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JOURNAL OF DAIRY SCIENCE

VOLUME XXXVIII October, 1955 NUMBER 10

INFLUENCE OF CERTAIN VITAMIN K COMPOUNDS ON LACTIC ACID DEVELOPMENT IN MILK[1]

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Certain of the vitamin K compounds are known for their antibacterial effects as well as anti-hemorrhagic properties. The inhibitory action of these compounds against bacteria has been found to be associated with the quinone structure. The earlier work of numerous investigators establishing that quinones exhibit bacteriostatic properties has been reviewed by Colwell and McCall *(4)*. Armstrong *et al. (2)* found that menadione (2-methyl-1,4-naphthoquinone), a synthetic vitamin K, also known as vitamin K_3, exhibited bacterial inhibition against Gram-positive organisms. Colwell and McCall *(3)* reported earlier that 25 p.p.m. of menadione was bactericidal to *Escherichia coli* in a chemically defined medium. Armstrong and Knutson *(1)* found that 3 to 5 mg. of menadione per 100 ml. saliva-glucose mixture and corresponding amounts of benzoquinone caused a progressive decrease in the *Lactobacillus acidophilus* counts in samples withdrawn at hourly intervals over a 4-hour incubation period.

Some of the vitamin K compounds have received consideration as preservatives for foods. Tomiyasu *et al. (10)* reported that vitamin K_3 preserved raw milk at 4° C. for 1 month at a dilution of 1:20,000. In investigating various substances for inhibition of mold growth in butter, Tanaka and Ishii *(9)* reported that best results were obtained with vitamin K but that it was necessary to avoid concentrations that affected the flavor. Faggioli *(5)* found that vitamin K_5 (4 amino-2-methyl-1-naphthol) exhibited anti-mycotic activity against growth of eight molds at a concentration of 0.1%. He reported that vitamin K_5 had the advantages of being low in toxicity and of not adversely influencing food flavors but had the disadvantage of oxidation by atmospheric oxygen, causing a darkening effect in broths, apricot sirup, tomato conserve, and wine.

Kelley and Dittmer *(7)* reported that cows receiving 25 mg. of menadione per day produced milk that remained sweet for 18 to 24 hours at 37° C., whereas milk from cows not receiving menadione generally soured in about 12 hours when held at the same temperature. They indicated also that direct addition of menadione to milk at the rate of 0.1 p.p.m. retarded the rate of souring of milk at 37° C. However, they call attention to the fact that during one period

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[1] Florida Agricultural Experiment Station Journal Series, No. 366.

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of the trials neither the feeding of menadione nor its addition to milk retarded the rate of souring. Other variations in results within the herd and for individual cows also were observed by them, and they therefore suggested the need for further research into the bacteriostatic properties of menadione before definite conclusions could be drawn relative to this compound as investigated by them.

This research was undertaken to investigate further whether feeding vitamin K compounds to cows has an influence on the rate of lactic acid fermentation in the milk produced by them and to establish concentrations of these compounds, when added to milk, that are required to retard lactic acid development by commercial dairy cultures.

EXPERIMENTAL PROCEDURE

The two vitamin K compounds used in this study were menadione (2-methyl-1,4-naphthoquinone) and menadione diphosphate (tetra-sodium-2-methyl-1,4-naphthohydroquinone diphosphoric acid ester). Menadione is soluble in fat but is only slightly soluble in water. Menadione diphosphate is readily soluble in water.

As there was some question relative to the rate at which these materials could be fed with safety to dairy cows, in the first trials menadione was fed at rates of 25 mg. per cow per day; Kelley and Dittmer *(7)* had successfully fed menadione at rates of up to 200 mg. per cow per day with no apparent ill effects. In other feeding trials, the rate was increased to the maximum of 600 mg. of menadione per cow per day. Information on the toxicity to cattle of menadione diphosphate when administered orally is not available, but Foster *(6)* reported that an injection of approximately 450 mg. per kilogram of body weight was lethal to mice.

Menadione was fed to 18 cows at levels ranging from 25 to 600 mg. per cow per day for periods ranging from 11 to 45 days. Menadione diphosphate was fed to six cows at levels ranging from 1 to 10 g. per cow per day for periods ranging from 5 to 24 days. The various feeding trials were started in mid-October, 1953, and continued through May, 1954. Cows selected for the feeding trials were Jerseys and Guernseys comprising part of the University of Florida Agricultural Experiment Station herd. The cows were being fed the regular herd concentrate mixtures calculated to meet the requirements of the Morrison *(8)* standards, and the roughage portion of the rations at different times included improved fertilized pasture grasses, corn silage, or alfalfa hay.

Individual portions of the vitamin K materials were weighed, mixed into approximately 50 g. corn meal and blended into 2 lb. of the concentrate that each cow was to receive. Usually all of this offering was consumed before the remainder of the concentrate was fed. Milk yields were not affected by such feeding practices. When the cows were fed at the rate of 600 mg. per cow per day, the menadione was given twice each day at the rate of 300 mg. per feeding. In all other trials the material was fed daily or once every other day at the time of milking. Most of the cows consumed the offerings without any apparent objections, but a few cows were somewhat hesitant in eating even with the lowest levels

of menadione in the feed. There were a few refusals when 300 mg. of menadione was offered in 2 lb. of concentrate, but when additional concentrate was sprinkled on the surface of the experimental ration, usually most of the offering was consumed. These observations would indicate that the bitter and irritating properties of menadione are detected by cows.

The regular practice of milking was followed, the milking machines being cleaned and sanitized in the manner usually practiced with large herds. One-quart samples of milk were collected by pouring the milk directly from the milking machine pail into glass quart bottles recently washed in a commercial bottle washer. Control samples were collected from other cows on the same feeding and milking schedule but not fed the vitamin K compounds. Samples were delivered to the laboratory in less than 1 hour after collection. As total bacteria counts generally ranged from 1,000 to 3,000 per milliliter, occasionally up to 10,000, and as preliminary studies had indicated that frequently the milk collected was almost, if not entirely, free of the lactic acid–producing microorganisms, it was considered desirable to divide the samples into two portions, one portion to be inoculated with commercial buttermilk cultures and the other not inoculated, the rate of inoculation being 0.0001% to assure the presence of an estimated 100 to 1,000 lactic acid–producing streptococci per milliliter of milk. Each portion was incubated at 30° C., and titratable acidity determinations were made periodically to determine the rate of lactic acid production.

RESULTS AND DISCUSSION

Feeding trials. Periodic titrations of the various samples of milk were made to determine the number of hours required to reach 0.25% acidity. In Figure 1 are shown the results obtained on samples of milk collected from one cow on experiment and on control samples from another cow during a 45-day feeding period. This is a typical example of the day to day variations in rates of lactic acid development. Although the data are for samples of milk of only one cow receiving menadione and one control cow, the general relationships are typical

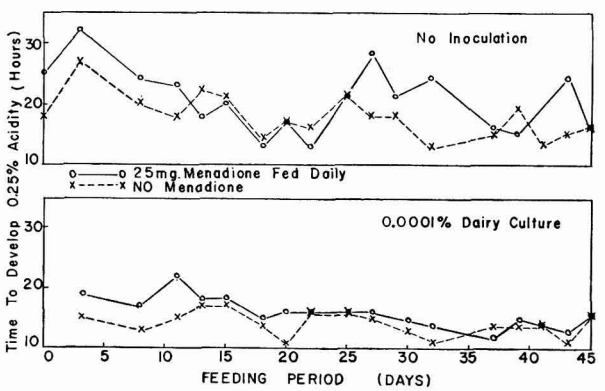

FIG. 1. Lactic acid development in raw milk at 30° C. when a cow received 25 mg. daily of menadione in the feed.

of the data for milks of the other cows receiving menadione and menadione diphosphate at the various levels. It was observed that there was considerable variation from day to day in the length of time required to reach 0.25% acidity for the noninoculated samples of milk from the control cow. Some samples required as long as 48 hours to develop 0.25% acidity. Apparently this was due to types of organisms other than the recognized lactic acid producers. After fermentation, very few samples showed the typical lactic acid fermentation characteristics but generally were gassy and had a putrid odor. Similar irregular daily variations were found in the different milks obtained at all levels of feeding. Because of these variations due to organisms other than lactic acid producers, it was not possible to draw any reliable conclusions relative to the rate of lactic acid development in the milk not inoculated with known amounts of lactic streptococci. The importance of inoculating the milk with small numbers of lactic acid–producing bacteria is shown in the two parts of Figure 1.

In the samples inoculated with lactic acid–producing organisms the rates of acid development were more reproducible from day to day, the rate depending upon the amount of inoculum used. Slight day to day variations inherent to this method of procedure could be expected when trials were conducted over a period of several months duration. The differences in the times required to reach 0.25% acidity in the samples of milk obtained from cows fed at all levels and corresponding controls in no instances exceeded the normal fluctuations of the controls. These studies, therefore, showed no effect in the rate of lactic acid production by lactic streptococci attributable to the feeding of menadione or menadione diphosphate to the cows.

Additive trials. Another phase of this investigation was concerned with determining quantitatively the antibacterial activity, against commercial dairy starters, of the vitamin K compounds when incorporated directly into milk. As menadione is practically insoluble in milk, levels from 0.3 to 50 mg. were dissolved in 1-ml. portions of ethyl alcohol before being added to 100-ml. portions of whole milk. Menadione-free controls, both with and without 1% alcohol, were included. Heat treatment was for 1 hour at 90° C., followed by cooling to 30° C. before inoculation. Trials were made with various levels of inoculation with lactic acid cultures, ranging from 0.0001 to 5%. A typical result of this phase of the study is shown in Figure 2. The effect of menadione in the milk at concentrations of 3 to 5 p.p.m. was evidenced by a measurable reduction in the rate of lactic acid development. Similar results were obtained when lower percentages of inoculation and lower incubation temperatures were used except that longer periods of incubation were required for the acid to develop. Similar results also were obtained when using skimmilk as the medium. The direct addition of 10 p.p.m. of menadione to milk extended the length of time to reach 0.25% acidity by approximately 3 hours; 50 p.p.m. extended it by about 6 to 8 hours. To preserve milk for several days, levels greater than 100 p.p.m. would be needed; such a practice would be of no value in the dairy industry, since levels of 100 p.p.m. or more of menadione cause a slight discoloration and also impart an undesirable flavor to milk.

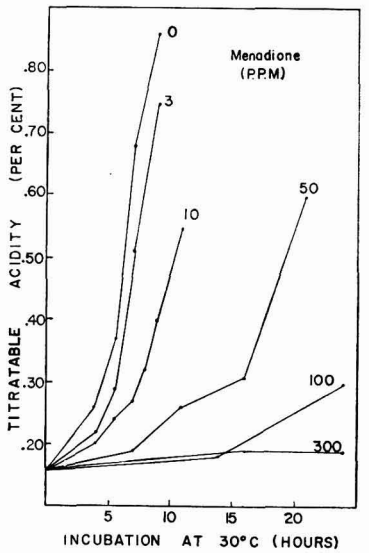

FIG. 2. Influence of menadione on rate of lactic acid development in milk inoculated with 3% dairy culture.

In trials with menadione diphosphate similar to those with menadione, it was found that this compound did not exhibit antibacterial activity even when used at rates as high as 1,000 p.p.m. in samples of milk inoculated with dairy starters.

SUMMARY

Bacteriological studies were made of milk obtained from cows fed various levels of menadione and menadione diphosphate. Twenty-four Jersey and Guernsey cows were used. The maximum levels fed per cow per day were 600 mg. of menadione and 10 g. of menadione diphosphate. Feeding periods ranged from 5 to 45 days. Samples of raw milk were incubated at 30° C. and titrated periodically for lactic acid content. To assure the presence of lactic acid–producing organisms in the raw milk, additional samples were inoculated with 0.0001% dairy culture. Data were compared with respect to the number of hours required to reach titratable acidities of 0.25%. None of the data obtained during the various feeding trials showed any reduction in the rate of lactic acid development that could be attributed to the practice of feeding menadione or menadione diphosphate to dairy cattle.

When added to milk in low concentration menadione exhibited antibacterial action against dairy starter microorganisms, as evidenced by a reduction in the rate of lactic acid development. Menadione added to milk in concentrations as low as 3 p.p.m. reduced the rate of lactic acid development. Levels of 100 p.p.m. or more were needed to stop acid development for periods as long as 24 hours at 30° C. Such concentrations in milk discolored it and imparted an undesirable flavor to the milk.

ACKNOWLEDGMENTS

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THE NUTRITIONAL REQUIREMENTS OF LACTIC STREPTOCOCCI ISOLATED FROM STARTER CULTURES. III. VARIATION IN GROWTH-PROMOTING PROPERTIES OF FRESH WHOLE MILKS

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Previous investigations *(1, 2)* demonstrated marked stimulation of lactic streptococci in synthetic media and milk after addition of liver fraction L and enzymatic digests of milk. Since liver L is considered an abundant source of peptides and similar compounds are liberated by enzymatic hydrolysis of milk proteins, the results suggested stimulation by peptides. It also was considered possible that a similar factor might be involved in the variation in rate of acid production frequently observed for lactic streptococcus starter cultures in different individual herd or cow samples of milk. This study, therefore, represents an attempt to correlate rate of activity of lactic streptococci with concentration in such milks of protein degradation products in the form of peptides, peptones, and proteoses. For the sake of brevity and to avoid misunderstanding in nomenclature in the following descriptions and discussion, the term peptides will be used to designate all the protein fragments occurring between amino acids and the complete proteins (caseins, albumins, and globulins).

EXPERIMENTAL METHODS

Two series of experiments were carried out to demonstrate that the peptide fraction in milk from different cows may cause marked variation in the growth of mixed strain lactic starter cultures and single strains isolated from them. The first or preliminary series represented an indirect approach in order to show the correlation between rate of growth in milk and an equated protein degradation factor (PDF) based on formol titrations and total nitrogen. The second represented a direct approach in which rate of growth was correlated with quantity of peptide in individual cow samples. In the preliminary study both normal and abnormal milks were included. Abnormal milks were considered those from animals with mastitis or animals on special rations or in early or late stages of lactation (less than 30 or more than 300 days). Results indicated that abnormal milks often did not show a positive correlation between rate of growth of lactic streptococci and the PDF. Therefore, the second series of trials included only normal milks.

In both series of studies pint quantities of representative samples were obtained from Holstein and Jersey cows in various stages of lactation. The milk from each animal was divided into two portions and immediately refrigerated. In order to minimize any microbiological or enzymatic action in the milk, it was

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chilled, skimmed, and tested as soon as possible. Five-ml. quantities of the skimmed milk from each animal were tubed in triplicate and heated for 5 minutes at 121.5° C. in the first series and pasteurized by steaming for 10 minutes in the second series. This milk was used for rate of growth tests of starter culture or individual strains.

Amino nitrogen determinations were made in triplicate on each of the refrigerated samples from each animal. It was observed that autoclaving (15 lb. for 5 minutes) or freezing milk altered the formol titration value slightly but did not change the sample sufficiently to affect activity of organisms used. All formol titration values used in the PDF calculations were obtained on raw, fresh skimmilk by the following modification of the formaldehyde titration for milk protein by Pyne *(4)*: Ten-ml. milk samples plus 10 ml. of water and 0.4 ml. of a saturated solution of aqueous potassium oxalate were dispensed into test tubes and allowed to stand for 2 minutes, after which an excess of quinhydrone was added and the solution was neutralized by titrating to pH 8.2 by means of a quinhydrone electrode; 2 ml. of 40% formaldehyde then was added to the neutralized milk. This was shaken and allowed to stand for approximately 1 minute and then titrated as before to the same endpoint. The number of milliliters of N/10 NaOH required to neutralize 10 ml. of milk was considered the formol titration value.

The total protein was determined on the same sample by using the micro-Kjeldahl procedure *(3)*. The protein degradation factor (PDF) against which acidities were plotted was derived as follows:

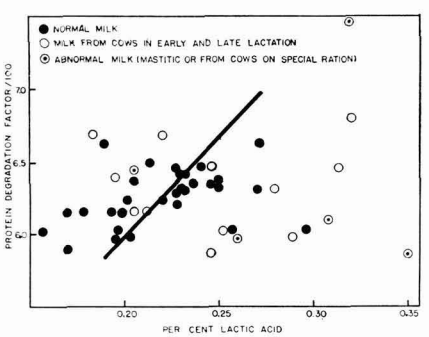
$$\frac{\text{Formol titration value}}{\text{Per cent total protein}} \times 1{,}000 = \text{PDF (protein degradation factor)}$$

The method recommended by Shahani and Sommers *(5)* for the determination of proteose peptone minus the nonprotein nitrogen was used to obtain peptide content of raw milks in the second series of trials. This method made it possible to correlate rate of growth with a defined fraction of the milk and eliminated the error due to presence of fractions such as nonprotein nitrogen.

Cultures employed for determining growth-promoting properties of the various milk samples included representative mixed strain commercial lactic streptococcus starters commonly used in this country for buttermilk, cottage cheese, and Cheddar cheese manufacture and single strains of *Streptococcus lactis* (SLE) and *S. cremoris* (144F and H-6). Activity of the cultures was maintained by daily transfer in reconstituted skimmilk and incubation at 21.5° C. Active 18-hour cultures were inoculated into experimental milks at the rate of 1%. The results are presented as per cent lactic acid calculated from the actual acidities produced by the organisms in a 5- or 8-hour incubation period at 30° C. At the end of the incubation period they were quickly frozen to prevent further growth and subsequently thawed, and titratable acidities were determined. The quantity of acid produced was used as an index of growth. This was correlated with the PDF or the peptide nitrogen, and results were plotted.

