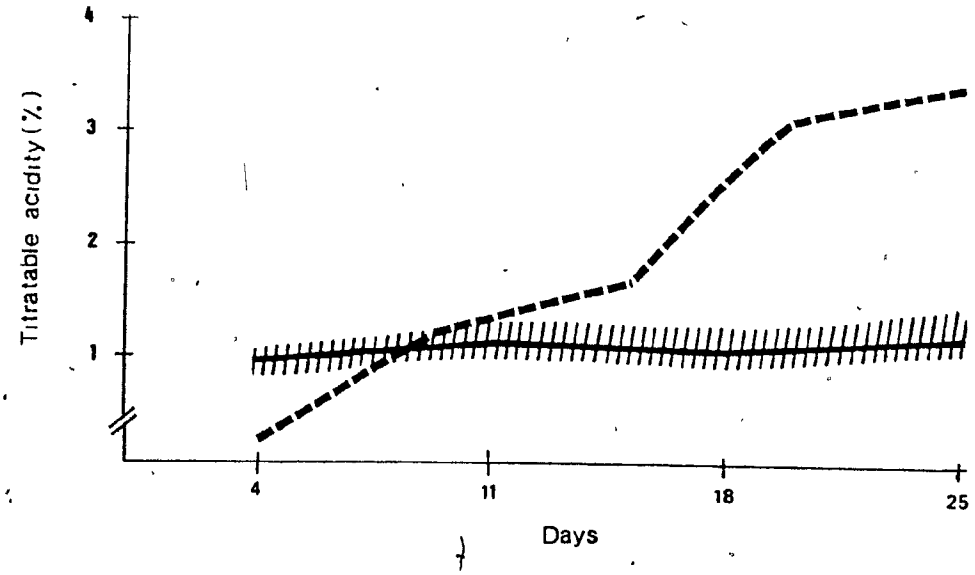
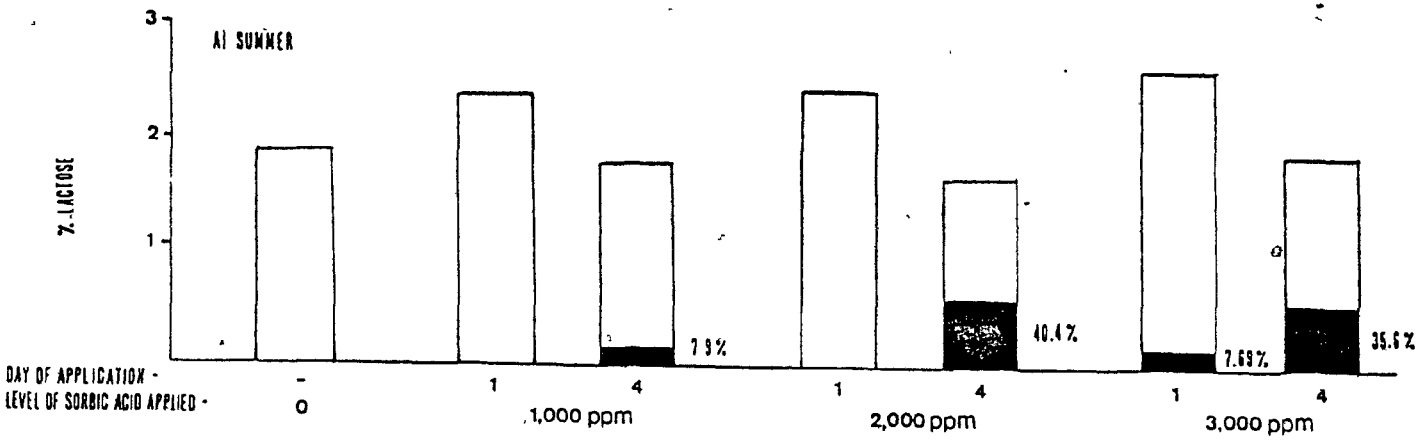

Titratable acidity (%)
4
3
2
1
4
11
18
25
Days

FIG. III -1.

INITIAL □ AND RESIDUAL ■ CONTENT OF LACTOSE IN COLOSTRUM TREATED WITH VARIOUS LEVELS OF SORBIC ACID, APPLIED AT DAY 1 OR 4 AND STORED FOR 28 DAYS IN SUMMER OR SIMULATED WINTER CONDITIONS

*INITIAL SAMPLES TAKEN AFTER 4 DAYS OF FERMENTATION



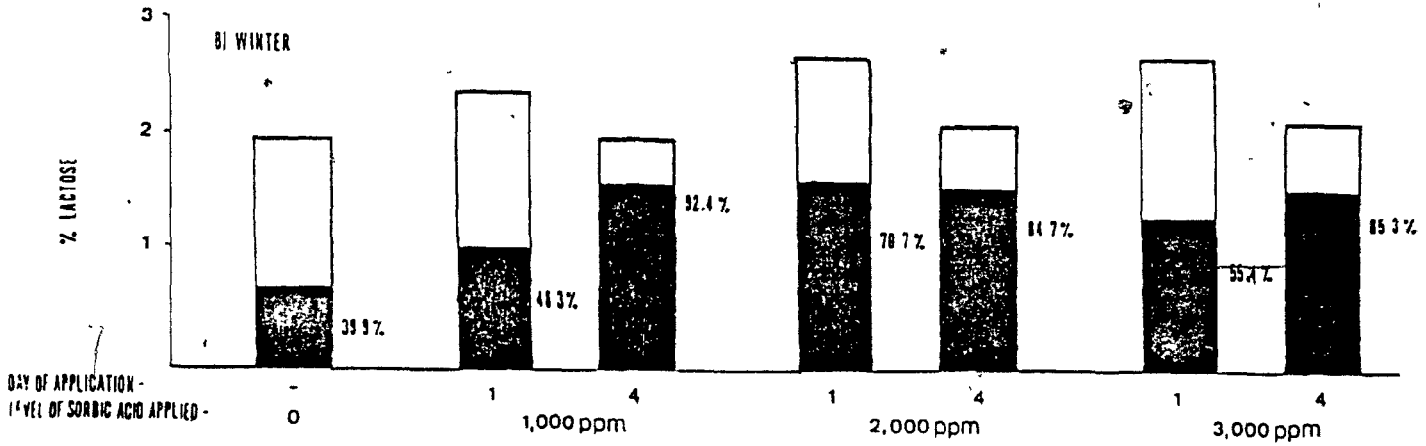


FIG III-2 INITIAL* ☐ AND RESIDUAL ■ CONTENT OF PROTEIN IN COLOSTRUM TREATED WITH VARIOUS LEVELS OF SORBIC ACID, APPLIED AT DAY 1 OR 4 AND STORED FOR 28 DAYS IN SUMMER OR SIMULATED WINTER CONDITIONS