RESULTS AND DISCUSSION

Figure 1 shows the correlation between the PDF and the acidities produced by culture A, a representative, widely used mixed commercial preparation. The normal milk shows a growth response giving a positive correlation, whereas that considered abnormal exhibits unpredictable results. Figures 2 and 3 present correlation between peptide nitrogen in milk and acid production of two mixed commercial cultures (B and C) and Figures 4, 5, and 6 show results obtained



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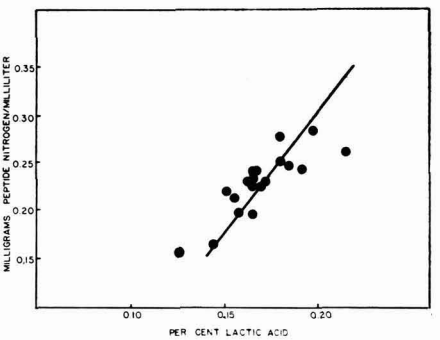

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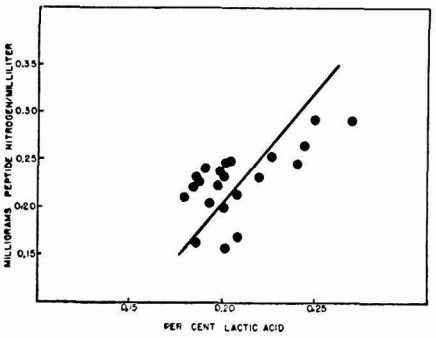
FIG. 1. Relation between protein degradation factor and acid produced by commercially available mixed strain lactic streptococcus starter culture A in individual cow samples of milk. Cultures were incubated at 30° C. for 8 hours. Regression equation is $y = -0.200 + 0.067x$ for normal milk where y is protein degradation factor and x is per cent lactic acid.

FIG. 2. Relation between peptide nitrogen and acid produced by commercially available mixed strain lactic streptococcus starter culture B in individual cow samples of milk. Cultures were incubated at 30° C. for 5 hours. Regression equation is $y = 0.0824 + 0.382x$ where y is peptide nitrogen in mg/ml and x is per cent lactic acid.

with three individual strains of lactic streptococci used in starters. Results indicate that normal milk from healthy cows varies in its growth-promoting property for the lactic streptococci according to its peptide content. There is a significant correlation for each of the cultures tested since the correlation coefficient r is significantly greater than 0.5 at the 1% level for peptides and greater than 0.4 at the 5% level for PDF.

Previous studies *(1)* indicated that not all commercial cultures or individual strains react similarly to peptide stimulation. However, all that were investigated in this study showed a positive response when cultured in an otherwise nutritionally complete medium. If, in the normal milk, the response to peptide was positive but no correlation existed, the cause usually could be traced to some individual strain nutritional requirement.

Additional data not shown in Figures 1 to 6 indicate that high protein content in milk does not necessarily favor more rapid acid production by starter organisms. In general, starter cultures and individual strains grew equally well



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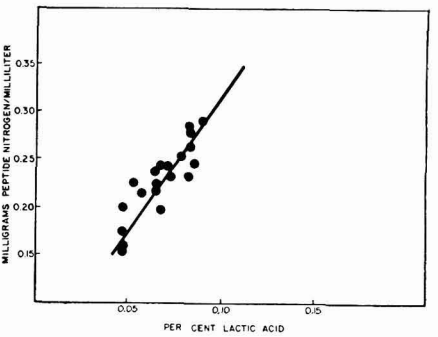

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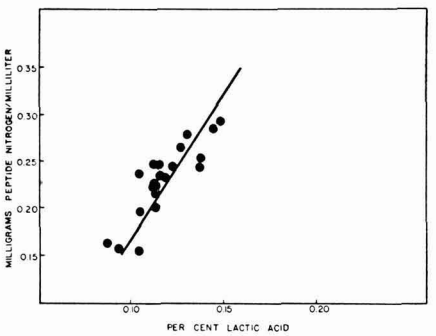
FIG. 3. Relation between peptide nitrogen and acid produced by commercially available mixed strain lactic streptococcus starter culture C in individual cow samples of milk. Cultures were incubated at 30° C. for 5 hours. Regression equation is $y = 0.112 + 0.424x$ where y is peptide nitrogen in mg/ml and x is per cent lactic acid.

FIG. 4. Relation between peptide nitrogen and acid produced by a single strain lactic streptococcus starter culture SLE in individual cow samples of milk. Cultures were incubated at 30° C. for 5 hours. Regression equation is $y = -0.00258 + 0.306x$ where y is peptide nitrogen in mg/ml and x is per cent lactic acid.

in high and low protein milk. The two animals on a ration deficient in certain essential nutrients (in this case, low carotene) produced milk which supported poor growth, although the protein as indicated by total nitrogen was normal. Other data also indicated no correlation between rate of acid production of lactic streptococci tested and content of nonprotein nitrogen in various samples of milk from individual cows.

The growth response of mixed commercial starter cultures to peptide variations was comparable to that of the individual strains. Some single cultures and one mixed commercial culture did not show a direct correlation, although the response to peptide nitrogen was positive in almost all instances. A possible explanation for this observation may be found in previous studies on the nutrition of the individual strains of lactic streptococci *(1)*, in which some cultures were shown to require certain purine and pyrimidine bases for optimum growth. It is possible that milk may vary in these constituents. Other studies *(2)* demonstrated that milk supplemented with adenine, guanine, or uracil accelerated the growth of the strains which previously had been shown to require one or more of the purine and pyrimidine bases. One fast-growing mixed-strain commercial starter did not respond as markedly as the others. It is possible that the strains used in the mixed culture in the present investigation initiate growth faster on less of the peptide fraction so that this does not become a limiting factor for growth in normal milk.

Some extreme variations in rate of growth in abnormal milks are shown in Figure 1. Milk from the late lactation period or from mastitic animals showed greater fluctuations in PDF than milk from healthy animals in the middle of the



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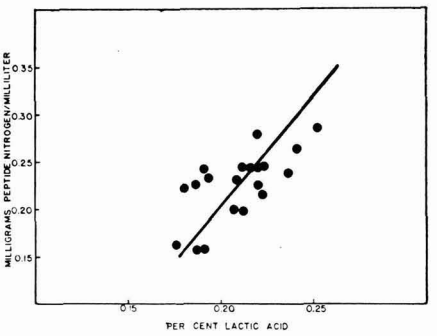

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FIG. 5. Relation between peptide nitrogen and acid produced by a single strain lactic streptococcus starter culture 144F in individual cow samples of milk. Cultures were incubated at 30° C. for 5 hours. Regression equation is $y = 0.0432 + 0.322x$ where y is peptide nitrogen in mg/ml and x is per cent lactic acid.

FIG. 6. Relation between peptide nitrogen and acid produced by a single strain lactic streptococcus starter culture H6 in individual cow samples of milk. Cultures were incubated at 30° C. for 5 hours. Regression equation is $y = 0.121 + 0.389x$ where y is peptide nitrogen in mg/ml and x is per cent lactic acid.

lactation period. Six animals with active mastitis were included in the original tests and, of these, the results of only three are shown in Figure 1. The milk from the others showed a higher PDF factor than the maximum limits of the scale in the figure, which indicates the possibility of some protein breakdown. The NPN was included in the total nitrogen determination used in calculating the PDF and may account for some of the observed discrepancies. Normal milk, which was characterized by small variations in NPN, exhibited good correlation between growth response and the peptide fraction or PDF.

There may be variations in other substances in milk besides the peptides which influence growth of lactic streptococci. Milk sampled early in the lactation period and milk from some cows producing a high volume provided slower growth of cultures than normal milk from the middle stages of lactation. This might be due to some inhibitory substance in milk from cows in early lactation. In the milk from abnormally high-producing animals there may actually be a suboptimal level of certain metabolites in the milk which are required for fast starter culture growth.

There was no significant difference between Jersey and Holstein milk in the amount of acid produced by starter cultures when the initial acidities were taken into consideration. Therefore, the amount of protein appears to have no appreciable influence on the rapidity of growth. The effect of increasing the milk protein was determined by the addition of various quantities of nonfat milk solids to fresh skimmilk. Amounts of powder so added ranged from 1 to 10% per 100 ml. of milk. The increase in initial growth rate by such a procedure was negligible. However, the addition of 0.1 to 1% of trypsin-hydrolyzed skimmilk

caused a marked increase in the initial growth rate of cultures in both Jersey and Holstein milk.

The variation in the peptide nitrogen appeared to be a characteristic of either the individual animal or the individual milking. There was no evidence that it was related to either of the breeds tested. The milk samples used were tested as soon as possible after being drawn to reduce natural enzymatic activity to a minimum. The presence of a natural proteolytic enzyme might conceivably increase the peptide content if the milk were stored. However, Shahani and Sommers *(5)* have shown such increases to be negligible over a 10-day period when stored at temperatures from 0° to 5° C. Further studies are planned to determine variation in peptide content of individual cow's milk from one milking to another and from herd to herd.

SUMMARY

Analysis of individual cow samples of milk indicated variation in peptide content. Rate of acid production by mixed strain commercial cultures and individual strains of lactic streptococci, in most instances, increased with increase in the peptide content of the milks from individual cows. Different cultures varied somewhat in their response to the peptide content of milk.

The peptide content appeared to exert greater effect on rate of growth of lactic streptococci than the protein content of milk. No significant difference was apparent between Jersey and Holstein milk from which the fat was removed. The correlation between rate of growth and peptide content was poor in a number of samples of milk from mastitic animals and from those in early or late stages of lactation.

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TOCOPHEROL CONTENT OF THE FAT OF DAIRY PRODUCTS AS AN INDEX OF ADULTERATION

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A rapid procedure for determining the tocopherol content of butterfat was developed in this laboratory *(2)* and has been successfully employed for 2 years in the detection of the addition of vegetable fats (except coconut) to butterfat. The application of the tocopherol procedure is based on the fact that the tocopherol content of butterfat is low (10 to 50 p.p.m.) *(1)* whereas that of most vegetable fats is considerably higher (300 to 1600 p.p.m.) *(2)*. In most cases, therefore, the addition of vegetable fats to butter will result in a significant increase in the tocopherol content of the adulterated butterfat. Adulteration with lard and tallow cannot be detected by this procedure.

It appeared of interest to extend this tocopherol procedure to the detection of vegetable fats in dairy products other than butter. It has been found that by using a modification of the butter procedure *(2)*, the adulteration of evaporated milk, condensed milk, whole milk powder, ice cream, and cheese with vegetable fats can be detected. In all products the isolated fat is analyzed as for butterfat, and only the method of isolating the fat has been modified. A variation of the Sager and Sanders *(3)* detergent procedure has been found satisfactory for liberating the fat from evaporated milk, condensed milk, whole milk powder, ice cream, and cheese. The details of these procedures are given in the following sections.

EXPERIMENTAL PROCEDURE

Reagents:

Detergent solution. Dissolve 35 g. sodium tetraphosphate[1] in approximately 200 ml. of warm water, add 15 g. of Triton x-100[2], dilute to 1,000 ml. with water and mix. Store this reagent in a refrigerator and use within 48 hours. If stored longer, low tocopherol values may be obtained for the isolated fat.

Methyl alcohol. Prepare 50% methyl alcohol by volume.

Isolation of fat from evaporated or condensed milk. Place 125 g. of sample in a 500-ml. Erlenmeyer flask, add 175 ml. of detergent solution, and mix. Place the flask in a boiling water bath, making certain that the water level in the bath is above the level of the liquid in the flask. Shake the flask several times during a period of 10 to 15 minutes or until the fat layer has completely separated. Allow the flask to remain in the boiling water bath for 5 minutes more, without

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[1] Sold as "Quadrafos" by the Rumford Division, Heyden Chemical Corp., Rumford 16, Rhode Island.

[2] Obtainable from Rohm & Haas Co., Philadelphia, Pa.

remixing. Remove the flask from the bath, add 50% methyl alcohol down the side until the fat layer reaches the bottom of the neck, and then add water until the flask is full. Immerse the flask to within 0.5 in. of the top of the neck in a water bath at 55° C. When no more fat rises, usually after 15 minutes, remove the flasks from the bath. Skim off any scum on the surface of the fat, draw off the fat with a warm pipette, filter it, and store it in a refrigerator. Proceed as directed for the determination of tocopherol content of butterfat *(2)*.

Isolation of fat from whole milk powder. Mix 40 g. of whole milk powder with 85 ml. of water. Treat this mixture as the evaporated milk.

Isolation of fat from ice cream. Allow the ice cream to melt. If fruit or nuts are present, filter the ice cream through a pad of glass wool and discard the fruit or nuts. Treat 100 g. of filtered ice cream as the evaporated milk.

Isolation of fat from cheese. Place 40 g. cheese in a Waring blendor, add 85 ml. of water and blend thoroughly. Treat this fluid product as the evaporated milk.

DISCUSSION

The Sager and Sanders *(3)* detergent reagent was employed at half the original concentration because it was found that emulsions form during the sulfuric acid extraction step if the full-strength reagent is employed.

No interferences have been encountered in the analysis of evaporated milk, condensed milk, or whole milk powder. In ice cream, however, the presence of flavorings, nuts, fruit, and confections might be expected to influence the apparent tocopherol content of the isolated fat, and it has been found that ice cream containing nuts yields unusually high tocopherol values. These results are caused by the high tocopherol content of the oil of most nuts (450 p.p.m. was found in walnut oil). Apparently, some of the nut oil is liberated from the surface of chipped nuts during the isolation of the fat and raises the tocopherol content of the extracted fat. This error can be minimized by filtering off the nuts from the melted ice cream before isolating the fat. The effect of nuts in ice cream upon the tocopherol content of the isolated fat is illustrated in Table 1.

TABLE 1

Effect of nuts in ice cream upon the tocopherol content of the isolated fat

	Tocopherol content of the isolated fat in p.p.m.	
Type of ice cream	Nuts present	Nuts filtered off
Butter pecan	50	37
Maple walnut	47	35

Many kinds of ice cream have been analyzed to determine whether other ice cream ingredients influence the apparent tocopherol content of the isolated fat. Since chocolate, maple, strawberry, and vanilla ice creams are widely sold, the majority of analyses were conducted on these flavors. The summary of 80 analyses is given in Table 2.

TABLE 2

Tocopherol content of the fat isolated from differently flavored ice creams

Flavor	No. samples analyzed	Av. tocopherol content of fat	Tocopherol range
		(p.p.m.)	*(p.p.m.)*
Banana	1	31	——
Butter pecan (filtered)	1	37	——
Butterscotch	2	32	29-34
Burgundy cherry	1	37	——
Cherry	2	44	43-44
Chocolate	11	45	38-58
Chocolate (3-flavored brick)	14	52	43-71
Maple	5	40	34-47
Maple walnut (filtered)	1	35	——
Orange	1	36	——
Orange sherbet	2	32	32
Pineapple	1	41	——
Strawberry	14	38	22-53
Vanilla	24	39	29-49
Butterfat at same time of year	48	42	35-49

These data indicate that the tocopherol content of maple, vanilla, and strawberry ice cream is in the same range as that of butterfat produced at the same time of year *(1)*. The fat isolated from chocolate ice cream, however, exhibited a significantly higher tocopherol content. This increase is attributed to the presence of the cocoa fat, which was found to have a tocopherol content of 150 to 200 p.p.m. The higher tocopherol content for the chocolate portion of three-flavored ice cream bricks was due to the higher chocolate content of this product.

The original tocopherol procedure for the analysis of the fat of butter was designed to avoid interferences due to carotenoids, Oil Yellow AB (F.D. & C. Yellow No. 3) and Oil Yellow OB (F.D. & C. Yellow No. 4). These are the only interferences that have been encountered in butter. In Cheddar cheese, annatto also may be present, but fortunately this vegetable dye causes no error in the tocopherol procedure. Of a large number of Cheddar cheeses examined, all yielded fat with normal tocopherol contents, indicating that the cheese making and aging processes have no significant effect on the tocopherol content.

Recently, a new type of cheese spread has appeared on the Canadian market which when analyzed yields abnormally high tocopherol contents. This peculiar behavior has been traced to the presence of other permitted oil-soluble color in the cheese, such as Orange SS (F.D. & C. Orange No. 2) or Oil Red XO (F.D. & C. Red No. 32). If such colors are present, they are not completely removed from the fat by the sulfuric acid extraction step *(2)* or by three or four additional extractions. Consequently, these unextracted colors contribute to the apparent tocopherol contents with the result that higher values are obtained.

In order to avoid erroneous conclusions from the use of the tocopherol procedure, its use should be restricted to the analysis of the fat from uncolored cheeses or to cheeses which contain only yellow AB (F.D. & C. Yellow No. 3), yellow OB (F.D. & C. Yellow No. 4), or annatto.

Application of the tocopherol procedure for the detection of adulteration. In addition to butterfat *(1)*, the tocopherol procedure has been employed success-

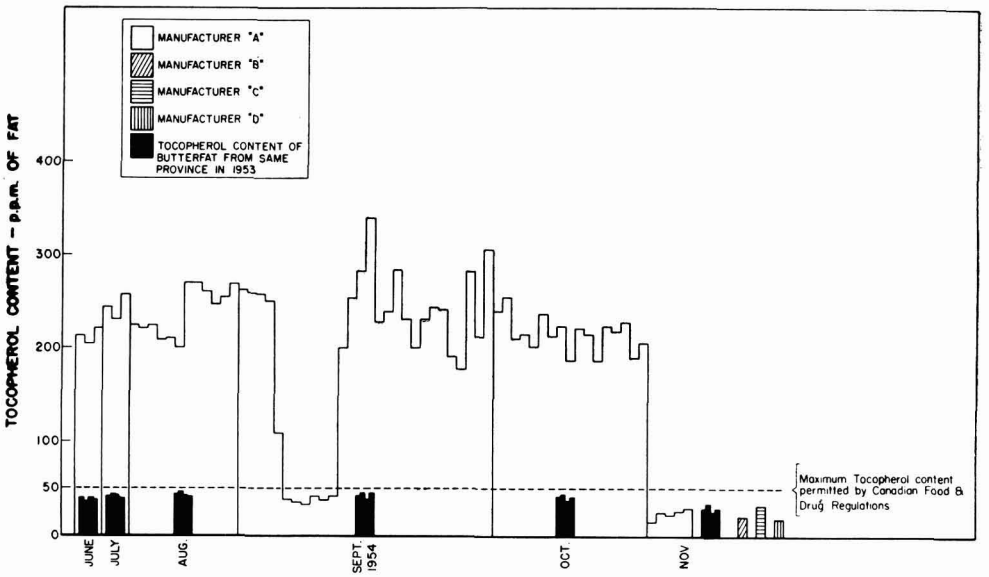

FIG. 1. Tocopherol content of the fat of evaporated milks.

fully to detect the adulteration of evaporated milk, condensed milk, and whole milk powder. Typical results on evaporated milk are illustrated in Figure 1.

These data indicate a wide variation in tocopherol content for the fat from evaporated milk produced by manufacturer A. A few fat samples in September exhibited normal tocopherol contents, when, according to testimony given in court, this manufacturer had run out of the adulterant fat. In addition, the samples produced in November, immediately after the seizure of the adulterated stocks, were found to have a normal tocopherol content. The average tocopherol content for the adulterated fat from evaporated milk produced by manufacturer A was 231 p.p.m., as compared to 31 p.p.m. for the samples considered to be unadulterated. These latter values are in close agreement with those from genuine butterfats produced at the same time of year, as well as with the fat from evaporated milk produced by manufacturers B, C, and D.

A sample of the fat obtained from the adulterant employed by manufacturer A was found to have a tocopherol content of 620 p.p.m., a Reichert-Meissl value of 2.5, and a Polenske value of 0.8. The adulterant consisted of vegetable fat homogenized with milk to produce a cream-like product with approximately 30% fat. It appears from the foregoing analytical data that approximately 30 to 35% of the normal milk fat in the evaporated milk had been replaced with the adulterant fat.

Similar data relating to the tocopherol content of adulterated whole milk powder and condensed milk are given in Figure 2.

These data again demonstrate the excessive tocopherol content of the fat from whole milk powder and condensed milk produced by manufacturer A. On the

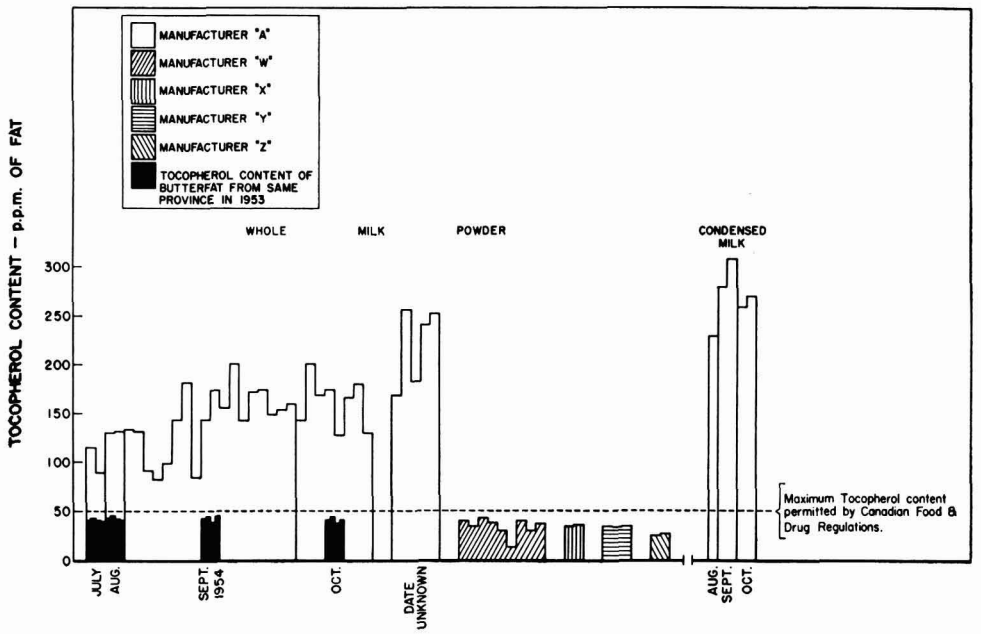

FIG. 2. Tocopherol content of the fat of whole milk powders and condensed milks.

other hand, the tocopherol content of the fat of whole milk powders produced by manufacturers W, X, Y, and Z was normal and of the same order as for genuine butterfat.

The increasing tocopherol content of the fat of whole milk powder produced by manufacturer A during the period July to October, 1954, is due to the increased percentage of adulterant employed. This observation is confirmed by the progressive reduction in Reichert-Meissl value of the fat as shown in Table 3. Available data would indicate that the fat of whole milk powder produced by manufacturer A contained approximately 12% adulterant in July and 25% in October, 1954. The fat from condensed milk produced by manufacturer A appeared to contain approximately 40% of adulterant fat, as shown in Table 4.

TABLE 3

Tocopherol content, Reichert-Meissl value, and Polenske value for fats isolated from adulterated whole milk powders produced by manufacturer A

Month of manufacture	Tocopherol content	Reichert-Meissl value	Polenske value
	(p.p.m.)		
August	131	21.5	1.4
September	148	19.5	1.4
September	153	19.0	1.4
September	159	19.5	1.2
October	180	18.9	1.3

TABLE 4

Tocopherol content, Reichert-Meissl value, and Polenske value for fat isolated from adulterated evaporated milk produced by manufacturer A

Tocopherol content	Reichert-Meissl value	Polenske value
(p.p.m.)		
201	19.3	1.1
244	17.6	1.1
242	16.2	1.1
224	15.0	1.0
239	14.5	1.1

The data in Tables 3 and 4 indicate that the level of foreign fat present was such that the Reichert-Meissl procedure could have been used to detect the adulteration. However, the tocopherol procedure was employed routinely, since its use results in a considerable saving of time.

The Food and Drug laboratories analyzed 6,270 commercial butterfat samples during 1954—with the results given in Figure 3. Only one butterfat sample contained a tocopherol value in excess of 50 p.p.m. (58 p.p.m.). The average

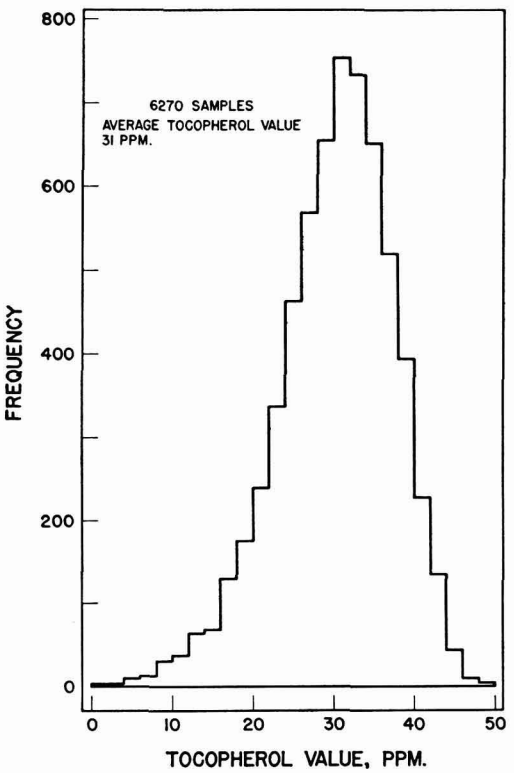

FIG. 3. Tocopherol values for 6,270 commercial butterfat samples.

tocopherol value was 31 p.p.m. and the range, excepting the aforementioned sample, was 2 to 50 p.p.m., which is in excellent agreement with previous data obtained on genuine butterfat samples *(1)*. It is believed that the analysis of butterfats from storage might have accounted for the few samples with tocopherol values of 2 to 9 p.p.m.

SUMMARY

The tocopherol procedure, originally developed to detect the adulteration of the fat of butter, has been extended to the analysis of the fat of evaporated milk, condensed milk, whole milk powder, ice cream, and cheese. This procedure will not detect lard, tallow, or coconut oil.

The tocopherol content of the fat from evaporated milk, condensed milk, whole milk powder, and Cheddar cheese is very close to that of butterfat produced at the same time of year.

Nuts and fruit, if present in ice cream, must be filtered off before isolating the fat or excessively high tocopherol contents may be found. The tocopherol procedure cannot be applied to chocolate ice cream owing to the error caused by the tocopherol content of the chocolate fat.

This procedure cannot be applied to processed cheeses that contain coloring materials other than annatto, Oil Yellow AB (F.D. & C. Yellow No. 3), or Oil Yellow OB (F.D. & C. Yellow No. 4). Other colors, if present, may cause abnormally high results since they are not completely extracted from the fat by the sulfuric acid extraction step.

Examples are given to demonstrate the effectiveness of the tocopherol procedure for detecting the adulteration of the fat of evaporated milk, condensed milk, and whole milk powder.

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THE PHOSPHATASE ACTIVITY OF CHOCOLATE MILK

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Chocolate milk generally is made by blending raw white milk with prepared sirups or powders and pasteurizing the mixture. Certain components of chocolate milk, not found in white milk, may influence phosphatase activity and color values so markedly as to make some "pasteurized" samples of chocolate milk unacceptable to health officials. These components, such as sugar and artificial flavors, have been shown by Hahn and Tracy *(2)* and Caulfield and Martin *(1)* to alter significantly the phosphatase values of ice cream.

For a variety of reasons a wide range of pasteurization temperatures has been recommended for chocolate milk by chocolate manufacturers. Temperatures of 145° to 185° F. for 30 minutes are suggested for vat pasteurization but for HTST, in certain instances, the same minimum treatment given to white milk is advocated. A search of the scientific and regulatory literature has failed to disclose the minimum heat treatment considered necessary to obtain negative phosphatase values for chocolate milk. As a result, the present study was projected to observe phosphatase activity in heated chocolate milks and to determine, within practical limits, the heat treatment required for inactivation of phosphatase in chocolate milk.

METHODS

The initial phase dealt with actual commercial pasteurization of chocolate milks at various temperatures. A Cherry-Burrell Super-plate HTST pasteurizer, capacity 3,600 lb/hour, and an R. G. Wright 30-gal. vat pasteurizer were used. Both pasteurizers were equipped with properly calibrated indicating and recording thermometers.

Mixed raw whole milk, averaging about 3.5% B.F., from the College herd and local producers was made into chocolate milk with sirup in proportions recommended by the manufacturer. For each experimental HTST run, 2,000 lb. of mixed raw milk was required. Of this quantity, 350 lb. was pasteurized as such at either 161.5° F. or 162° F. for 15.5 seconds, and the remainder was made into chocolate milk, which immediately followed through the HTST pasteurizer. Successive portions of this chocolate milk were pasteurized at various levels from 161.5° to 171.5° F. for 15.5 seconds. Immediately after being cooled to 50° F. or lower in the cooling section, approximately 1-l. samples were drawn from the pasteurizer for testing.

Vat pasteurization was accomplished by heating 30-gal. quantities of raw chocolate milk to 150° F. ± 0.5°. Small samples were withdrawn at appropriate time intervals, followed by quick cooling to 40° F. in ice water.

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[1] Now serving with the United States Army.

Samples of chocolate milk were laboratory pasteurized by means of a series of water baths, thermostatically controlled, similar to the immersion method of Holland and Dahlberg *(3)*. Milk samples in 25-ml. sterile test tubes and 125-ml. flasks were completely immersed in hot water for the appropriate times and temperatures, followed by rapid cooling to 40° F.

Analysis of phosphatase activity for all samples was by the Cornell 24-hour method *(4)*. One slight change was made in the method for chocolate milk in that final blue color was developed at room temperature (20-24° C.) for 15 minutes instead of at 37° C. Colors were read visually and also by using a Bausch and Lomb colorimeter, 660 mμ, on butyl alcohol extractions. In addition to the Cornell phosphatase method, a number of samples were examined by using the ether extraction modification of the Kay-Graham phosphatase procedure *(5)*.

RESULTS

Phosphatase activity of commercial chocolate milks after HTST pasteurization. Phenol values (representing phosphatase activity) on two large lots of chocolate milk made from different sirups and pasteurized at varying temperatures are presented in Table 1. Raw chocolate milks with a sugar content, other than lactose, of about 5 to 6% displayed significantly higher phenol values than white milk when both were subjected to the same heat treatment. This phosphatase activity in chocolate milk at the 161.5°-162.2° F. level, attributable in some degree to the enzyme sparing effect of sugar, represents a raw milk equiva-

TABLE 1

The phosphatase activity of commercial chocolate milk after HTST pasteurization

Milk	Temp. with 15.5 sec. holding time	Phosphatase activity by Cornell method[a]		Phosphatase activity by ether extraction modification of Kay-Graham	
		Normal sugar[b]	3% added sucrose	Normal sugar[b]	3% added sucrose
	(° F.)	(γ phenol per 0.5 ml.)		(mg. phenol per 0.5 ml.)	
		Chocolate milk made with sirup A			
Plain white	162.2	0.0	—	0.000	—
Choc.	162.2	38.4	45.0	0.019	0.022
Choc.	163.7	25.4	30.0	0.014	0.014
Choc.	166.0	11.5	10.1	0.010	0.011
Choc.	169.0	1.2	5.5	0.012	0.005
Choc.	171.5	0.0	0.0	0.000	0.001
		Chocolate milk made with sirup B			
Plain white	161.5	1.2	—	0.000	—
Choc.	162.0	26.3	26.3	0.026	0.035
Choc.	163.5	20.8	22.6	0.020	—
Choc.	165.5	10.5	13.5	0.018	0.031
Choc.	167.5	6.7	8.8	0.015	0.022
Choc.	169.5	3.8	6.2	0.008	0.011
Choc.	171.5	2.4	4.6	0.003	0.006

[a] Any value above 5.0γ/0.5 ml. is considered either underpasteurized or raw, when Cornell phosphatase method is used.

[b] Normal sugar content of chocolate milk from sirup A—5.7% (44% sucrose, 55% invert). Normal sugar content of chocolate milk from sirup B—6.8% (78% sucrose, 22% invert).

lent on the order of 0.3-0.5%. A temperature of at least 169° F. for 15.5 seconds was necessary before phenol values of normal sugar chocolate milk became comparable to those of white milk properly pasteurized at 161.5° or 162° F. for 15.5 seconds (Table 1).

To observe the effects of excess concentration of sucrose in chocolate milk, a duplicate lot of raw chocolate milk with 3% added sucrose was pasteurized in the HTST apparatus at the same temperatures as normal sugar chocolate milk. As observed in Table 1, excess sugar has a tendency to increase the protective action over the phosphatase enzyme against heat, although in general the over-all effect, at these levels, was not extremely pronounced. Phenol values comparable to properly pasteurized white whole milk were attained with excess sugar chocolate milks at temperatures of 170° to 171.5° F. for 15.5 seconds. In practice such high levels of sugar, 8-9% over-all, other than lactose would appear undesirable because of the resulting intense sweetness.

Measurement of phosphatase activity by the ether extraction modification of the Kay-Graham method (Table 1) was introduced to observe differences by means of yet another method based on different principles and reagents. This method, used successfully on ripened cheese, has not previously been applied to other dairy products. In chocolate milk the critical value for determining underpasteurization is undetermined, although it would appear from values in Table 1 that 0.005 or 0.01 mg. phenol per 0.5 ml. might be satisfactory. Data obtained here with this secondary method reflect the general trend obtained by the Cornell phosphatase method.

Vat pasteurization of chocolate milk. Heating of raw chocolate milk to 150° F. in a vat pasteurizer resulted in phenol values below 5.0γ/0.5 ml. at the end of 18 minutes for chocolate milk A and at the end of 22 minutes for chocolate milk B

TABLE 2

The phosphatase activity of commercial chocolate milk after vat pasteurization

Holding time at 150° F.	Phosphatase activity by Cornell method[a]	
	Normal composition choc. milk made with sirup A[b]	Normal composition choc. milk made with sirup B[b]
(min.)	*(γ phenol per 0.5 ml.)*	
8	21.6	19.3
10	15.6	17.6
12	11.7	10.7
14	10.7	8.6
16	10.7	7.7
18	3.6	7.7
20	3.9	7.7
22	3.8	3.9
24	0.0	0.0
26	0.0	0.0
30	0.0	0.0

[a] In the Cornell phosphatase method, any milk having value greater than 5.0γ/0.5 ml. is considered underpasteurized or raw.

[b] Normal sugar content of chocolate milk from sirup A—5.7% (44% is sucrose and 55% invert).

Normal sugar content of chocolate milk from sirup B—6.8% (78% is sucrose and 22% invert).

(Table 2). Enzyme activity was reduced to zero with both lots of chocolate milk when a temperature of 150° F. was sustained for 24 minutes.