*INITIAL SAMPLES TAKEN AFTER 4 DAYS OF FERMENTATION

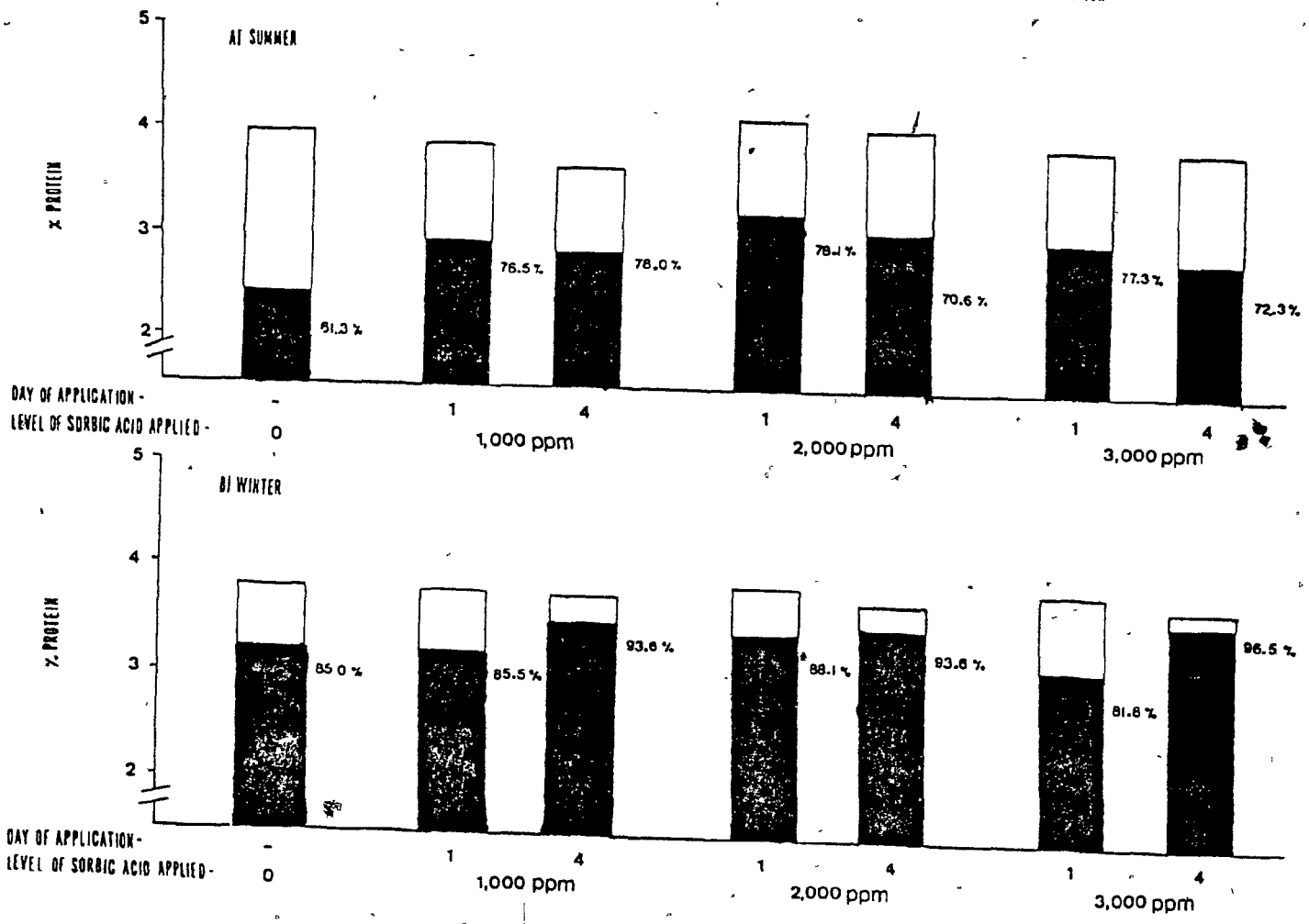


FIG VII-3. INITIAL* ☐ AND RESIDUAL ■ CONTENT OF FAT IN COLOSTRUM TREATED WITH VARIOUS LEVELS OF SORBIC ACID, APPLIED AT DAY 1 OR 4 AND STORED FOR 28 DAYS IN SUMMER OR SIMULATED WINTER CONDITIONS

*INITIAL SAMPLES TAKEN AFTER 4 DAYS OF FERMENTATION

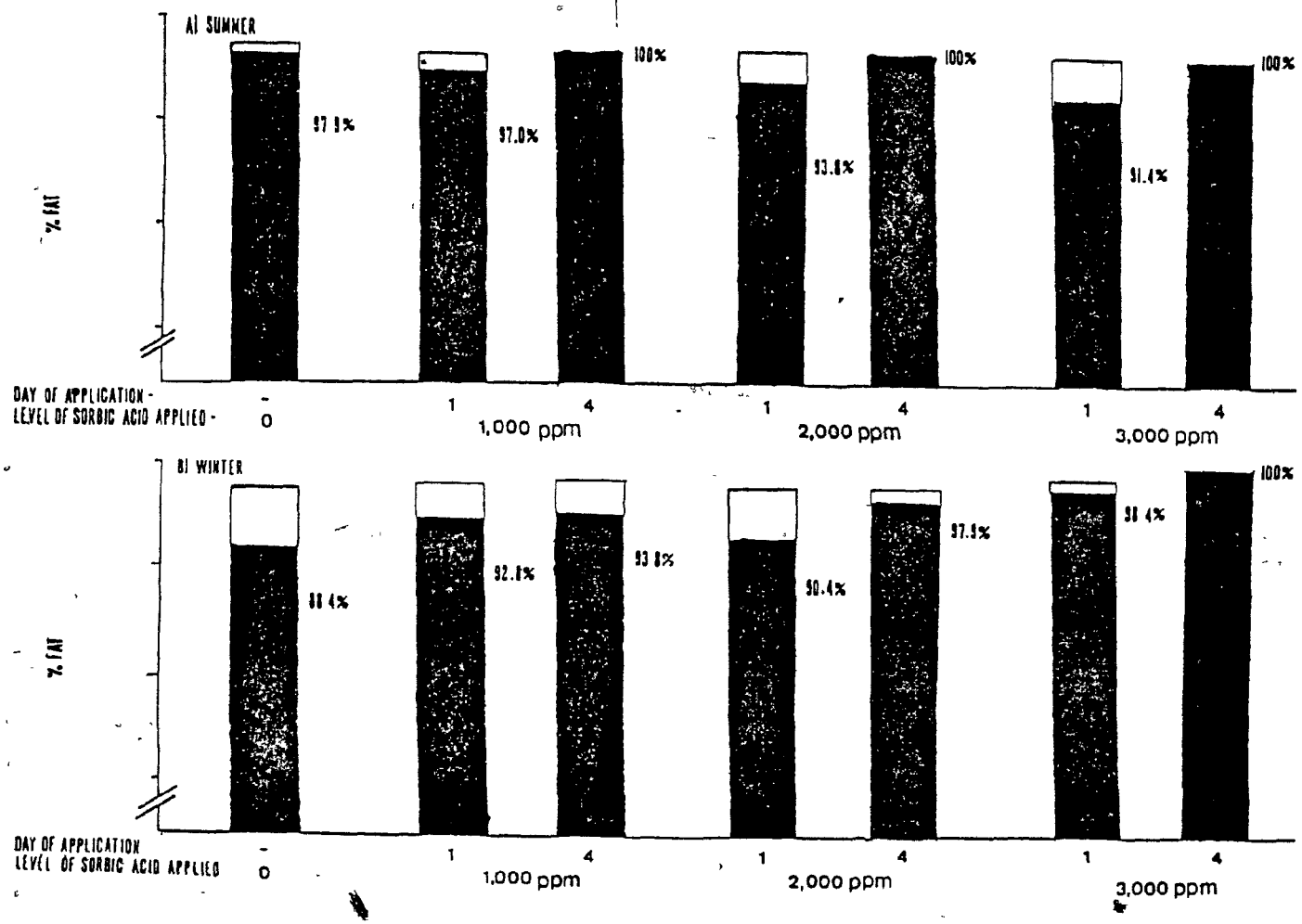


FIG.IV - 1.