Pasteurization of chocolate milks in the laboratory. Twenty-three different chocolate sirups and powders from nine independent sources were mixed with raw whole milk in proportions recommended by their manufacturers. These individual chocolate milks were laboratory pasteurized at various times and temperatures to note the degree of variation in phosphatase values.

Laboratory pasteurization at 170° F. for 15 seconds and 150° F. for 25 minutes (Table 3) reduced phenol values of most chocolate milks to a point comparable to heated, white whole milk. Even with the exceptions that exist, the highest phenol value was only about 10γ/0.5 ml. These exceptions in actual practice represent less than 0.1% raw milk equivalent by the Cornell method. Similar temperatures used in conjunction with commercial equipment would more than likely lower these values even further *(1)*.

The use of pasteurization treatments ordinarily associated with white whole milk, 160° F. for 15 seconds or 145° F. for 30 minutes, showed very marked phosphatase activity in chocolate milk samples (Table 3).

TABLE 3

Phosphatase activities of chocolate milks made from commercial chocolate sirups and powders[a] and laboratory pasteurized at different temperatures

Choc. milk made with commercial sirup or powder	Chocolate milk laboratory pasteurized at				
	170° — 15 sec.	150° — 25 min.	160° — 15 sec.	145° — 30 min.	Controls[b]
(No.)	Phosphatase values by Cornell method *(γ phenol/0.5 g.)*				
1	2.5	4.4	73.1	26.0	16.6
2	1.0	7.0	>144.0	45.0	14.4
3	7.2	10.2	92.0	73.0	12.0
4	10.8	1.8	>144.0	56.0	3.6
5	2.4	3.2	66.0	51.0	3.6
6	8.4	4.8	>144.0	>144.0	5.0
7	7.2	4.0	66.0	43.0	4.8
8	4.8	4.4	>144.0	66.0	9.6
9	3.6	4.0	123.0	32.0	9.6
10	0.0	4.8	>144.0	>144.0	9.6
11	3.6	0.6	>144.0	73.0	4.8
12	0.6	9.6	>144.0	92.0	1.8
13	2.2	3.2	92.0	34.0	4.2
14	1.2	0.0	123.0	74.0	1.8
15	4.8	6.4	136.0	79.0	2.4
16	3.6	3.0	>144.0	123.0	14.4
17	4.2	1.8	92.0	14.0	13.2
18	0.0	3.4	>144.0	43.0	14.4
19	0.4	1.3	56.0	34.0	14.4
20	0.3	4.2	>144.0	32.0	5.0
21	2.4	4.8	>144.0	30.0	6.0
22	0.0	6.0	>144.0	36.0	3.6
23	2.8	3.7	62.0	23.0	3.0
White milk	1.8	2.4	17.0	0.0	0.0

[a] Range of components of chocolate sirup or powders—chocolate, cocoa, sucrose and invert sugar, corn sirup, dextrose, salt, carrageen, corn starch, vanillin, nonfat milk solids, vitamin D, irradiated yeast, artificial flavor.

[b] Controls obtained by heating chocolate milk to 190° F. for 2 minutes and incubating with buffer substrate. These values have been subtracted to obtain the tabulated data.

DISCUSSION

It is evident that the composition of the average chocolate milk is of such nature as to afford significant protection to alkaline phosphatase. The question of whether the same degree of protection is given to pathogenic bacteria possibly existing in chocolate milk has received prior consideration *(7)*, but this phase was not within the scope of the present investigation. The data obtained in this study point to 170° F. for 15.5 seconds or 150° F. for 30 minutes as being practical minimum values for the pasteurization of chocolate milk.

A point should be made concerning phosphatase control values of chocolate milk, which are more variable and higher than found with white milk (Table 3). These high control values result from such interfering compounds in chocolate milk as artificial flavors. They appear relatively uniform in value for the products of the same manufacturer but vary rather widely between the various manufacturers' products. This is easily understood, as sirups or powders formulated by one manufacturer may contain the same specific flavor compound at the same levels for all of his products. Such variability between the different manufacturers' products emphasizes the need for making control checks on all individual samples of chocolate milk when testing for phosphatase.

The protective influence of sugar upon phosphatase during heating, previously observed in ice cream, has been demonstrated here in chocolate milk. Not shown previously is the influence of sugar on the mechanics of the phosphatase test. It was observed that the final blue color developed in chocolate milk samples behaved very differently from blue color resulting in white milk at 37° C. Though blue color eventually will fade even for white milk in phosphatase tests, its rate of fading is relatively slow, requiring many hours. In chocolate milk samples when blue color developed at 37° C., the peak of color intensity was reached rapidly. At about 5-6 minutes the color began to fade, progressing from the bottom of the test tube upward. Generally the contents of the test tube became completely colorless in a few minutes. Extraction with butyl alcohol when peak color was developed arrested color fading completely. This effect appeared directly attributable to the sugar in the chocolate milks, as the samples with higher sugar concentrations tended to increase in rate of color disappearance. The effect was also noted with the Sanders-Sager phosphatase test but was not as pronounced. It was not observed with the ether extraction method of Kay-Graham as the ether does not extract sugar and testing is done on a sugar-free solution. Concurrently, a similar type of reaction was observed in phosphatase testing of ice cream by Pyne *(6)*, who considered that not only were sugars responsible but the rate of fading was affected by the type of sugar in the mixture.

The present authors have found that, for chocolate milk, ice cream, or any dairy product containing added sugar, blue color development proceeds normally if allowed to develop at room temperature (22-25° C.), instead of 37° C., for 15 minutes. At room temperature, maximum blue color developed in 5 to 8 minutes with Cornell reagents, and no problems resulted in color fading, as the blue color was stable for at least 30 minutes.

SUMMARY

Phosphatase activity of chocolate milk was significantly higher than the activity in its white whole milk counterpart when both milks were pasteurized at minimum temperatures required for the latter. Sucrose was shown to contribute to the protection of the phosphatase enzyme during heating of chocolate milk.

Heating of chocolate milk under commercial and laboratory conditions indicated that the practical minimum times and temperatures required for the proper pasteurization of chocolate milk, using white whole milk as a comparison, should be 170° F. for 15.5 seconds or 150° F. for 30 minutes.

The importance of proper controls and the interesting effects of sugar on color development in the phosphatase test used are briefly discussed.

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ANTIBIOTICS AS GROWTH STIMULANTS FOR DAIRY CATTLE: A REVIEW[1]

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The feeding of antibiotics to farm animals has brought about a new era in livestock production. The beneficial effects of adding small quantities of various antibiotics to poultry and swine rations have been well established. The merits of feeding antibiotics to ruminants are still of considerable interest to rumen nutritionists, physiologists, and bacteriologists.

Most of the present interest in antibiotics as supplements to animal feeds was initiated in early 1950 and stemmed from the production of vitamin B_{12} and the feeding of crude fermentation products as sources of this vitamin *(52)*, although Harned *et al. (36)* reported in 1948 that Duomycin (aureomycin) did improve the growth rate of chicks. These products were commonly named animal protein factor (A.P.F.) concentrates, and one of the products in general use was the fermentation product of *Streptomyces aureofaciens*.

Stokstad *et al. (117)* reported in early 1949 that a fermentation product of *Streptomyces aureofaciens* produced a growth response in chicks which was greater than the growth response obtained from vitamin B_{12}. Early in 1950, Stokstad and Jukes *(116)* observed that crystalline aureomycin produced similar results. This observation was confirmed by others working with chicks and pigs *(16, 26, 43, 51, 90, 97, 126)*. Luecke *et al. (76)* also reported that streptomycin improved the growth rate of young pigs. Since these early reports it has been well established that various antibiotics, namely, aureomycin (chlortetracycline), terramycin (oxytetracycline), penicillin, bacitracin, and streptomycin (dihydrostreptomycin), stimulate the growth rate of monogastric farm animals, such as the chick, turkey, and pig.

The effect of antibiotics on ruminants might be expected to be different from that on simple-stomach animals since ruminants depend basically on bacterial synthesis for proper nutrition. The results of antibiotic feeding to ruminants at first appeared to be contradictory. Workers at the Louisiana *(99)*, Kansas *(3)*, and Cornell *(73)* stations found that aureomycin promoted growth and possibly helped to eliminate scours in young dairy calves. On the other hand, Bell *et al. (10)* reported adverse effects from the feeding of aureomycin to beef steers, and Colby *et al. (24)* found that aureomycin depressed feed consumption and reduced the growth of lambs. Colby *et al. (23)* also found that these adverse effects could not be overcome by the feeding of various members of the vitamin B-complex group in addition to aureomycin. These workers *(22)* had found earlier that aureomycin lowered the blood level of vitamin B_{12} in lambs, thus suggesting an interference with rumen vitamin synthesis. Penicillin and strep-

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tomycin were also fed, but these did not cause the severe reactions of aureomycin, although no beneficial effects were observed. Bell *et al. (11)* found that the feeding of 600 mg. daily of crystalline aureomycin to 620-lb. Hereford steers produced a marked anorexia and diarrhea within 48-72 hours. The level was reduced to 200 mg. daily and was fed without serious reactions to all but two steers, which still reacted to the treatment. The digestibility of dry matter was reduced 60% and the digestibility of crude fiber 45% by aureomycin. Aureomycin also increased the blood levels of urea nitrogen significantly. It was proposed by these workers that aureomycin had a depressing effect on the cellulolytic microorganisms in the gastro-intestinal tract.

With these conflicting reports it became apparent that there was a major difference in the reaction among ruminants to antibiotic treatment. In these early studies the major difference was apparently the result of the varying ages of the experimental animals. Bell *et al. (10)* used about 600-lb. yearling beef steers, and Colby *et al. (22)* in one trial used fattening lambs and in another trial used lambs ranging in ages from 4 to 12 weeks and in weight from 7.5 to 68.0 lb. In the latter trial an animal protein factor concentrate containing aureomycin was used rather than crystalline aureomycin. In contrast to these studies, Bartley *et al. (3)*, Loosli and Wallace *(73)*, and Rusoff *(99)*, all working with young dairy calves, observed beneficial effects from the feeding of aureomycin. Since the young calf is essentially a monogastric animal and not a ruminant, it was speculated that these conflicting results depended on whether or not the microflora of the rumen was developed at the time of antibiotic administration.

In this review an attempt will be made to adequately consider all antibiotics in respect to all classes of dairy cattle to the extent of published data. Some unpublished data have been included since their inclusion helped to complete the discussion.

The subject of antibiotics as components of dairy cattle rations will be discussed principally according to class of dairy cattle, namely young calves, growing animals, and mature animals, such as lactating cows, steers, and dairy bulls. Throughout this review the trademark names of the antibiotics will be used rather than the chemical name of these products since the trademark name is the one in general use.

YOUNG CALVES

The principal interest concerning antibiotics for young dairy calves has been their effect on *(a)* growth, *(b)* calf scours, *(c)* feed consumption and feed efficiency, and *(d)* metabolism and the possible mode of action of antibiotics. These various points of interest will be discussed separately insofar as possible, but when it seems advantageous they will be combined with each other.

A. *Growth.*

1. *Various antibiotics.* The feeding of antibiotics to farm animals has been a continuation of the interest in the role of vitamin B_{12} in their nutrition since animal protein factor (A.P.F.) concentrates were used as sources of vitamin

B_{12} rather than the crystalline vitamin. Because of this some early workers fed various A.P.F. concentrates which contained factors other than vitamin B_{12}, and these later were found to be antibiotics. Unfortunately, in a few reports the source of the A.P.F. was not given; therefore, the antibiotic which it might have contained is not known.

a. *Aureomycin.* Rusoff and Haq *(108)* in June, 1950, questioned the feeding value of A.P.F. concentrates in the rations of young dairy calves. The concentrate (Merck & Co., No. 3) used by these workers was added to an all-plant calf starter at a rate sufficient to supply 10 mg. of vitamin B_{12} per ton. No advantages could be found by including this concentrate in the ration of the calves. The antibiotic content of the A.P.F. was not stated. Williams and Knodt *(128, 129)* reported similar findings, using two A.P.F. concentrates, one supplied by Merck and Co., and the other by Lederle Laboratories. These supplements were added to milk replacement rations containing 60% milk products. The antibiotic content of these products was not indicated.

In November, 1950, Bartley *et al. (3)* at the Kansas station reported that an A.P.F. concentrate containing aureomycin stimulated the growth rate of dairy calves from birth to 42 days of age. These calves were fed the equivalent of about 15 mg. daily of aureomycin per 100 lb. of body weight. The growth rate of the aureomycin-fed calves was the same as that of Ragsdale standards *(95)*, but the growth of the control calves was considerably less. The control calves gained 18.0 lb. in 42 days and the A.P.F.-fed calves gained 30.8 lb., or 71.1% faster. The incidence of calf scours was much lower in the aureomycin-fed calves, and these calves also showed more thriftiness and an improved over-all condition. These data caused the authors to conclude that aureomycin enhanced the growth of the calves by preventing scours. Certainly it would appear that the disease problem in this study was great and that aureomycin was beneficial under these conditions. Rusoff *(99, 100)* reported also in November, 1950, that an A.P.F. concentrate containing aureomycin improved the growth rate of calves. However, the Louisiana worker was using dairy calves which were 14 weeks of age, and some of these calves had previously been fed an A.P.F. concentrate containing vitamin B_{12}. This trial was conducted from the time the calves were 14 weeks of age until they were 34 weeks of age. Aureomycin increased the growth rate of the calves over the control calves 60, 36, and 30% after 2, 4, and 6 weeks of the experiment. After 8 weeks of aureomycin feeding, the response had decreased to 8% and after 20 weeks there was no difference between the two groups of calves. This work indicated that aureomycin did not cause anorexia or diarrhea and apparently had no effect on rumen function of ruminating calves. These results were in contrast to the results of Bell *et al. (10)* and Colby *et al. (24)*, who indicated that the feeding of antibiotics produced adverse effects.

Also in 1950, Loosli and Wallace *(73)* reported a growth response in young calves both from an A.P.F. concentrate containing aureomycin and from crystalline aureomycin. Both of these supplements were fed in a milk substitute at the rate of 10.0 g. per ton of the crystalline antibiotic. The control calves gained 0.92 lb. and the antibiotic-fed calves 1.11 lb. daily for the first 8 weeks. As was

reported by the Kansas workers *(3)*, a significant reduction in calf scours was observed from the feeding of aureomycin. This work is of special interest since the A.P.F. concentrate and crystalline aureomycin produced similar growth responses. Seemingly, the growth response from the A.P.F. supplement was due to its aureomycin content rather than to the vitamin B_{12} which it contained. These workers postulated that the reason Rusoff and Haq *(108)* and Williams and Knodt *(128, 129)* had not observed a significant growth response from the A.P.F. concentrates used by them was that a small number of calves was involved in their studies and the product was not fed in the milk; therefore, it was not consumed in large enough quantities early in life. The Cornell workers *(73)* employed 27 pairs of calves in their study. Although this possibly could have been true, the type of A.P.F. concentrate fed could also have been a factor. In both studies no indication of the antibiotic content of the supplements was given. In the report of Williams and Knodt it will have to be assumed that consumption of the A.P.F. supplement did occur, since it was fed as a part of the milk replacer ration. The growth responses produced by crystalline aureomycin or aureomycin supplements (Aurofac) in young calves were reconfirmed by various workers within 12 months after the first results were published *(9, 48, 75, 85, 88, 103, 105)*. Rusoff *et al. (103, 105)* fed both the crude aureomycin supplements and crystalline aureomycin and obtained similar results with both products.

Interest has been shown concerning the effect of the calf's age on the growth response produced by antibiotic feeding. The Louisiana workers *(105)* observed that aureomycin-fed calves began to gain in weight faster than the control calves at about 5 weeks of age. An improvement in over-all condition was noted from the feeding of aureomycin. These workers also observed a breed difference in the response to aureomycin supplementation. Aureomycin produced a 25% increase in growth in Jersey calves but only an 18% increase in Holstein calves. These differences, however, were not statistically significant. Loosli *et al. (75)* and Loosli *(72)* reported the results of antibiotic studies involving 40 pairs of Holstein calves fed a milk replacer ration early in life and hay and calf starter in addition. Half of the calves were fed an aureomycin supplement the first 8 weeks of life. For the first 8 weeks the antibiotic-fed calves gained approximately 22% faster, but growth data to 16 weeks of age indicated that no further growth improvement was made by the aureomycin-fed calves. Murley *et al. (88)* evaluated aureomycin in the rations of calves fed either a whole milk, hay, and concentrate ration or a reconstituted skimmilk, hay, and concentrate ration. A greater growth response was obtained when aureomycin was added to the whole milk ration, 31.9% as compared with 17.2%, but part of this difference probably can be explained by the fact that the whole milk–fed calves not given any antibiotic gained at a slower rate than calves fed reconstituted skimmilk and no antibiotic.

Bartley *et al. (9)* reported an extensive trial comparing two levels of aureomycin supplementation, 3 and 9 g. of an aureomycin supplement per 100 lb. of body weight daily. This trial was conducted from 1 week of age to 22 weeks of age. At the end of the 22nd week the control calves had gained 356%; the

calves fed 3 g. of aureomycin supplement, 410% ; and those fed 9 g. of aureomycin supplement, 400% of their initial body weight. Ragsdale growth standards *(95)* indicated that the calves should have increased in weight 340%. In contrast to the earlier Kansas study *(3)*, the control calves made satisfactory gains in this experiment; thus, by comparison the supplemented calves showed a response to aureomycin when the level of infection was not high. Murdock *et al. (85)* reported results which showed that aureomycin feeding produced a growth response up to 6 weeks of age but by the time the calves were 12 weeks of age both the controls and the aureomycin-fed calves had made similar gains. Morrison and Deal *(84)* approached the question of feeding antibiotics to young dairy calves in a slightly different manner than had been done previously. These workers, believing that the first 2 weeks of a calf's life were the most critical time, attempted to evaluate antibiotics during this period. An aureomycin supplement was fed in the milk for the first 2 weeks after the calf was removed from the dam at 3 days of age. The supplement was fed at the rate of 1% of the dry matter of the milk. The amount of pure aureomycin fed was not noted. The supplement had no effect on the average daily gain to 12 weeks of age, general health, incidence of scours, or feed consumption. No indication was given as to the level of infection that was present; therefore, unless calf scours or other infections were problems, it is doubtful if any advantages of the antibiotic could be expected during this short period of supplementation.

b. *Terramycin.* Not until the summer of 1951 were there any indications that other antibiotics might be of value in stimulating the growth rate of calves or reducing the incidence of scours. Cason and Voelker *(17)* fed two levels of a terramycin supplement, to supply 15 and 30 mg. of terramycin per 100 lb. of body weight daily, for the first 8 weeks of the calf's life. In this study a growth response was obtained, but it was not great enough to be statistically significant. The terramycin supplement did appear, however, to aid in the control of calf scours. The same workers *(120)* reported three experiments with antibiotics in November of 1951. In Experiment 1, the feeding of 30 mg. of terramycin per 100 lb. of body weight daily increased the growth rate of calves about 21%, which was significantly greater than the growth of the control calves. No indication was given as to the length of the trial. In Experiment 2, calves fed 100 mg. of terramycin daily gained 28% faster than the control calves. Murdock *et al. (85)* reported that terramycin improved the growth rate of calves from birth to 6 weeks of age, but when terramycin supplementation was discontinued at this age the antibiotic-fed calves grew at a slower rate during the period 6-12 weeks of age than the control calves, resulting in a slight growth depression for terramycin-fed calves over the entire 12-week feeding period.

The information available on the value of terramycin in the rations of calves is not nearly so voluminous as that concerning aureomycin. Several reports *(54, 55, 65, 69, 78, 83, 127)* in recent years have added much needed information concerning the feeding value of this antibiotic. Kesler and Knodt *(55)* fed Holstein calves a milk replacement ration supplemented with 20 mg. of terramycin per 100 lb. of body weight daily. This ration was fed for 8 weeks, at which

time one-half of the control calves were fed terramycin, and terramycin feeding was discontinued on one-half of the terramycin-fed calves. The calves fed terramycin were 22.7% heavier at 8 weeks than the control calves but their gains were about equal to those of a third group fed whole milk. Cessation or initiation of terramycin feeding at 8 weeks of age had very little effect on growth, thus showing that the maximum growth response was obtained by the time calves were 8 weeks old.

In a later report, which apparently included some of the data previously reported *(54)*, the statement was made that terramycin improved the growth rate of the calves up to 8 weeks of age but not to 16 weeks of age. It was also stated in the later report that starting terramycin feeding at 7 weeks of age reduced the growth rate of the calves. In a second trial calves were fed a milk replacement ration containing 2 g. of crystalline terramycin per 100 lb. of milk replacement. Terramycin was fed for only 7 weeks, although the trial was 12 weeks in length. In this study terramycin stimulated the growth rate of calves, as measured by both weight gain and increase in height at withers.

MacKay *et al. (78)* reported that a terramycin supplement improved the growth rate of Holstein, Ayrshire, Guernsey, and Jersey calves over the growth rate of control calves. Since several breeds were involved and it was difficult to balance both groups in respect to breeds, the increase in growth rate was compared to Ragsdale standards *(95)*. The control calves gained 13.6% and the terramycin calves 20.2% faster than Ragsdale standards. The length of the trial was from birth to 12 weeks of age, and terramycin was fed at the rate of 30 mg. per 100 lb. of body weight daily. The calves consumed from 19 to 82 mg. of terramycin daily. The antibiotic-fed calves appeared to be in better condition than the calves not fed terramycin, and the feces of the terramycin calves were firmer than those of the control calves.

Moody *et al. (83)* reported that terramycin stimulated the growth rate of female calves more than that of male calves and found no evidence that small calves responded more to terramycin supplementation than did the larger calves. Lassiter *et al. (69)* found that crystalline terramycin did not improve the growth rate of Holstein and Jersey calves fed a limited amount of milk and an all-plant starter, whereas a terramycin supplement had stimulated the growth rate of calves 12% in a previous trial *(65)* when a similar system of feeding had been employed. A slight beneficial effect on calf scours was noted from the feeding of crystalline terramycin. Williams and Jenson *(127)* observed no improvement in the growth of calves when terramycin was included in a milk replacement feed.

c. *Penicillin.* The third antibiotic to receive consideration was penicillin. Because it has proven to be of considerable value in stimulating the growth rate of chicks and, to some extent, swine, it might be expected to perform similarly in the rations of young dairy calves. The results to date are very much to the contrary. Bloom and Knodt *(12)* and Knodt and Bloom *(58)* reported that potassium penicillin when fed at the rate of 10 p.p.m. in a milk replacement feed significantly lowered the gains of Holstein calves. Some deaths were recorded, and penicillin lowered the consumption of starter considerably. The

Pennsylvania workers *(60)* later reported results of feeding both potassium and procaine penicillin in a milk replacement feeding program. Potassium penicillin was fed at a level of 0.5 g. per 100 lb. of milk replacement feed. Procaine penicillin was fed at the levels of 0.1 g., 0.3 g., 0.9 g., and 2.7 g. per 100 lb. of milk replacer. All levels of procaine penicillin and the one level of potassium penicillin reduced the average daily gains of the calves, as compared with the control calves, from birth to either 8 or 12 weeks of age. All levels of procaine penicillin with the exception of the 0.3 g. level produced improved gains over those of calves receiving the 0.5 g. level of potassium penicillin. In this study there did not appear to be a depression in feed consumption, as was observed in the earlier studies *(12, 58)*. The incidence of scours was high among calves of all groups but was about twice as severe in penicillin-fed calves as in calves not fed penicillin. Penicillin appeared to increase the incidence of scours rather than to reduce it, as had been shown to be true of aureomycin and, possibly, of terramycin. Gardner *et al. (32)* fed veal calves procaine penicillin at the rate of 15 mg. per pound of milk up to 56 days of age. Under these conditions penicillin had no effect on the growth rate of the calves. Voelker and Jacobson *(121)* fed procaine penicillin G to calves from 4 to 88 days of age on a whole milk replacement diet. The calves fed penicillin gained only 86% as much as the control calves and consumed slightly less concentrates. The calves were fed penicillin at the rate of 40 mg. daily for the first 60 days and 80 mg. daily from the 61st to the 88th day of life. Hibbs *et al. (41)* found that procaine penicillin did not improve significantly the growth rate of calves fed a hay to grain ratio of 3:2, although a slight improvement in growth was observed.

In contrast to these studies, Hogue *et al. (44)* reported favorable results from the feeding of a mixture of bacitracin and penicillin (4:1), and Kon *et al. (62)* reported similar results with procaine penicillin. In the Cornell study *(44)* the mixture of bacitracin and penicillin significantly increased the growth rate of calves for the first 7 weeks (18%) and decreased the days of abnormal feces. However, at 16 weeks of age there was no difference between the calves fed the antibiotic and the control calves as far as weight gain was concerned. It is of interest to note that the bacitracin-penicillin fed calves consumed less starter than the control calves. This is in agreement with the results of Bloom and Knodt *(12)*. Although in the Cornell studies a group of calves fed only penicillin was not included, the reduction in starter consumption by the bacitracin-penicillin calves was probably due to the presence in the mixture. Kon *et al.* conducted two experiments with procaine penicillin with calves from birth to 12 weeks of age. Penicillin was fed at the level of 80 mg. per calf daily. In Experiment 1 the control calves gained 1.04 lb. daily, whereas the penicillin-fed calves gained 1.25 lb. daily. In the second experiment a slight increase in growth was obtained, but it was not nearly so great as in the first experiment, the control calves gaining 1.30 lb. and the calves fed penicillin 1.40 lb. daily. These growth differences were not statistically significant. Aureomycin was used in a third experiment, and both aureomycin and penicillin caused a marked reduction in the incidence of calf scours. These English workers

also observed a greater growth response from the feeding of antibiotics to calves born during the fall months than with calves born during the spring months. It was believed that this observation could be related to the seasonal incidence of infection, which was believed to be highest during the fall months.

It is difficult to explain the conflicting reports in the literature concerning the feeding of penicillin to young dairy calves. Kon *et al. (62)* fed 80 mg. of penicillin per calf daily, whereas Knodt and Ross *(60)* fed from 0.2 to 19 mg. daily, depending upon the age of the calf, the level of penicillin fed, and the amount of milk replacement fed, since the penicillin was included in the milk substitute at various levels. This shows that actually Kon *et al.* fed penicillin at a higher level than Knodt and Ross. It is difficult to understand how a lower level of penicillin would cause a growth depression and a higher level of the same antibiotic would produce a growth stimulation. Such factors as climate, environmental conditions, feeding programs, level of infection present, and the location of the two experiments must be considered in the interpretation of these data. Voelker and Jacobson *(121)* fed 40-80 mg. daily and Hibbs *et al. (41)* fed 0.9 mg. of penicillin per calf daily for the first 7 weeks and 1.0 mg. per pound of feed the remainder of the trial.

d. *Other antibiotics.* Available data on the value of other antibiotics in the rations of young dairy calves are extremely limited. Hogue *et al. (44)* fed streptomycin at levels of 10, 20, and 40 mg. per 100 lb. of body weight daily up to 7 weeks of age. The control calves gained 0.78 lb. daily, whereas the streptomycin-fed calves gained 0.92 lb. daily, or about 18% faster. The trial was conducted for 16 weeks, and at the end of this period the average daily gain over the entire period was the same for both groups. The aureomycin-fed calves gained 1.00 lb. daily, or 28% faster, than the control calves. All antibiotics significantly increased daily gain and heart girth and decreased the days of abnormal feces up to 7 weeks of age. Rusoff *et al. (111)* reported on the effect of soluble streptomycin on the growth of young dairy calves. Streptomycin was fed at levels of 30 and 50 mg. per calf daily to 12 weeks of age. The 50-mg. level improved the growth rate of the calves 15%, but the 30-mg. level failed to have any effect on average daily gain. During the first 8 weeks the 50-mg. streptomycin-fed calves had very loose feces and long, rough haircoats. Results were presented which indicated that streptomycin stimulated the growth of the calves mostly after 8 weeks of age. Owen and Allen *(92)* fed calves a whey product milk replacement feed from 4 to 88 days of life and supplemented this ration with terramycin, bacitracin, chloromycetin, or arsonic acid. Terramycin stimulated daily gain 33%, bacitracin 24%, and chloromycetin only 10%. All antibiotic-fed calves increased faster in height at the withers, but not in chest and barrel circumference, than the control calves. Rusoff and Davis *(104)* found that neither tyrothricin nor bacitracin stimulated the growth of calves. Ellsworth *et al. (30)* observed that aureomycin improved the growth rate of calves 23% and bacitracin improved the growth rate 14% from 3 days to 100 days of age. These calves were fed considerable quantities of either whole milk or reconstituted skimmilk. The growth of the control calves was 24% greater than Ragsdale standards.

By 1952 it had been established that various antibiotics, particularly aureomycin, and probably terramycin, did stimulate the growth rate of calves early in life. Since that time, studies which added more information as to the value of antibiotics in the rations of calves under varied feeding conditions were reported by MacKay *et al. (77)*, Lassiter *et al. (64)*, Voelker and Jacobson *(121)*, Murley and Pou *(89)*, and King and O'Dell *(57)*. In the study by Murley and Pou aureomycin did not improve the growth rate of the calves. Considerable interest has been manifested as to the factors which might influence the response obtained from the feeding of antibiotics to calves.

2. *Effective antibiotic level.* One of the most important questions concerning the feeding of antibiotics to dairy calves is the amount of antibiotic which must be fed to obtain a maximum growth response and to have a significant effect on the incidence of scours. Information of this nature is extremely difficult to interpret because the methods of feeding antibiotics have not been consistent. Only data on the oral administration of antibiotics will be discussed at this point because this method appears to be the only practical means of administration. Also, in most cases only data on the value of various levels of antibiotics obtained in the same experiment have been included in order to avoid the effect of different feeding regimes and other uncontrollable factors.

As previously mentioned, Bartley *et al. (9)* fed 3 and 9 g. of an aureomycin supplement per 100 lb. of body weight daily. The amount of pure aureomycin fed is not known, but both levels produced similar improvements in growth. In a later study, these workers *(2)* fed calves 15 and 45 mg. of aureomycin per 100 lb. of body weight daily from birth to 25 weeks of age. The 45-mg. level was more effective in controlling scours and colds and stimulated growth more than the 15-mg. level. The control calves gained 291% of their initial body weight; the 15-mg. fed calves, 314%; and the 45-mg. level calves, 349%. Evidence was presented which indicated that the higher level of antibiotic feeding reduced the difference between sexes in the growth response to the antibiotic. Another interesting observation was that Holstein calves responded to the 45-mg. level as measured by increased growth rate only up to 10 weeks of age, whereas Holstein calves fed the 15-mg. level responded from birth to 25 weeks of age. Jersey calves responded to both levels from birth to 25 weeks of age. When the growth data of Holstein calves fed 45 mg. of aureomycin per 100 lb. of body weight was considered from birth to 25 weeks of age, a significant response from the antibiotic was not observed, although a significant growth response was obtained through the first 10 weeks of the trial. The greatest response from aureomycin feeding was obtained during the first 5 weeks and during the 22-25 week period. No explanation was given for the stimulation in growth during the latter period. In a third study *(7)*, the Kansas workers fed 45 mg. and 90 mg. of crystalline aureomycin daily from birth to 25 weeks of age. Both aureomycin treatments reduced the incidence of infections. The 45-mg. level produced significantly greater gains than the 90-mg. level up to 12 weeks of age. Based on the results of this experiment, these workers proposed that the optimum level of aureomycin feeding should be 45 mg. per 100 lb. of body weight daily for the first 12 weeks and 15 mg.

from 13 to 25 weeks of age. Pritchard *et al. (93)* found no advantage of feeding 60 mg. per 100 lb. of body weight over 15 mg. of terramycin daily. Both levels stimulated the growth of calves slightly over that of the control calves, but this increase was not statistically significant. These workers, however, made a significant observation on the continuation of feeding antibiotics in a dairy herd: The percentage response from the feeding of antibiotics over the growth of the control calves had reduced over a 3-year period. The lack of growth response was not due to poorer growth of antibiotic-fed calves but to improved growth of control calves with each succeeding year. This can probably be explained by the fact that the level of infection of the housing facilities was reduced over the 3-year period by the continued feeding of antibiotics. This supports the initial work of Bartley *et al. (3)*, in which a 70% growth response was obtained from the feeding of aureomycin. This extremely high percentage growth response from aureomycin feeding was probably due to the high incidence of calf scours, which resulted in below normal growth of the control calves. Hogue *et al. (44)* reported the results of studies involving the feeding of aureomycin, streptomycin, and a mixture of bacitracin and penicillin (4:1) at levels of 10, 20, and 40 mg. per 100 lb. of body weight daily. No advantage was observed for any particular level of feeding. All levels of each antibiotic improved growth up to 7 weeks of age. These authors concluded there were no advantages of feeding antibiotics to calves past 7 weeks of age.

Mochrie *et al. (82)* studied the value of various levels of aureomycin in a calf starter upon the growth rate of calves. An aureomycin supplement was fed at levels equivalent to 9, 18, 36, and 54 mg. of aureomycin per pound of starter. Half of the calves were housed in a temperature-controlled barn and the other half in a barn with uncontrolled temperature. The length of the trial was from 2 to 119 days of age of the test animals. Aureomycin at all levels increased daily gain, height at withers, heart girth, and girth of paunch, but these increases in growth were not statistically significant. There was very little difference between the growth rate of the calves fed the various antibiotic levels. Although it was not pronounced, the data indicated that aureomycin stimulated the growth rate of the calves housed in the unheated barn more than that of the calves in the heated barn. Apparently this fact was not related to the level of infection under the two systems of housing because none of the levels of aureomycin had a significant effect on the incidence of scours.

Pennsylvania workers *(13, 14, 59, 61)* have conducted a series of trials testing various levels of aureomycin in milk replacement feeds. Knodt and Ross *(59)* fed levels of 0, 2, 4, 6, and 10 g. of aureomycin per 100 lb. of milk replacement. All levels of aureomycin improved the growth rate of the calves at 8 and 12 weeks of age, although the stimulation from the 6-g. level was very slight. It was not stated but is assumed that aureomycin was fed for only the first 8 weeks. The 2-g. level of aureomycin per 100 lb. of milk replacement improved growth more than any other level at 8 weeks of age and was a close second over the 12-week period. Based on these data, it appears that 2 g. of aureomycin per 100 lb. of milk replacer was adequate to produce a maximum growth response.