REGRESSION OF D.E. CONSUMPTION ON ADG FOR CALVES

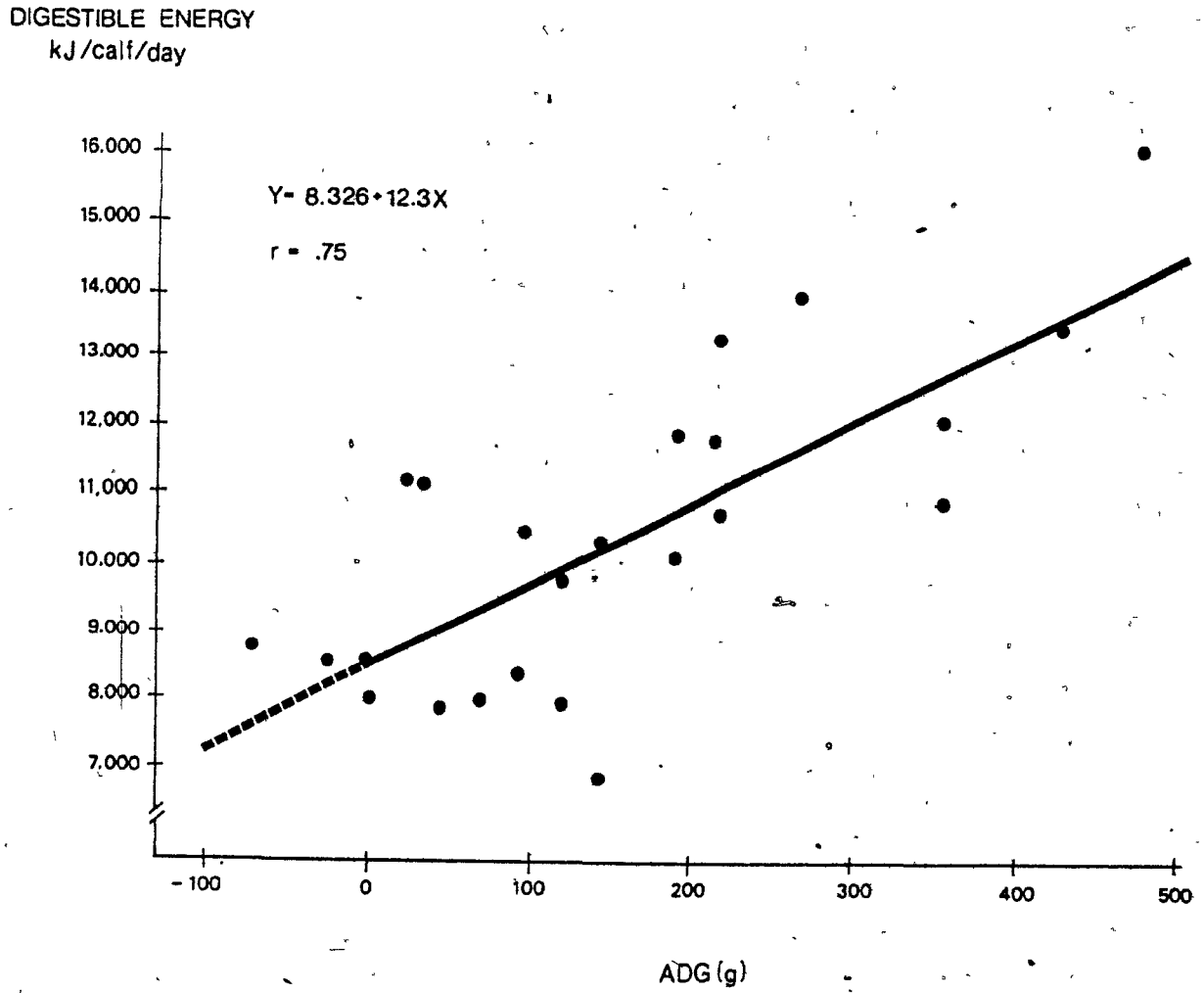
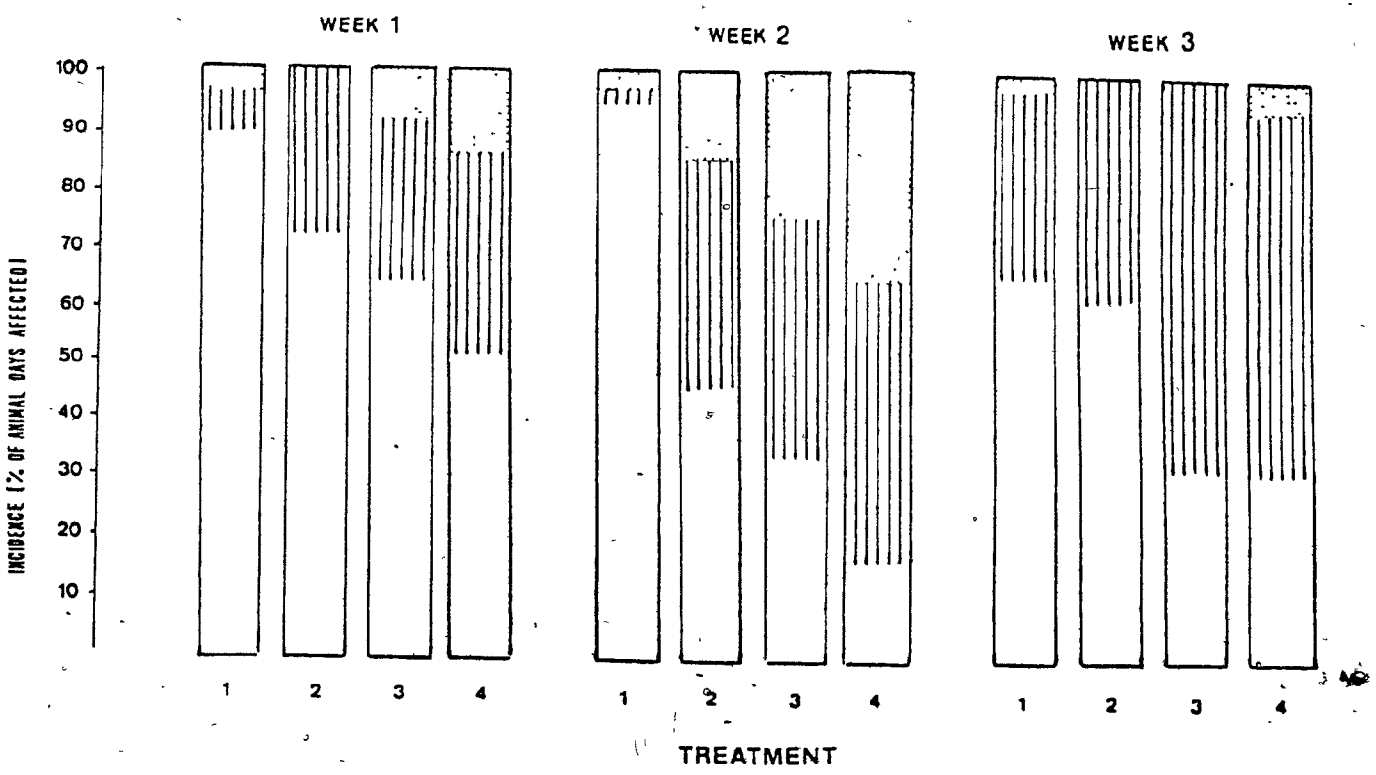


FIG.IV - 2.