Bloom and Knodt *(13)* in a later trial fed 0.5 g., 1.0 g., and 2.0 g. of aureomycin per 100 lb. of milk replacement in both the pure and supplement form. In this study the greatest growth stimulation was obtained during the first 4 weeks of the trial. A growth response was produced by all levels of aureomycin feeding from both the crystalline and the crude form of aureomycin. When the calves were 8 weeks of age, the greatest response was obtained from the feeding of 1.0 g. of aureomycin in the pure form and 0.5 g. and 1.00 g. of aureomycin in the crude form. However, at 12 weeks there was no difference between the levels of aureomycin feeding in the crude form, but the 1.0 g. level produced the greatest growth response when aureomycin was fed in the crystalline form. As can be seen from these data, there appears to be no consistent relationship between the levels of aureomycin fed and the growth response obtained. Knodt *et al. (61)* reported similar results from the feeding of 2, 6, and 10 g. of aureomycin per 100 lb. of milk replacement supplied by an aureomycin supplement. All levels of aureomycin stimulated the growth rate of the calves over the control calves, but there was no significant difference between the various levels of aureomycin.

The Pennsylvania workers fed aureomycin at various levels of a milk replacement ration. In all other studies with one exception *(82)* the antibiotic was fed so that each calf received a given amount daily or at a certain level per 100 lb. of body weight daily. Mochrie *et al. (82)* fed aureomycin at various levels in a calf starter. Since the basis for the feeding of aureomycin has varied, an attempt will be made to correlate the data of the Pennsylvania workers with other available data on the optimum level of antibiotic that should be fed. Since in the Pennsylvania work the amount of antibiotic consumed daily varied with the age of the calf, according to the milk replacement feeding program, the amount of antibiotic fed at various ages will be calculated. Usually, the amount of milk replacement fed was 0.4 lb. for the first 10 days, 1.0 lb. from the 11th to the 28th day, 1.2 lb. from the 29th to the 42nd day, and 1.4 lb. from the 43rd to the 49th day *(61)*. This means that calves fed the antibiotic at the rate of 1.0 g. per 100 lb. of milk replacer received 4 mg. daily for the first 10 days, 10 mg. daily for the 11th-28th day, 12 mg. daily for the 29th-42nd day, and 14 mg. daily for the 43rd-49th day. When Bloom and Knodt *(13)* fed 0.5 g. of aureomycin per 100 lb. of milk replacement, the calves received only 2 to 7 mg. of aureomycin daily over the 7-week trial. In their study when this amount of aureomycin was supplied by crystalline aureomycin a satisfactory growth response was not obtained, but the same amount from a crude supplement did produce a significant growth response.

In studies by the Pennsylvania workers *(13, 59, 61)* a satisfactory growth response was obtained when aureomycin was included in the ration at the rate of 2.0 g. of aureomycin per 100 lb. of ration. The calves fed this level consumed from 8 to 28 mg. of aureomycin daily. When this level of antibiotic feeding was converted to an amount of antibiotic per 100 lb. of body weight, estimated values were used since the weights of the calves are not known for any particular age. However, if it is assumed that the Holstein calves weighed 90 lb. at less than 10 days of age and 160 lb. at 7 weeks of age, the calves received about 9 mg. of

aureomycin per 100 lb. of body weight daily during the period from birth to 10 days of age and 18 mg. per 100 lb. of body weight daily during the period from the 43rd to the 49th day. These values agree closely with the results of Hogue *et al. (44)*, which showed that the feeding of 10 mg. per 100 lb. of body weight daily stimulated growth as much as 40 mg. per 100 lb. of body weight. Pritchard *et al. (93)* found that the feeding of 15 and 60 mg. of antibiotic per 100 lb. of body weight daily stimulated growth similarly. Bartley *et al. (2)*, however, found that 15 mg. did not produce the same growth response as that obtained by the feeding of 45 mg. of aureomycin per 100 lb. of body weight. In a later study 45 mg. was as effective as 90 mg. per 100 lb. of body weight as a growth stimulant. Mochrie *et al. (82)* found that 9 mg. of aureomycin per pound of starter possessed the same growth-promoting properties as 54 mg. Based on these studies, it appears that the minimum effective level of aureomycin, and probably of terramycin, for feeding young dairy calves is approximately 15-20 mg. of antibiotic per 100 lb. of body weight daily. This conclusion does not agree with the observations of Warner *(123)*, who stated that the optimum level of aureomycin feeding was about 30 mg. per calf daily, but it should be remembered that at the time Warner established this level of aureomycin feeding, much of the data used by the present author was not available. It should be pointed out, however, that a slightly higher level than 15-20 mg. per 100 lb. of body weight probably should be used by farmers to combat high level infections that might exist on some farms. Present data indicate that milk replacement feeds should contain about 20 mg. of antibiotic per pound of feed.

The previous discussion applies only to aureomycin and terramycin. Data available at the present time on other antibiotics are not sufficient to draw valid conclusions as to proper levels of supplementation. It is very difficult to establish a satisfactory value for the amount of antibiotic that should be included in calf starters. This is true because the consumption of starter by the calf early in life is small and variable. The work of Mochrie *et al. (82)* indicated that 9 mg. of aureomycin per pound of starter produced a maximum growth response, but apparently in this study calf scours was not a problem. It is possible that an added growth response would have been obtained from the feeding of aureomycin if the antibiotic had been included in the milk as well as in the calf starter. At the present time it seems that these antibiotics should be included in calf starters at levels of 10 to 20 mg. per pound of starter.

3. *Method of administration.* Several research workers have been interested in the effect that the method of administration of an antibiotic might have on the growth response produced. Rusoff *et al. (106)* studied this problem by supplementing the rations of one group of calves with aureomycin in the milk and the calf starter and giving a second group of calves weekly intramuscular injections of 400 mg. of aureomycin. In this study orally administered aureomycin increased the average daily gain of the calves 24%, whereas the injected aureomycin improved the growth rate only 15% over that of the control calves. The injected calves showed an improvement in condition over the control calves at 9-10 weeks of age, but the orally fed calves did not show this improvement in

condition until 12-16 weeks of age. It was also found that the injection of aureomycin caused a significant increase in the fat content of the 12th rib and that oral administration caused some increase in fat percentage, but to a lesser extent. In a later study with aureomycin the same workers *(107)* found that injections increased the growth rate of calves 30% and oral feeding 20%. Although these results were contrary to their earlier report, it should be pointed out that the difference between the growth rates of the aureomycin groups was not statistically significant in this study. Richardson *et al. (98)* found that daily or weekly oral feeding of aureomycin was about equally effective in stimulating the growth rate of calves over that of the controls, but this was not true of weekly subcutaneous implantations or weekly intramuscular injections of aureomycin. Bartley *et al. (8)* observed that aureomycin given as one weekly intramuscular injection (125 mg.) was not so effective in improving the growth rate of the calves as daily oral administration (45 mg. daily). Based on these reports, it appears that oral administration of antibiotics is as effective, if not more so, than other methods of administration.

Bloom and Knodt *(14)* and Bartley *et al. (8)* studied various methods of oral feeding of aureomycin. Bloom and Knodt attempted to determine whether the antibiotic should be placed in a milk replacement and the calf starter, the milk replacement only, the calf starter only, or various combinations of these two. The results of this study indicate that it is not necessary to include the antibiotic in the milk replacement feed if the calf starter contains adequate amounts of the antibiotic and if the starter is consumed in adequate amounts. It should be mentioned that the calves in this experiment consumed about 2 lb. of starter daily from birth to 8 weeks of age, which would be considered very good consumption of feed by calves of this age group. It is possible that this amount of starter may not be consumed by calves under farm conditions and, therefore, if a milk replacement is fed it would probably be advantageous to include an antibiotic in this feed.

Bartley *et al. (8)* were interested in the same problem. In a study with calves from birth to 8 weeks of age these workers found that the control calves gained 176% of the starting weight; calves fed 1% of an aureomycin supplement (Aurofac 2A) in a calf starter, 185%; calves fed 45 mg. of aureomycin daily by capsule, 191%; calves fed 45 mg. of aureomycin daily in the milk, 207%; and those given weekly injections of 125 mg. of aureomycin, 183%. These results show that when aureomycin is fed only in a calf starter some growth stimulation will be produced but not so much as when the antibiotic is placed in the milk. These data show the necessity of insuring adequate consumption of aureomycin by calves early in life. It appears that when the antibiotic is fed only in a calf starter, maximum effect may not always be obtained, owing apparently to the low consumption of starter early in the life of the calf. These workers explained the lack of maximum growth response from aureomycin administered by capsule by the fact that the capsule probably went into the rumen upon administration, whereas milk enters the abomasum.

Gaunya *et al. (33)* observed that aureomycin did not improve the growth

rate of calves or influence the incidence of calf scours when included in a calf starter at the rates of only 4.5 and 9 mg. of aureomycin per pound of starter. Apparently the calves did not consume adequate amounts of starter to insure the consumption of the minimum amount of aureomycin required to produce a growth response or to reduce the incidence of calf scours. Certainly, more research is needed to determine more effective means of achieving maximum beneficial effects from the feeding of an antibiotic to calves when it is to be fed in a calf starter.

4. *Influence of type of ration.* Some consideration has been given to the effects of various types of rations on the growth response produced by an antibiotic. Rusoff *et al. (101)* in one study found that the type of plant protein in a calf starter, i.e., soybean oil meal, hydraulic cottonseed oil meal, or degossypolized cottonseed oil meal, did not affect the amount of growth response produced by aureomycin. In a later experiment by these workers *(102)*, which appears to be a continuation of the earlier study, it was found that aureomycin stimulated the growth rate of calves to a greater extent when added to a starter containing soybean oil meal than when added to a starter containing either one of the cottonseed oil meals. Lassiter *et al. (65)* found that an aureomycin supplement improved the growth rate of calves 14% over that of the control calves when added to a calf starter containing corn distillers dried solubles (plant protein) but only 9% when added to a starter containing nonfat dry milk solids (animal protein). The growth rate of calves fed the animal protein ration without an antibiotic was about 10% greater than that of calves fed the plant protein ration without an antibiotic; this may account partly for the greater growth response from aureomycin when added to the plant protein ration. These results are in agreement with those of Sanford *(113)* and Heuser and Norris *(38)*, working with chicks.

Hibbs and Conrad *(39)* and Hibbs *et al. (42)* observed that aureomycin improved the growth of calves raised on a high roughage system but found that variations in the hay to grain ratios (4:1, 3:2, and 2:3) had no effect upon the response from the antibiotic. Elliott and Ellsworth *(29)* found that the greatest response from aureomycin feeding of lambs was obtained on a high-roughage, low-grain ration and the poorest response on a low-roughage, high-grain ration. Clawson *et al. (21)* observed that when calves were fed aureomycin to 10 weeks of age the types of hay fed, alfalfa or prairie, had no effect on the growth response produced. However, from birth to 16 weeks of age, calves fed alfalfa hay responded more to aureomycin supplementation than did calves fed prairie hay. Actually, the data which were shown indicated that aureomycin caused a growth depression in calves fed prairie hay between the 10th and 16th weeks of the experiment. This was true also of calves fed aureomycin for the first 10 weeks, with alfalfa hay feeding starting at 8 weeks of age. In all of these comparisons aureomycin was fed only for the first 10 weeks.

Gardner *et al. (32)* and Swanson and Hinton *(118)* studied the effect that heavy milk feeding might have on the growth response produced by aureomycin. Gardner *et al.* found that aureomycin supplementation of a whole milk–veal calf

ration improved the growth rate of milk-fed veal calves. The exact amount of improvement was not stated. Swanson and Hinton fed calves a commercial milk replacer containing 60% milk products at very high levels until the calves were 30 days old and then reduced the milk replacement feeding until they were removed from milk feeding at 51 days of age. Under these conditions aureomycin improved the growth of the calves 24.1% for the first 50 days and 17.5% over a 16-week experimental period. As shown by these two studies, apparently aureomycin will improve the growth rate of calves even when large quantities of milk are fed. McGilliard *et al. (79)* found that calves fed aureomycin for 35 days followed by cud inoculations on the 36th and 41st days gained as fast during a second 35-day period as a control group of calves and faster than calves fed aureomycin to 35 days and then discontinued without cud inoculations. The growth of all groups of calves was approximately the same at the end of 16 weeks. Although data were reported on only three calves per group, these data indicate that aureomycin possibly caused some disturbance in normal rumen function.

5. *Combination of antibiotics vs. single antibiotics.* Some interest has been shown in the feasibility of feeding a combination of antibiotics rather than the single antibiotic. Edgerly *(28)* found that aureomycin and terramycin improved the growth rate of calves about the same, 17%, but an equal mixture of aureomycin, terramycin, bacitracin, and procaine penicillin improved the average daily gain of the calves over their controls about 27%. It was also observed that aureomycin- and terramycin-fed calves did not grow as fast as control calves 2-3 weeks after the removal of the antibiotic from the ration at 12 weeks of age, but with calves fed the mixture of antibiotics this reduction in the growth curve was not noted when the antibiotics were removed from the ration. Lassiter *et al. (67)* found that the feeding of a combination of aureomycin and terramycin supplements did not produce a greater growth response or afford other advantages than those produced by the single antibiotics.

6. *Crystalline vs. crude antibiotics.* Some workers have questioned the relative feeding value of crystalline antibiotics and the crude supplements. Loosli and Wallace *(73)* found that crystalline aureomycin produced a growth response similar to that secured from an A.P.F. supplement containing aureomycin. Rusoff *et al. (103, 105)* observed similar results with aureomycin. Bloom and Knodt *(13)* fed three levels of both crystalline aureomycin and aureomycin supplements. In every respect equivalent amounts of both forms of aureomycin were fed. Although the growth differences between levels or between methods of supplying aureomycin were not statistically significant, the calves fed the crude aureomycin supplement gained faster than the crystalline aureomycin-fed calves at two of the three levels of aureomycin feeding. Bartley *et al. (7)* fed groups of calves no aureomycin, 45 mg. daily of crystalline aureomycin, 45 mg. daily of aureomycin from Aurofac, and 90 mg. daily of crystalline aureomycin. Growth data from birth to 12 weeks of age of the calves receiving 45 mg. of aureomycin from Aurofac showed that they gained significantly faster than calves fed an equivalent amount of crystalline aureomycin, but from 13 to 25 weeks of age

those fed 45 mg. of crystalline aureomycin gained significantly faster than all other groups. Calves fed 90 mg. of crystalline aureomycin daily at no time gained faster than did calves fed 45 mg. of aureomycin. These workers postulated the presence of a factor(s) in Aurofac which was not in crystalline aureomycin and which benefited the calves until rumen synthesis provided the factor(s).

7. *Skeletal growth.* Throughout this discussion very little has been mentioned about the effect of antibiotics on growth other than gain in body weight. Early workers in this field were concerned as to whether the increase in body weight was due to increased fatness and condition or whether skeletal growth was affected. In general, antibiotics seemingly stimulate skeletal growth as well as body weight. Voelker and Cason *(120)* reported that aureomycin-fed calves showed more structural body growth than did control calves. Since this report, several workers *(42, 44, 46, 47, 54, 61, 64, 65, 67, 106, 107)* have confirmed the observation of the Arkansas workers. Murley and Pou *(89)* observed that aureomycin did not increase skeletal growth, but in this trial neither did it increase body weight gains. Rusoff *et al. (106)* conducted carcass studies of antibiotic-fed calves which showed larger muscles and skeletal size of calves fed aureomycin.

B. *Calf scours.* Bartley *et al. (3)* reported that the feeding of an A.P.F. concentrate containing aureomycin to young dairy calves lowered the incidence of scours. These workers therefore proposed that aureomycin enhanced the growth of calves by preventing scours. Loosli and Wallace *(73)* reported similar results, observing that control calves scoured an average total of 6.9 days per calf from birth to 8 weeks of age and calves fed aureomycin scoured only an average of 2.5 days. Several workers *(1, 6, 7, 13, 44, 49, 59, 62, 68, 103)* have confirmed these early reports. Morrison and Deal *(84)*, MacKay *et al. (77)*, Voelker and Jacobson *(121)*, Murley and Pou *(89)*, Mochrie *et al. (82)*, Knodt *et al. (61)*, Swanson and Hinton *(118)*, and Warner *(123)* reported that aureomycin either had no effect on the incidence of scours or that scours and other types of infections were not a problem under the conditions existing in their studies. In the study by Warner the experiment was conducted in a portion of the housing facilities that had never been occupied by dairy calves, and the level of infection was extremely low. Bortree *et al. (15)* conducted some field trials on the use of aureomycin for the prevention and treatment of scours. In one trial, 58 cases of scours were treated with 51 animals responding immediately, five gradually, and two dying. In another trial, 67 calves were given a 500-mg. oblet of aureomycin at birth. Forty of these calves remained healthy and 27 developed scours of varying severity. Twenty-one of these calves responded to a single treatment of 500 mg. of aureomycin, six required further treatment, and one calf died. These data are the results of field observations, but evidence is presented which at least indicates that antibiotics, in this case aureomycin, are effective in the prevention and treatment of scours.

Voelker and Cason *(120)*, MacKay *et al. (78)*, and Lassiter *et al. (69)* observed that terramycin probably helped to reduce the incidence of scours, although apparently none of these studies was conducted under conditions where scours was a major problem.

Very little information is available concerning the use of other antibiotics in the control of scours. Hogue *et al. (44)* observed that calves fed either aureomycin, streptomycin, or a mixture of bacitracin and penicillin had fewer days of abnormal feces than calves which received no antibiotic for the first 7 weeks of life. Kon *et al. (62)* reported that procaine penicillin reduced the incidence of scours. On the other hand, Knodt and Ross *(60)* found that procaine or potassium penicillin increased the incidence. In an earlier study Knodt and Bloom *(58)* found that penicillin-fed calves had a higher incidence of respiratory ills than did control calves and two penicillin-fed calves died during the experiment, whereas there were no deaths among calves not fed penicillin. Gardner *et al. (32)* and Voelker and Jacobson *(121)* failed to observe that penicillin had any effect on the incidence of scours.