PHYSICAL APPEARANCE OF FECES



APPENDIX I

PRACTICAL IMPLICATIONS

(a) Step by step procedure (Appendix 2) for farmers was prepared and distributed at the occasion of Eastern Ontario Dairy Day, 1978.

(b) Approximately 350 calves were raised at Kemptville College of Agricultural Technology on various fermented colostrum programs, of which only one was lost prior to weaning. Low incidence of digestive disorders, characterizing the fermented colostrum feeding system, described in Appendix 2, seems to be an additional feature of the system.

(c) It is estimated that approximately 60 farmers in Ontario are applying the program in its entirety (inoculation with S. lactis and application of sorbic acid).

(d) It appears to be an ideal system for raising Holstein male calves, destined for a heavy calf production or feedlot stockers. Daily intake of nutrients from colostrum was calculated so that it meets approximately the maintenance requirements of newborn calves. As the daily allotment is fed once a day, the abomasum remains devoid of colostrum for approximately 16 hours per day. All this results in early intake of calf starter. The reticule-rumen is functioning early (2-3 weeks) and the calves are known for highly efficient conversion of inexpensive solid diet into gains.

APPENDIX 2

FERMENTED COLOSTRUM FEEDING - STEP BY STEP PROCEDURE

- Immediately after birth, the calf's navel should be disinfected with iodine or an organic disinfectant such as Hibitane. The calf should be given a 2cc (ml) injection of vitamin A, D, and E preparation and 1 ml of selenium and vitamin E combination as protection against White Muscle Disease.

- As soon as possible, preferably within 2 hours after birth, make sure that the calf sucks its mother to get a good feeding of the first colostrum.

- Following is the feeding schedule up to weaning:

Day	Fresh Colostrum		Streptococcus Lactis Fermented Colostrum		Mixed Hay	18% Starter
	A.M.	P.M.	A.M.	P.M.		
1st	sucking	4 lb				
2nd	4 lb	4 lb				
3rd	4 lb	4 lb				
4th	4 lb	4 lb				
5th			5 lb	5 lb	free choice	free* choice
6th up to 25th			5 lb			

* To encourage the intake of starter, offer it occasionally by hand for first few days.

- Obtain a 16 gallon plastic pail, with a lid, and make sure it is thoroughly cleaned and disinfected.

- During the first four to five days of the calf's life, the surplus colostrum from the calf's mother should be collected and stored in the plastic pail. About 110 to 120 lbs will be needed for successful calf weaning. Most cows will produce upwards of 150 lbs of unmarketable but extremely nutritious milk after calving.

- To ensure the correct fermentation, leading to more palatable, uniform and well preserved product, add one cupful of Streptococcus lactis cultured buttermilk to the first surplus colostrum. This is ordinary buttermilk which can be purchased at any dairy or dairy bar. Keep the colostrum at 25 - 32°C for first 4 days and at 10°C or lower thereafter.

- To keep the surface mold growth under control and increase the daily gains of calves, add 0.1% of potassium sorbate to the volume of colostrum and mix it thoroughly. For 120 lbs of colostrum you should add approximately 55 g or 2 ounces of potassium sorbate. (For details, contact Kemptville College of Agricultural Technology, Animal Science Section).

- It is best to have separate containers for the colostrum for each cow. Normally, this would be for the first 6 - 8 milkings. It is not essential that calves be restricted to their own mother's colostrum.

- From day 5 to day 25, fermented colostrum is the only "milk" fed. While once a day feeding has given satisfactory results, twice a day feeding may be more acceptable. Feed 3 lbs of fermented colostrum twice a day. Add 2 lbs of hot water as it is necessary to warm it to body temperature. On the once a day feeding do not dilute the colostrum with water, but dip the pail into another container with hot water. The capacity of a calf's abomasum at an early age is about 5 lbs.

- At each feeding time, the contents of the plastic container should be thoroughly mixed so that the quantity fed will be of uniform content.

- When the calf reaches 25 days of age, it should be consuming sufficient calf starter (500 g/head/day), hay and water that it may be weaned. This may or may not be in accordance with the supply of fermented colostrum available.

- High quality palatable calf starter and good hay should be offered to calves at about 5 days of age. Shortly after colo-

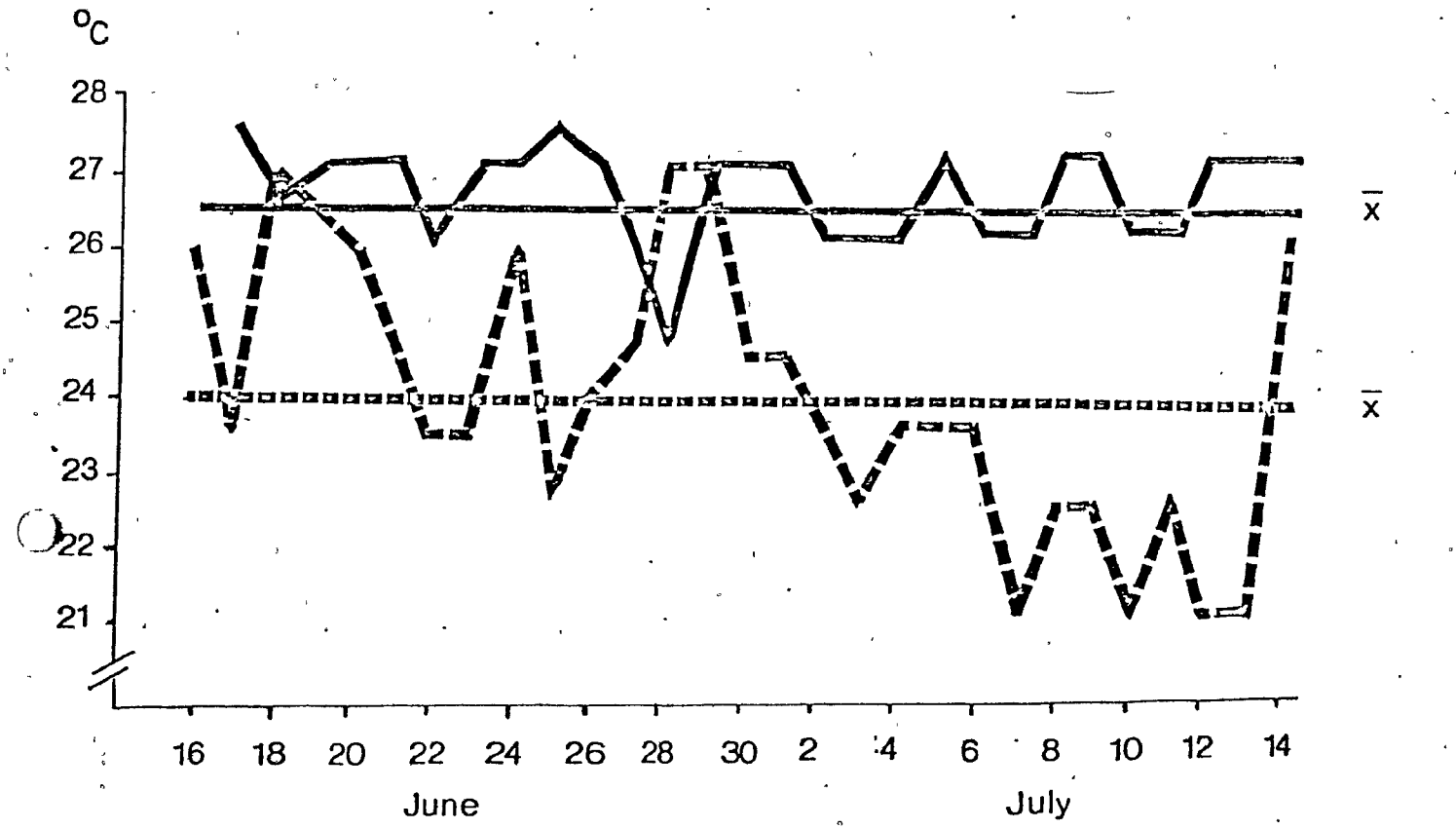
strum feeding, when the calf is still looking for additional feed, it should be offered a handful of starter so that it will get acquainted with solid food as soon as possible. Starter which was found very attractive to calves on fermented colostrum has the following composition:

Ingredient	Lbs
Shelled corn (ground through ½" screen)	520
Barley (rolled)	500
Wheat Bran	300
Soybean Oil Meal	600
Molasses	200
Limestone	40
Salt	10
Vitamin ADE	2 - 2½

The general texture of the mixture should be coarse rather than fine.

- Calf pails used for colostrum and starter should be cleaned out daily so as not to become musty with old feed.

FIG. III-4. TEMPERATURE VARIATIONS DURING SUMMER STORAGE



Legend:

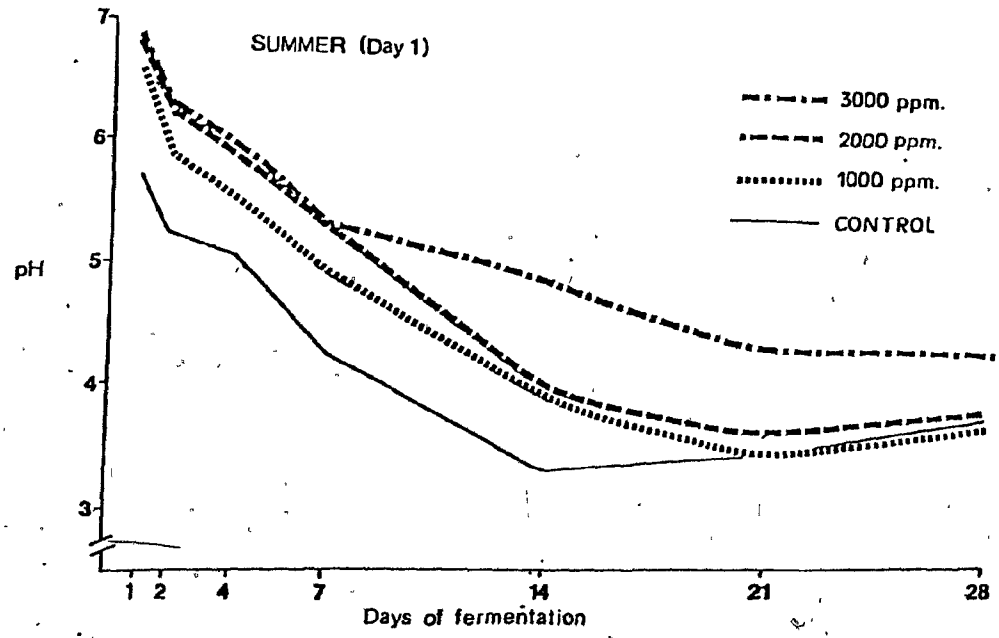