Unfortunately, sufficient data are not available concerning the effect of antibiotics on scours when it was a major problem. Several investigators have obtained indications that various antibiotics aid in the control of scours, but very few experiments have been conducted under conditions when disease problems were encountered. Apparently, the first experiment reported by Bartley *et al. (3)* was conducted under such conditions. In this study it was clearly demonstrated that aureomycin did reduce the incidence of scours.

C. *Feed consumption and feed efficiency.* There is good agreement among investigators as to the effect of antibiotics on the consumption of feed by dairy calves and how well this feed is utilized. Research work in general indicates that all of the antibiotics studied, with the possible exception of penicillin, increase the consumption of hay, starter, or both, and improve the efficiency of feed utilization over that for calves not fed antibiotics.

Rusoff *et al. (105)* reported that aureomycin-fed calves consumed more starter than did calves not fed aureomycin. Loosli *et al. (75)* and Loosli *(72)* found that aureomycin-fed calves consumed 30-40% more starter and required less TDN per pound of gain than calves which were not fed aureomycin. These workers found that aureomycin had no significant effect on hay consumption. Murley *et al. (88)* observed an increase in feed efficiency from the feeding of aureomycin to young calves. Several workers *(6, 9, 13, 30, 39, 49, 59, 65, 67, 82, 118)* have confirmed the early reports that aureomycin does stimulate the consumption of grain or starter. Morrison and Deal *(84)*, MacKay *et al. (77)*, Murley and Pou *(89)*, Bartley *et al. (7)* and Knodt *et al. (61)*, however, observed that aureomycin had very little effect on starter or grain consumption. Several investigators *(7, 59, 61, 75, 82, 84, 107)* found that aureomycin had very little effect upon consumption of hay by young dairy calves, but Murley *et al. (87)* observed that aureomycin-fed calves consumed 21% more hay than did control calves. All calves were fed a basal ration of either whole reconstituted skimmilk, hay, and grain. Jacobson *et al. (46)* confirmed this observation with calves which were 16 weeks of age at the start of the experiment and remained on trial for 12 weeks.

The effect of terramycin on feed consumption and feed utilization by calves has been similar to that reported for aureomycin. Kesler and Knodt *(55)* observed that the addition of terramycin to a milk replacement ration improved

the appetite of the calves. Kesler *(54)* later reported that terramycin appeared to stimulate the appetite of the calves for starter but not for hay. In fact, terramycin-fed calves failed to consume as much hay as did the control calves. MacKay *et al. (78)* reported greater starter consumption for calves fed terramycin than for the controls. Pritchard *et al. (93)* and Lassiter *et al. (69)* failed to find that the feeding of terramycin to calves had any effect on the consumption of hay or grain and the utilization of feed. It should be mentioned that very little improvement in growth was observed by Pritchard *et al.* and that terramycin reduced slightly the growth rate of calves as compared with that of their controls in the study reported by the latter workers.

Disagreement exists concerning the effect of penicillin on dairy calf feed consumption. Bloom and Knodt *(12)* and Knodt and Bloom *(58)* reported that the inclusion of 10 p.p.m. of potassium penicillin in a milk replacement feed reduced feed consumption. The control calves consumed, on the average, 211.3 lb. of starter for the first 10 weeks of the trial, whereas calves fed penicillin during this same period consumed only 102.2 lb. of starter. During the next 2 weeks very little difference in starter consumption was observed. Penicillin did not appear to have any effect on hay consumption. In a later study Knodt and Ross *(60)* again observed a reduction in the growth rate of penicillin-fed calves as compared with calves not fed penicillin, but the growth depression appeared to be due to a higher incidence of scours among penicillin-fed calves rather than to a reduction in feed consumption, although the control calves were slightly more efficient in feed utilization than the penicillin-fed calves. Voelker and Jacobson *(121)* observed that procaine penicillin G when fed to calves caused a slight reduction in the consumption of starter. On the other hand, Kon *et al. (62)*, Gardner *et al. (32)*, and Hibbs *et al. (41)* found that penicillin had very little effect on the consumption of feed or feed utilization by calves.

Data concerning the effect of the other antibiotics on feed consumption are extremely limited. Ellsworth *et al. (30)* found that a bacitracin supplement when fed to calves had no effect on starter or hay consumption but did increase the efficiency of feed utilization. Hogue *et al. (44)* found that both streptomycin and a mixture of bacitracin and penicillin (4:1) increased feed efficiency of the supplemented calves over the control calves. In this study the calves given bacitracin and penicillin consumed slightly less starter than did those fed aureomycin, streptomycin, or no antibiotic.

On the basis of these data, aureomycin, terramycin, and possibly bacitracin and streptomycin seemingly increase the consumption of feed and/or improve the efficiency of feed utilization in young dairy calves. There appears to be very little difference in the effect of aureomycin and terramycin, although it must be remembered that considerably more research is needed to substantiate the stimulatory effects of terramycin on feed consumption.

In this discussion the term "efficiency of feed utilization" signifies the units of feed required to produce a unit of gain in body weight. This usage should not be confused with an improvement in the utilization of feed as might be determined by metabolism studies. Since antibiotics in general improve the rate

of gain of calves, an improvement in feed efficiency might be expected unless a corresponding increase in feed consumption occurred. Apparently this does not occur, at least not in all cases; therefore, an increase in feed efficiency usually results. These conditions pose the question as to whether the improvement in feed efficiency from the feeding of antibiotics is due to an increased growth rate by an antibiotic-fed calf or an actual improvement in the utilization of a unit of feed. Unfortunately, data are not available concerning young dairy calves as to whether an increase in growth would be obtained if the control calves and antibiotic-fed calves were fed equal amounts of feed throughout the aureomycin feeding period. Eating habits of young calves make this type of data extremely difficult to obtain.

D. *Metabolic processes and the possible modes of action of antibiotics.* Several workers have studied the effects of antibiotics on various metabolic processes in the young calf in an effort to determine why antibiotics improve the growth rate. Investigators working with chicks have established that antibiotics in the ration probably cause some change in the intestinal flora of the chick that is responsible for the improvement in the growth rate.

Loosli *et al. (75)* observed that aureomycin when fed to calves under 16 weeks of age had very little effect on the total bacteria count of the rumen or the types of bacteria present. Rusoff *et al. (105)* failed to find any evidence from the examination of rumen smears that aureomycin had any effect on the microscopic flora of the rumen. Voelker and Cason *(120)* made bacteriological studies of colon material and found no consistent differences in bacterial population between terramycin-fed calves and those not fed terramycin. Ellsworth *et al. (30)* observed that neither aureomycin nor bacitracin had any significant effect on the total number of bacteria of either the coliform or streptococci groups in the feces of calves. Bartley *et al. (9)* and Rusoff *et al. (101, 102)* reported results similar to the data discussed previously, indicating that aureomycin has no consistent effect on the intestinal microflora of the young calf.

Several investigators have been interested in the effect of antibiotics on the establishment and function of rumen microorganisms in the young calf. Kesler and Knodt *(55)* found that rumen inoculum taken from calves not fed terramycin digested cellulose in the artificial rumen at the rate of 67.4%, whereas the inoculum from terramycin-fed calves had a cellulose digestive power of only 24.2%. These samples were taken from calves 12 and 16 weeks of age. In another trial it was observed that calves not fed terramycin had a cellulose digestive power of 49.5%, but after they were fed terramycin the cellulose digestion in the artificial rumen dropped to 12.3%. When terramycin supplementation was discontinued, cellulose digestion returned to normal in 6 days. It was further found that neither the thiamin nor the riboflavin level in the rumen of calves at 6, 12, or 16 weeks of age was affected by terramycin feeding. Hibbs and Conrad *(39)* observed that aureomycin had no effect on the average total steam volatile fatty acids or acetic acid content in rumen juice of calves 12 weeks old. Rumen propionic acid was slightly lower and butyric acid slightly higher in aureomycin-fed calves than in control calves. In a later study, Hibbs *et al. (42)*

conducted metabolism studies with 13-week-old calves which had been fed aureomycin since birth. These studies indicated that aureomycin had no effect on dry matter, cellulose, or protein digestion. Aureomycin-fed calves did have a slightly higher retention of nitrogen. In those studies and in a subsequent experiment *(25)* aureomycin had no significant effect on the riboflavin and thiamin content of rumen juice and urine of calves. After aureomycin feeding was discontinued, a slight reduction in growth occurred. During this period a change in "hay group" bacteria of the rumen was noted, which was possibly the reason for the reduction in growth during this period.

Radisson *et al. (94)* in artificial rumen studies found that rumen samples taken from aureomycin-fed calves had a lower digestion of filter paper cellulose with or without starch or grain, and/or when grass juice was added than rumen samples taken from calves not fed aureomycin. When alfalfa hay replaced filter paper only a slight, if any, inhibition was noted. These workers postulated the presence of a factor in alfalfa hay not in filter paper which prevented the depressing effect of aureomycin on cellulose digestion.

Rusoff *et al. (107)* reported that aureomycin had no effect on rumen environment (pH or Eh), fiber digestion in the artificial rumen, rumen flora, or B-vitamin levels of rumen fluid. These workers found that the injected aureomycin was excreted mainly in the urine and the oral-administered aureomycin in the feces. Some aureomycin was found in the bile of injected calves, which was thought to explain why aureomycin was found in the feces of injected calves. These workers stated that this probably signified that the rumen had been bypassed; also, since a growth response was produced by the injection of aureomycin, they postulated that the rumen was not the site where aureomycin stimulated growth. It was also stated that the small amount of aureomycin in the intestine of injected calves indicated that probably the intestinal flora was not involved in the growth stimulation. Another interesting observation was that aureomycin-fed calves showed heavier weights and were larger in size of carcass and bone than control calves.

Mann *et al. (80)* studied the effect of feeding aureomycin to calves on the establishment of normal rumen microflora and microfauna. They found that aureomycin-fed calves had a less acid rumen content and produced a rumen pH (less than 6) needed for rumen bacterial and protozoan activity at a much earlier age than calves not fed aureomycin. Aureomycin had very little effect on the final formation of a typical rumen viable streptococcal population. These workers believed that when aureomycin was given orally it did not act directly on the rumen microorganisms. Bartley *et al. (6)* found that aureomycin had no influence on the strength and number of rumen movements or rumen tone in young calves.

Murley *et al. (86)* found that aureomycin had no effect on the reducing sugars and nitrogen content of the urine of calves or on the fecal excretions of dry matter, reducing sugars, nitrogen, ether extract, and ash. Murley *et al. (87)* also observed that when calves were fed a restricted diet, aureomycin produced a slight improvement in the utilization of carbohydrates, nitrogen, ash, and ether

extract, but these differences were not significant. Lassiter *et al. (68)* studied the effect of aureomycin on the digestion of feed nutrients by young dairy calves, checking digestibility at 5, 8, and 11 weeks of age. Although uniform feed intakes were extremely difficult to obtain at the 5- and 8-week-old trials, it was contended that these trials were needed since it has been shown that the greatest stimulation in growth by aureomycin occurs by the time calves are 8 weeks old. These studies showed that aureomycin had no effect on the digestion of any feed nutrient over the 12-week trial or for any single digestion period.

Numerous investigators have studied the influence of antibiotics on various blood constituents. Murley *et al. (86)* observed that blood sugar levels rose slightly more rapidly and exhibited a greater increase in aureomycin-fed calves than in the control calves. These differences were, however, not significant. Hibbs *et al. (39, 40)* observed that 8- to 12-week-old calves fed aureomycin maintained a blood sugar level of about 9 mg. per 100 ml. higher than that found for the control calves. Voelker *et al. (122)* studied 1,500 blood samples from animals fed aureomycin (200-240 mg. daily) and found that aureomycin had no effect on blood glucose levels. The age of the animals used in this study was not stated, but it is believed that the dairy animals involved were growing heifers and not young calves such as those used by the Ohio workers *(39, 40)*.

Various workers *(31, 50, 82, 92, 112, 114, 115, 121)* have studied the effect of antibiotics on such blood constituents as blood erythrocyte count, hemoglobin percentage, packed cell volume, red blood cell counts, corpuscular volume, corpuscular hemoglobin, corpuscular hemoglobin concentration, plasma "Allen" fat levels, plasma calcium and inorganic phosphorus levels, and the blood levels of various vitamins but have found none of these blood constituents to be affected significantly by the feeding of aureomycin to calves. Lassiter *et al. (68)* found that aureomycin did not affect blood levels of urea nitrogen but did cause a significant depression in blood nonprotein nitrogen levels during the first 7 weeks of the calf's life. It was also during this period that the greatest stimulation in growth occurred from the feeding of aureomycin. Since aureomycin did not produce an increase in the digestibility of protein, these data were interpreted to indicate a greater utilization of absorbed nitrogen by calves fed aureomycin. Unfortunately, nitrogen balance values were not obtained in these studies.

Rusoff *et al. (112)* observed that aureomycin had no effect on the weight of the pituitary, thyroid, or thymus glands or the liver when recorded as a percentage of body weight. However, it was found that aureomycin caused a decrease in the thickness of the duodenal and jejunal sections of the intestine and an increase in the thickness of the ileum section of the intestine as compared with those of the control animals. Hester *et al. (37)* studied the distribution of orally administered and injected aureomycin in the bodies of dairy calves that had shown a growth response from aureomycin supplementation. In the injected calves the highest concentration was in the urine, with measurable amounts in the liver and kidneys. None was found in the spleen, thymus, pituitary glands, or muscles of any calves. When aureomycin was injected, none was found in the rumen. In the small intestine the concentration increased from the upper to the

lower portions and decreased in concentration in the large intestine between the lower small intestine and the anal end of the large intestine. Voelker *et al. (119)* concluded from in vitro studies that the growth-promoting action of aureomycin and terramycin was not due to a lowering of the surface tension of the intestinal tract contents since these antibiotics do not lower the surface tension of water, whereas penicillin and various surface-active agents do, yet these latter products did not increase the growth rate of calves.

Little discussion has been devoted to the possible modes of action by which antibiotics stimulate the growth rate of young dairy calves. Two general types of action have been suggested. Bartley *et al. (3)* proposed that aureomycin improved growth by reducing the incidence of scours. Growth responses have been reported *(13, 32, 59, 61, 67, 77, 123)*, however, from the feeding of antibiotics to calves when the incidence of scours was not a problem. However, such reports do not rule out the possibility of subclinical infections which would probably never result in scours, yet if they were reduced by feeding an antibiotic an increase in growth of the calves would probably result. Considerably more research, possibly under farm conditions, is needed on the effect of antibiotics on the control of scours when the level of infection is high.

Loosli *(72)* and Jacobson *et al. (47)* have proposed that antibiotic-fed calves grow faster because of increased appetite. The question arises as to whether such calves eat more because of the action of the antibiotic or because of their larger size.

TABLE 1

Effect of antibiotics on the consumption of hay and grain by young dairy calves

Reference	Consumption of feed (daily)		Length of trial
	Control calves	Antibiotic calves	
Aureomycin	*(lb. feed per lb. body wt.)*		*(wk.)*
Loosli *et al. (75)*	0.59	0.66	8
Bartley *et al. (6)*	0.27	0.37	7
Ibid.	0.91	0.93	12
Lassiter *et al. (67)*	1.29	1.34	12
Bloom and Knodt *(13)*	1.33	1.31	12
Hibbs *et al. (42)*	1.11	1.21	12
Mochrie *et al. (82)*	0.37	0.36	5
Ibid.	2.09	2.15	16
Murley *et al. (87)*	1.09[a]	1.08[a]	16
Swanson and Hinton *(118)*	2.02	2.07	16
Terramycin			
Lassiter *et al. (67)*	1.29	1.29	12

[a] TDN/lb of body weight.

The data presented in Table 1 are a further attempt to answer this question. The feed consumption (hay and grain) data for supplemented and calves not fed antibiotics from several experiments have been recalculated in terms of daily feed consumption per pound of body weight. In fairness to the original authors it should be stated that these figures have been calculated from the data presented and were not included in the original reports. In a few cases an average starting weight of the calves was assumed to be the average for the breed

of calves involved in the study. The daily feed consumption per pound of body weight was calculated by dividing the total amount of feed consumed over the experimental period by the end body weight of the calves and by the number of days in the trial. It was impossible to calculate the consumption of feed per pound of body weight at various stages of growth since the weight of the calves and the consumption of feed were quoted only in terms of average daily gain and total feed consumption over the entire experimental period. In the experiment by Bartley *et al. (6)* this information was already stated in terms of feed consumption per 100 lb. of body weight on a weekly basis.

When the feed consumption data from these experiments are calculated in this manner some very interesting results are revealed. In every experiment, with the exception of the studies by Bartley *et al. (6)* and Hibbs *et al. (42)*, there was very little difference in the consumption of feed by antibiotic-fed calves and by control calves when the weight of the calves was considered. In the study by Bartley *et al.* aureomycin-fed calves consumed more hay and grain per pound of body weight during the first 7 weeks of age but not from birth to 12 weeks of age. Mochrie *et al. (82)* presented data on calves from birth to 5 weeks of age as well as from birth to 16 weeks of age, but in this study aureomycin improved the growth of the calves only about 5% over the 16-week trial. These data certainly do not answer completely the original question but do show that much of the increased feed consumption by antibiotic-fed calves may be due to the larger size of such calves. If antibiotics affected the appetites of calves directly, one might expect calves fed antibiotics to consume more feed per pound of body weight.