Afternoon temperature

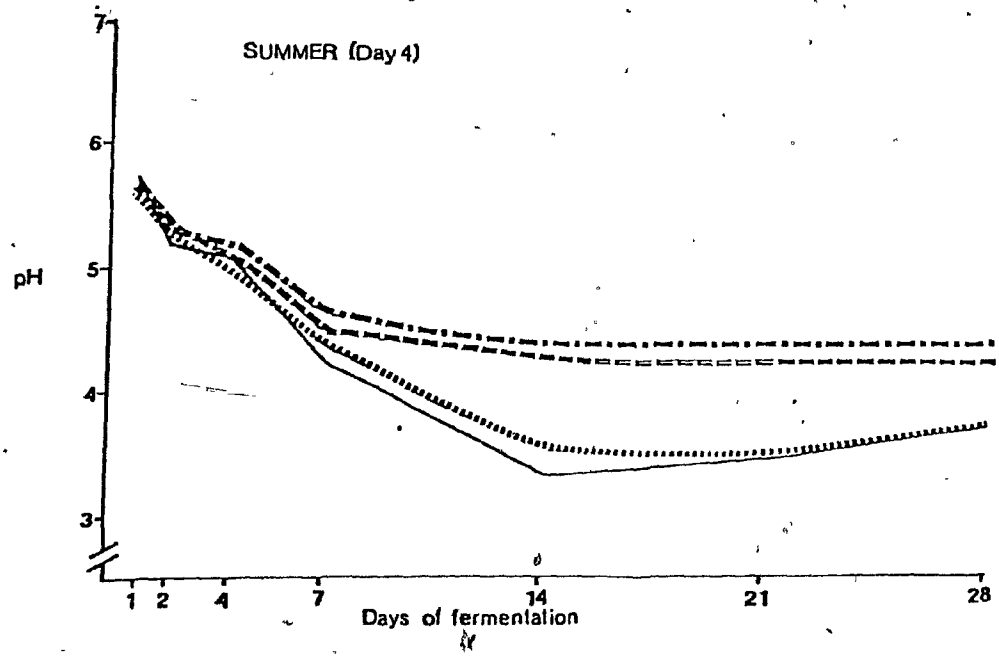
Forenoon temperature

APPENDIX 3

FIG. III - 5

EFFECT OF TIME OF STORAGE AND LEVEL OF APPLICATION OF SORBIC ACID ON pH CHANGE

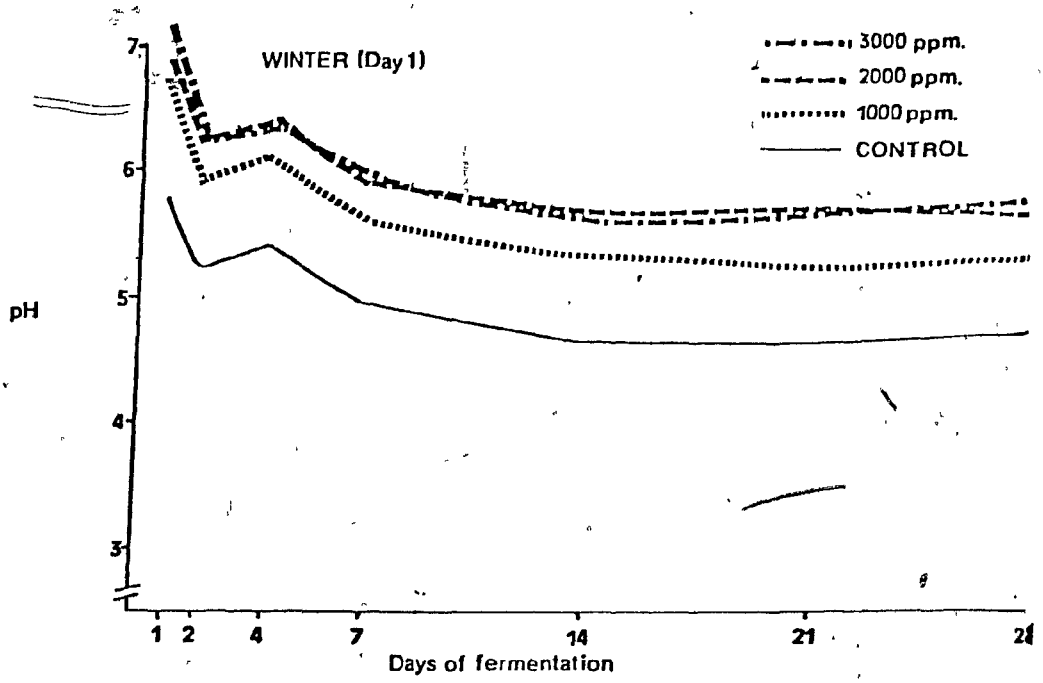


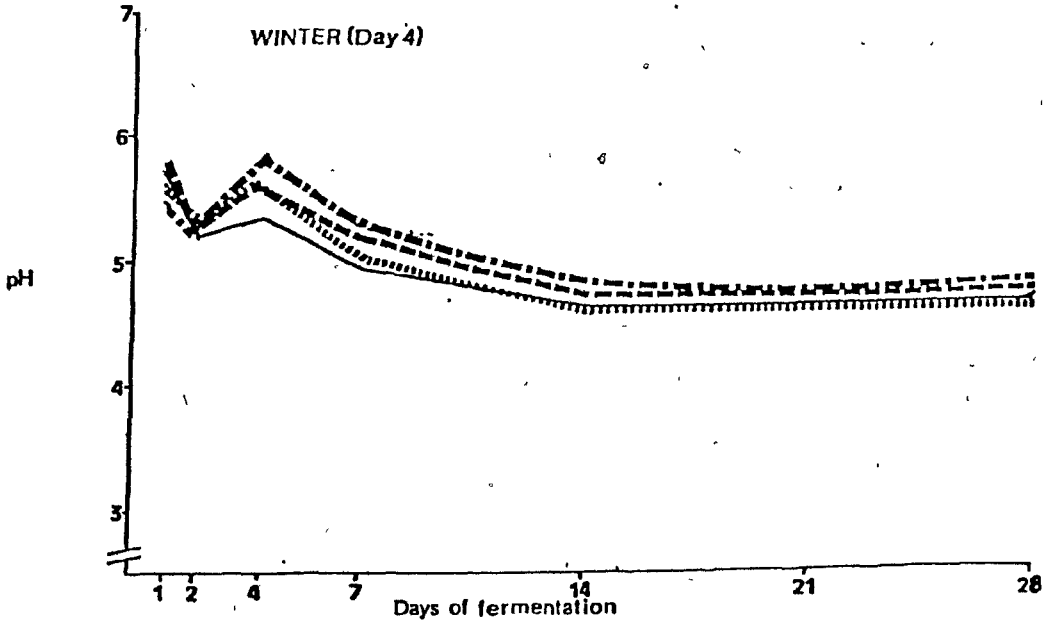


APPENDIX 3

Fig III-6.

EFFECT OF TIME OF STORAGE AND LEVEL OF APPLICATION OF SORBIC ACID ON pH CHANGES



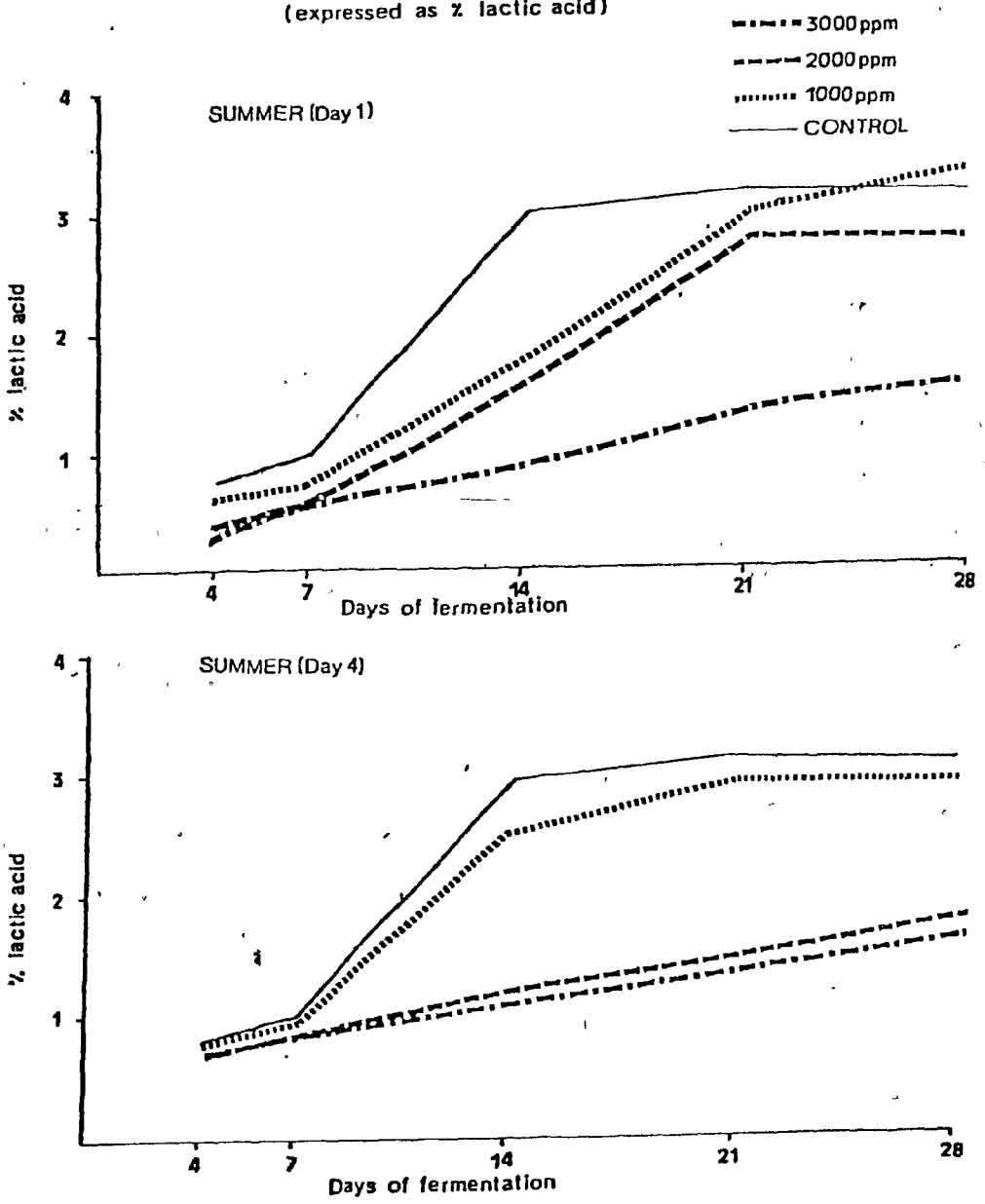


APPENXIX 3

FIG III-7.

EFFECT OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON TITRATABLE ACIDITY

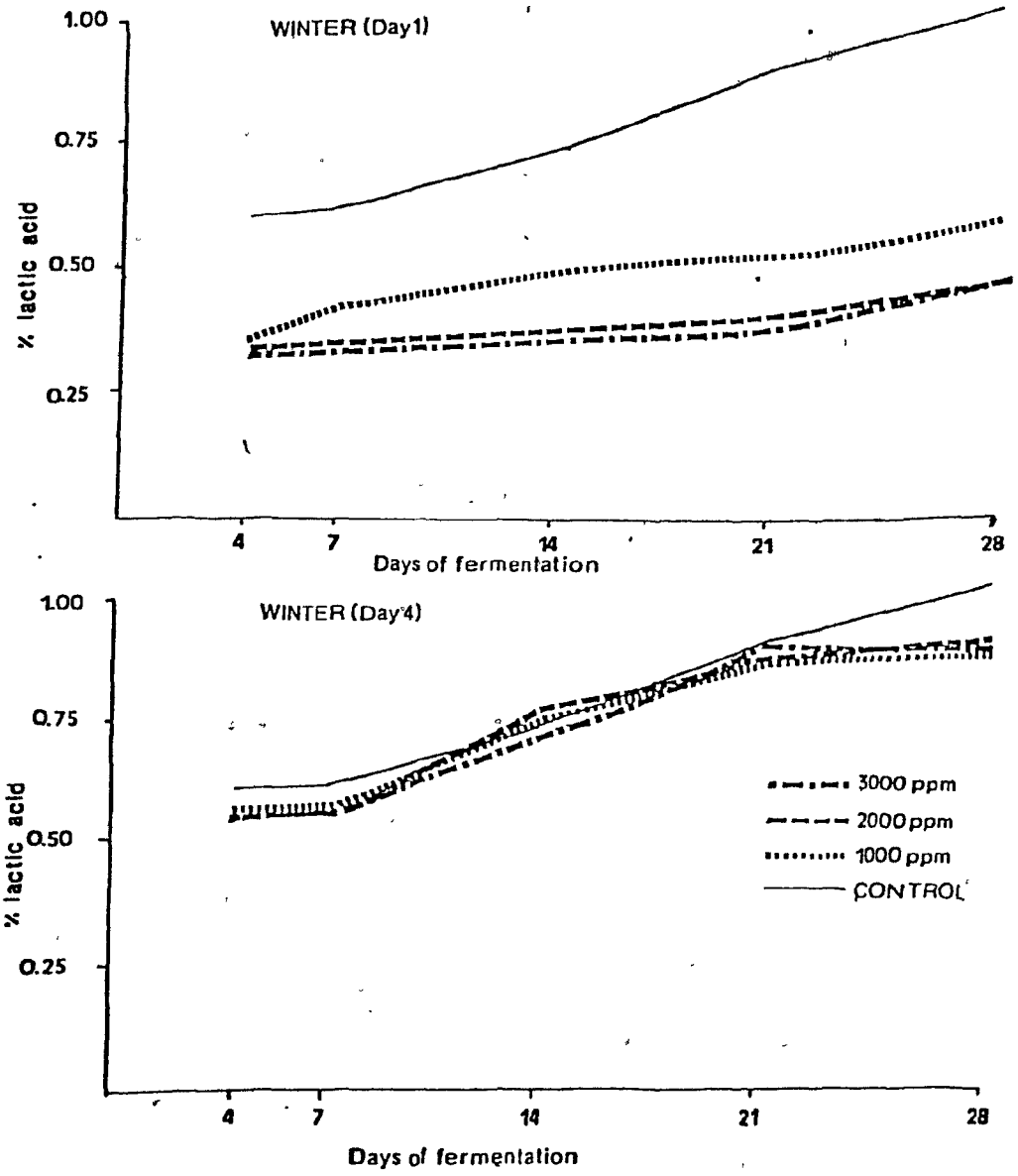
(expressed as % lactic acid)



APPENDIX 3

FIG III-8.

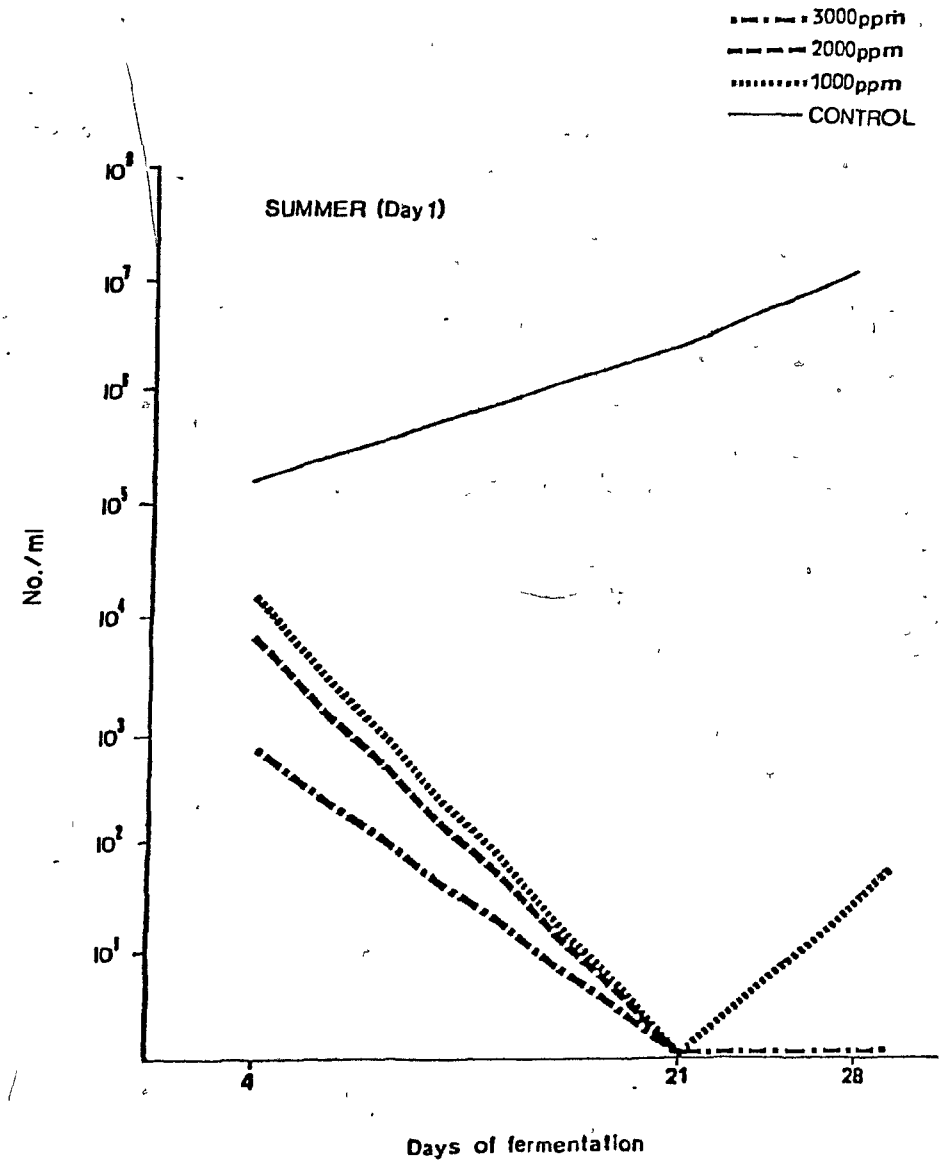
EFFECT OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON TITRATABLE ACIDIT
(expressed as %. lactic acid)



APPENDIX 3

FIG 811-9.

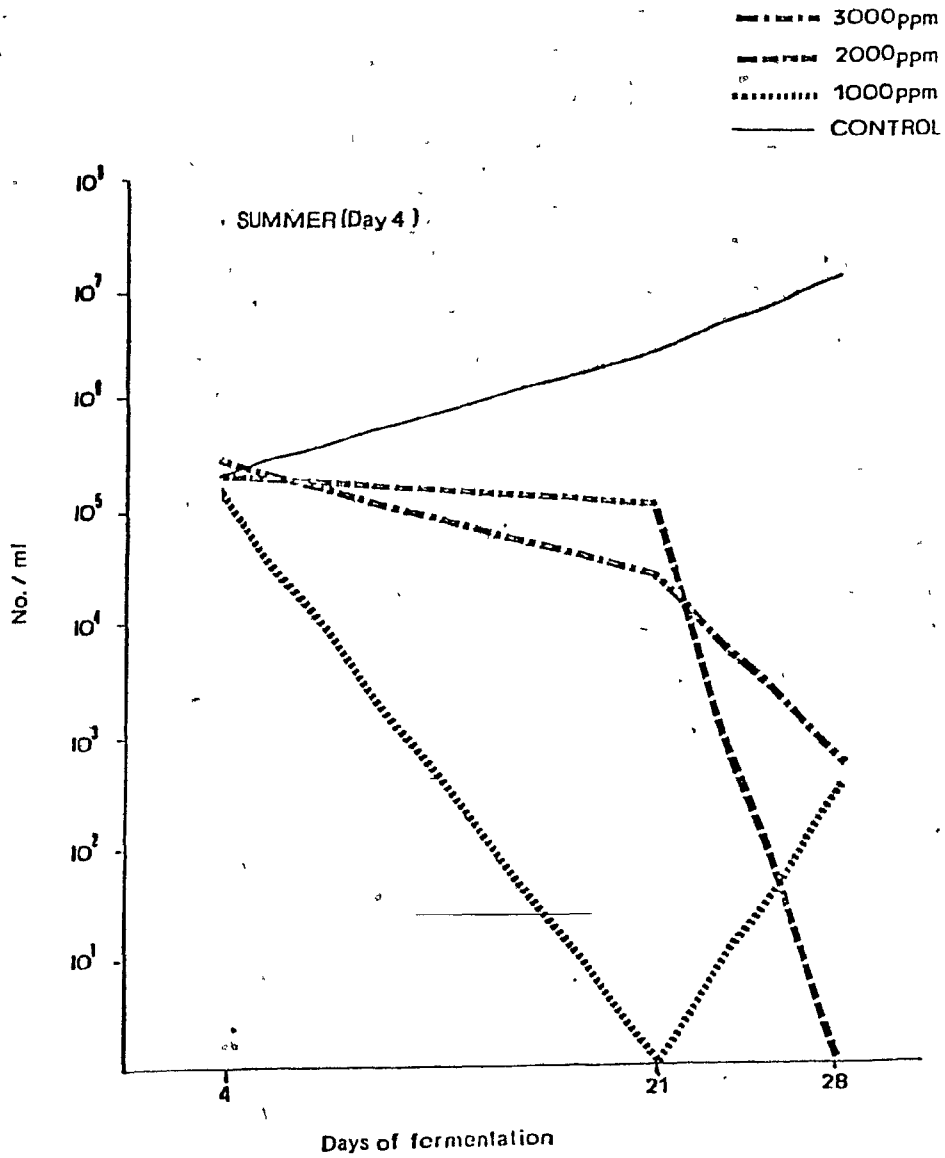
EFFECT OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON THE NUMBER OF YEASTS AND MOLDS



APPENDIX 3

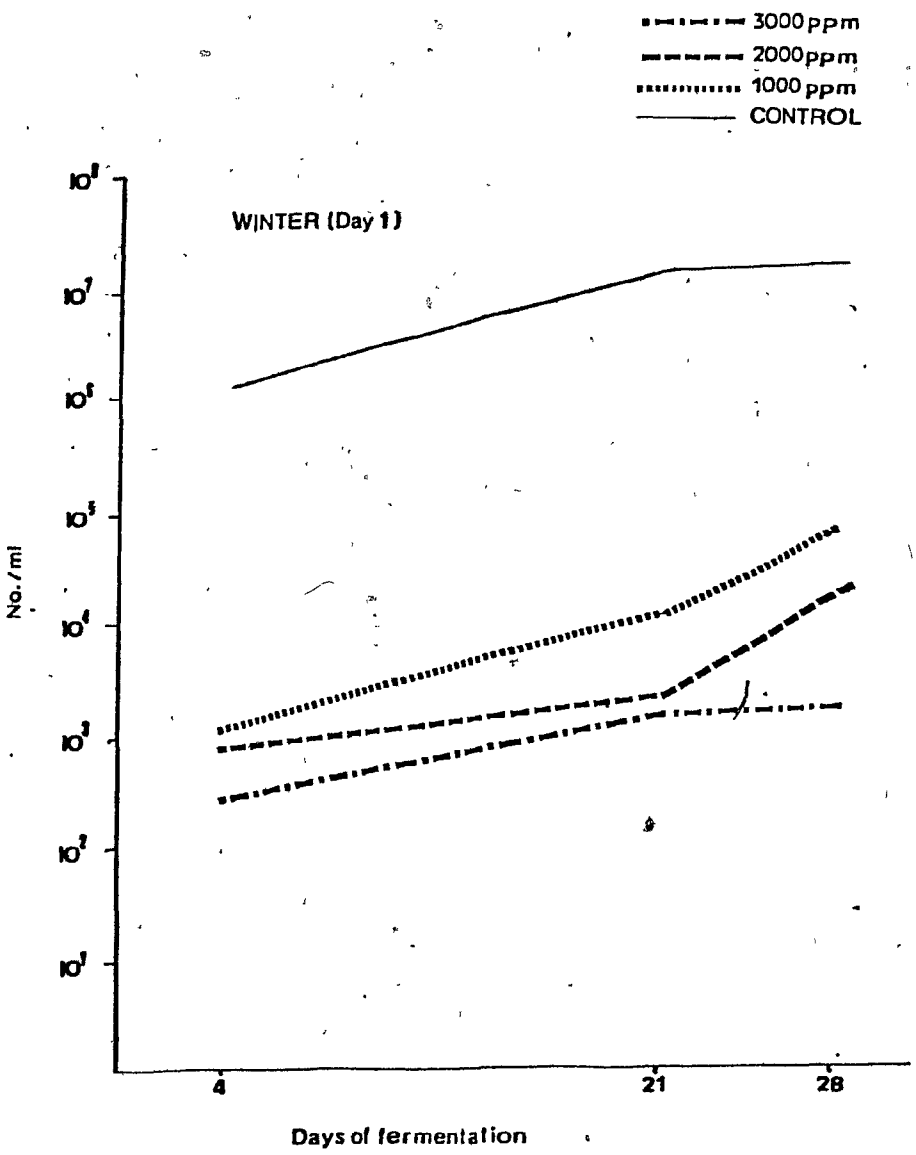
RS 111-10

EFFECTS OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON THE NUMBER OF YEASTS AND MOLDS



APPENDIX 3

FIG. III - 11.
EFFECT OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON THE NUMBER OF YEASTS AND MOLDS



APPENDIX 3

FIG. III-12.
EFFECT OF DAY AND LEVEL OF APPLICATION OF SORBIC ACID ON THE NUMBER OF YEASTS AND MOLDS