Generally, very few data at present indicate that the microflora of the intestinal tract has any great connection with the growth response produced by antibiotics in dairy animals. This does not mean, however, that antibiotics do not cause changes in the flora of the digestive tract of the calf, because it is doubtful if bacteriological methods have been perfected well enough that the microflora of the digestive tract can be characterized accurately to determine all the changes that could occur as a result of antibiotic feeding.

Two definite modes of action have been postulated, and apparently there is very little relationship between these two. Rusoff *et al. (107)* have presented evidence which indicates that in some manner aureomycin might stimulate the pituitary gland to produce more growth hormone, resulting in greater bone metabolism and over-all growth. These workers rule out the rumen of the calf as being the site of action since growth responses from injected aureomycin were observed. A small amount of aureomycin was found in the intestinal tract and feces of injected animals, but this suggested that the bile was probably the means of excretion into the intestinal tract, thus by-passing the rumen. It should be pointed out that Kraus *et al. (63)* found aureomycin in the saliva of humans after oral administration; this would provide a pathway of excretion that would not by-pass the rumen. In a later study these Louisiana workers *(37)* could not detect any aureomycin in the rumen of injected calves. This finding certainly makes it appear that the rumen is probably not the site of action of antibiotics.

Another supporting reason for this statement is that several workers have shown that the maximum growth response obtained from the feeding of antibiotics occurs before the calf is 8 weeks of age. It is generally accepted *(56)* that the rumen does not begin functioning until the calf is about 3 weeks old and is not fully developed until some time later. Knodt and Bloom *(13)* found that the greatest growth stimulation from aureomycin feeding occurred during the first 4 weeks of the calf's life.

Rusoff *et al. (107)* also attempted to rule out the intestinal tract as the site of action of antibiotics by the fact that only very small amounts of aureomycin were found in the intestinal tract and feces of calves injected with aureomycin. These calves were injected with 400 mg. of aureomycin weekly. It should be remembered that this amount of aureomycin, about 57 mg. daily, was about two and one-half times the amount of aureomycin that is believed to be the effective oral level. It is possible, therefore, that enough aureomycin was excreted into the digestive tract by way of the bile ducts to cause a stimulation in growth. In a later study *(37)* the Louisiana workers did not find any measurable amounts of aureomycin in the pituitary gland. One might expect to find aureomycin in this gland if that is the means by which this antibiotic improves the growth of calves.

Hibbs *et al. (42)* suggested that the mode of action of antibiotics is one of an alteration in energy metabolism, possibly involving the microflora of the rumen. These workers found that aureomycin-fed calves had significantly higher blood sugar levels than control calves during the 8- to 12-week-old period. By chromatographic separation it was found that 12-week-old aureomycin-fed calves had a lower percentage of propionic acid and a higher percentage of butyric acid in the rumen juice than did control calves. It was believed there was a relationship between these rumen short-chain fatty acid values and blood sugar levels. Therefore, it was suggested that a possible mode of action of antibiotics was through energy metabolism. It was further stated that aureomycin possibly has an energy-sparing effect manifested through its effect on the microflora of the rumen and lower digestive tract. These were very interesting findings, but two things should be remembered in light of these data. Voelker *et al. (122)* analyzed 1,500 blood samples of dairy animals fed aureomycin and found no effect on blood sugar levels. It is possible that these values were obtained with older animals than the 8- to 12-week-old calves used by Hibbs *et al. (42)*. The other point is the relationship between the time the maximum antibiotic stimulus occurs and the time Hibbs *et al.* observed this alteration of energy metabolism. It is not known, of course, when the stimulation in growth from the feeding of aureomycin occurred in this experiment, but most workers agree that the greatest stimulation from aureomycin feeding results during the first 7-8 weeks of the calf's life. Hibbs *et al.* did not observe this energy relationship until the calves were 8-12 weeks old. These blood and rumen values were checked before the calves were 8 weeks old, but no differences between the aureomycin-fed calves and the control calves existed. Therefore, the time of maximum growth improvement and the time when this energy relationship existed are not the same. It is

possible that the effect of aureomycin on blood sugar and rumen fatty acid levels is a secondary one rather than the primary reason why aureomycin improves the growth rate of calves.

The only other clearly demonstrated effect of aureomycin on metabolic processes in the calf was the depressing effect of aureomycin feeding on blood nonprotein nitrogen levels in calves under 7 weeks of age, as reported by Lassiter *et al. (68)*. It was believed that these findings indicated a greater utilization of absorbed nitrogen by aureomycin-fed calves, but these data are too preliminary to draw any conclusions as to the mode of action of antibiotics in the calf. As can be deduced from the foregoing discussion, considerably more information is needed on the effect of antibiotics on the metabolism of the calf before it will be possible to state with any degree of certainty their true mode of action.

GROWING DAIRY ANIMALS

A problem foremost in the minds of dairymen is the question of feeding antibiotics to growing heifers and the long-time effects of feeding antibiotics to dairy animals. Rusoff *(100)* fed Jersey male calves, ranging from 14 to 34 weeks in age, 90-180 mg. of aureomycin daily. A 60% increase in growth occurred for the first 2 weeks after the initiation of aureomycin feeding, followed by increases in growth of 36% after 4 weeks, 30% after 6 weeks, and 8% after 8 weeks. Over the entire 20-week feeding period there was no difference in the growth rate of the control calves and those fed aureomycin. Aureomycin apparently had very little effect on rumination, and no anorexia or diarrhea was observed in the aureomycin-fed calves. Voelker and Cason *(120)* fed 4.8-month-old heifer calves on pasture aureomycin for 8 weeks. During the first 6 weeks of this study the aureomycin-fed calves gained 17% more than the control calves. In another trial heifers were fed 200 mg. daily of crystalline aureomycin plus 2.5% of an aureomycin supplement in the grain ration without any harmful effects. Jacobson *et al. (47)* reported the feeding of aureomycin to calves up to 200 days of age. This experiment was started when calves were 16 weeks old, but half of them had received aureomycin from birth. The group of calves that received aureomycin from birth to 200 days of age were the largest calves at the end of the 28-week trial. Those that received aureomycin from 16 weeks to 200 days of age were the next heaviest animals, followed by the calves that were fed aureomycin from birth to 16 weeks of age; the smallest calves were those that were not fed aureomycin. These workers concluded that previous feeding of aureomycin had some effect on the gain of animals over 16 weeks of age but not so much as the actual feeding of aureomycin from 16 weeks of age to 200 days of age.

Fincham and Voelker *(31)* divided 40 pairs of heifers into two groups and fed one group 80 mg. of aureomycin daily from birth to 200 days of age and 240 mg. daily to about 2½ years of age. They found that most of the growth stimulation from the feeding of aureomycin occurred before the heifers were 6 months old, but the growth advantage obtained was maintained afterwards. At 18 months of age the control heifers weighed 755 lb. and the supplemented heifers, 794 lb. The aureomycin-fed heifers were bred 19 days earlier than the

control heifers but required 1.6 services per conception as compared with 1.4 for the controls. In a later report Jacobson *et al. (49)* fed one heifer of each of 17 pairs 240 mg. of aureomycin daily for a minimum of 15 months prior to parturition and during the first lactation. The average body weight for the control heifers 10 days after calving was 939 lb. and for the aureomycin-fed heifers it was 995 lb. Aureomycin feeding had no effect on milk and butterfat production during the first lactation. The average birth weight of calves born to the control animals was 66 lb. and that of calves born to the aureomycin-fed heifers was 76 lb. There was no difference in the average age of the animals in each group at first calving. The calves from each group of heifers were divided into two comparable groups, one receiving aureomycin to 16 weeks of age. The weight gain of the aureomycin-fed calves was 26% greater than that of the unsupplemented calves and apparently was not affected appreciably by the dams' rations.

Bartley *et al. (5)* reported on the feeding of aureomycin to dairy calves from birth to 13 months of age. Their data concern the period after the calves were 6.6-7.0 months old, although this was a continuation of a previous study. During the next 6 months the control calves gained 233 lb., a 79% increase over the starting weight, while the aureomycin-fed animals gained 237 lb., or a 68% increase over the 6.6 month starting weight. The aureomycin-supplemented calves weighed 57 lb. more when 6.6-7.0 months old than the control calves. These data show that the growth advantage of feeding aureomycin to calves early in life was maintained but there was no further stimulation in growth. It was not known, however, whether it is necessary to feed aureomycin to maintain this early advantage. These data also do not answer the question whether growth stimulation would have been obtained at this older age if aureomycin had not been previously fed. These workers *(6)* reported that calves fed aureomycin from birth to 7 weeks of age increased in weight 199% of their starting weight to 12 weeks of age as compared with 190% for calves not fed aureomycin and 228% for calves fed aureomycin continuously from birth to 12 weeks of age. At 7 weeks of age aureomycin-fed calves and control calves had increased in weight 155% and 138%, respectively, of their starting weights. Huffman *(45)* fed one heifer of a set of anastomose twins aureomycin from 42 days of age to about 19 months of age. At the end of this period the twin fed aureomycin had gained 781 lb. and the control twin 743 lb. The calves consumed about the same amount of TDN during the 18-month feeding period.

Bartley *et al. (9)* fed daily doses of 200-800 mg. of aureomycin per 100 lb. of body weight to calves 12-16 weeks old without any detrimental effects. One calf 16 weeks old that had not previously received aureomycin was fed 2,500 mg. of this antibiotic daily for 4 weeks without any harmful effects.

MATURE DAIRY ANIMALS

The reluctance of research workers to feed antibiotics to mature dairy animals is based primarily on the adverse effects of aureomycin on rumen function, as reported by Bell *et al. (10)* in beef animals and Colby *et al. (24)* in sheep. Wasserman *et al. (124)* conducted in vitro studies on the effect of various anti-

biotics on cellulose digestion in the artificial rumen by rumen microorganisms. These workers found that low concentrations of penicillin (5.0 and 7.5 units per milliliter) stimulated cellulose digestion but that 15 units per milliliter caused an inhibition of the digestion of cellulose. All levels of neomycin (6.25, 12.5, and 25.0 units per milliliter) increased cellulose digestion, but these effects were inversely proportional to the concentrations of neomycin. Streptomycin had no influence on digestion at 12.5 γ per milliliter but decreased digestion at 25.0 or 50.0 γ as well as did chloromycetin at levels of 25.0 and 50.0 units per milliliter.

Lodge *et al. (71)* fed dairy animals 240 mg. of aureomycin daily from an early age to maturity and found that cellulose digestion in the artificial rumen was decreased from 83% for animals not fed aureomycin to 72% for those fed aureomycin. When 80 mg. of aureomycin was fed to 4-month-old calves daily, the digestion of cellulose was reduced to 58%, compared to 78% for the control animals. The addition of 1.6 γ of aureomycin per milliliter to the fermentation mixture when inocula from cows not fed aureomycin were used severely inhibited cellulose digestion. This same addition had little effect on the digestion of cellulose by the inocula from aureomycin-fed cows. Jurtshuk *et al. (53)* found that the presence of both aureomycin and terramycin (100 γ/100 ml.) reduced the ability of rumen bacteria in the "resting state" to utilize the carbohydrates xylose, arabinose, glucose, maltose, and cellobiose in vitro.

Haq *et al. (34, 35)* and Rusoff and Haq *(109)* fed lactating cows aureomycin and observed no significant effect on milk and fat production, milk composition, bacterial count of milk, appetites of the cows, or rumination. In one study *(34)* similar results were observed for tyrothricin. No measurable amounts of the antibiotic were found in the milk. Rusoff *et al. (110)* fed mature dairy bulls 300 mg. of aureomycin daily for 10 weeks without having a significant effect on the number of ejaculations, volume of semen per ejaculate, per cent initial motility, motile sperm per ejaculate, or breeding efficiency as measured by the percentage of 60-90 day nonreturns. Warner *(123)* and Loosli and Warner *(74)* found that the feeding of 700 mg. daily of aureomycin to cows had no effect on feed consumption, but the feeding of 1,000 mg. daily caused feed refusals. No aureomycin was found in the milk of the cows fed 700 mg. of aureomycin daily for 10 days. No effect on cheese starter activity was observed when 500 mg. of aureomycin was fed daily to cows for 6 weeks. The feeding of 100 mg. of aureomycin daily to milking cows produced neither harmful nor beneficial effects on milk production and feed consumption.

Bartley *et al. (4)* observed that the feeding of 32 mg. of aureomycin per 100 lb. of body weight daily to lactating dairy cows had no effect on the general health and well-being of the animals, milk or fat yield, consumption of feed, body weight changes, pulse rate, body temperatures, or rumination. The amount of aureomycin fed ranged from 300 to 500 mg. daily per cow. In a later report *(81)* Kansas workers found that the feeding of this amount of aureomycin to cows had no significant effect on the total plate counts of milk or the amount of acid developed in the milk inoculated with starter. No aureomycin was detected in the milk, and the feeding of aureomycin did not appear to have any effect on the quality

of Cheddar cheese made from the milk. Huffman *(45)* fed as much as 4 g. of bacitracin daily from a bacitracin supplement to lactating cows and as much as 3 g. of aureomycin from Aurofac without affecting the production of milk or the appetites of the cows. Streptomycin also was fed at levels of 1, 2, and 10 g. daily without any adverse effects. Similar results were found with the feeding of neomycin.

Lassiter *et al. (66)* fed yearling dairy steers 500 mg. daily of crystalline aureomycin alone and in combination with a surfactant, Ethomid C/15, to study the effect of aureomycin and surfactants on the digestion of feed nutrients. It was found that crystalline aureomycin decreased dry matter digestibility from 64.0% on the basal ration to 60.5%. This reduction was not statistically significant. Crude fiber digestibility was reduced from 35.5 to 22.7%, and this reduction was significant. When aureomycin and Ethomid C/15 were fed in combination, the depressing effect of aureomycin on dry matter and crude fiber digestion appeared to be lessened. In this study aureomycin had no significant effect on the appetites, consistency of feces, over-all condition of the steers, or digestion of dry matter, crude protein, ether extract, and nitrogen-free extract.

Chance *et al. (18, 19, 20)* reported a series of studies on the effects of aureomycin on rumen digestion and synthesis. Two fistulated dairy steers were fed 0.5 g. daily of crystalline aureomycin for 15 days, followed by a similar feeding period when 1.0 g. of aureomycin was fed daily. The aureomycin feeding periods were preceded by a 15-day period when no aureomycin was fed. Aureomycin had no effect on the appetites of the steers or body weight changes and did not cause digestive disturbances. The rate of the removal of dry matter, crude fiber, crude protein, and nitrogen-free extract from the rumen was highest when 0.5 g. of aureomycin was fed *(19)*. The feeding of 1.0 g. of aureomycin daily caused an accumulation of dry matter, crude protein, and nitrogen-free extract at the 0-hour of feeding, which was the end of one 24-hour period or the beginning of the next 24-hour period. Both levels of aureomycin caused an accumulation of ether extract at the 0-hour of collection.

In a second study *(18)* the effect of aureomycin on the concentration and synthesis of amino acids and B-vitamins in the rumen was studied. The 0.5 g. level of aureomycin feeding produced a lower concentration of amino acids at the 6-hour collection period, which was probably due to the increased rate of removal of protein from the rumen during this period. The evidence of any amino acid synthesis was lacking. The amount of riboflavin in the rumen was reduced when 0.5 g. of aureomycin was fed daily. Both levels of aureomycin appeared to lower the synthesis of nicotinic acid.

In a third study *(20)* the effect of aureomycin on rumen microorganisms with special reference to the streptococci and coliform groups was studied. It was observed that when aureomycin was included in the ration a higher pH existed in the rumen than when the steers were not fed aureomycin. When 0.5 g. of aureomycin was fed daily, an increase in total rumenal bacteria count resulted but not when 1.0 g. of aureomycin was fed daily. Both levels of aureomycin caused a reduction of rumen streptococci, but at the 0.5 g. level of aureomycin

feeding an increase in coliform bacteria occurred in one steer but not in the other. It was noted that at this level of aureomycin feeding the largest amount of nutrients was removed from the rumen. Aureomycin caused an increase in the total bacteria count of fecal material but had no effect on the number of streptococci or coliform groups in the feces. This was believed to be due to the ability of these organisms to adapt themselves quickly to aureomycin, but their metabolism could have been altered. The results of these studies indicated a correlation between the increase in coliform population of the rumen when 0.5 g. of aureomycin was fed and the increased removal of dry matter, crude fiber, crude protein, and nitrogen-free extract from the rumen.

In all of these studies relating to mature dairy cattle it is interesting to note that aureomycin apparently had no pronounced effect on the appetites or the well-being of the animals, whereas the initial data reported with mature ruminants indicated very adverse effects from the feeding of low amounts of aureomycin (200 mg. per day).

DISCUSSION

Considerable discussion has been devoted to the effects of antibiotics on the growth, metabolism, health, and well-being of dairy cattle. Consideration has been given to the relative nutritional merits of the various antibiotics in the light of present-day knowledge. The feeding of antibiotics to dairy cattle appears to be justified only in the rations of young dairy calves, or those under 16 weeks of age. The feeding of antibiotics to older animals does not appear to afford any economic advantage and may possibly have some harmful effects.

When antibiotics are included in the rations of young dairy calves the following changes are likely to occur in the normal life span of the calf: *(a)* increased growth rate as measured by both body weight gains and skeletal growth, *(b)* lower incidence of scours, *(c)* increased feed consumption, particularly of concentrates, at an earlier age, *(d)* improvement in feed efficiency as measured by the pounds of feed required to produce a pound of gain in body weight, and *(e)* improvement in the condition and the well-being of the young calf. The amount of growth response produced by an antibiotic depends upon many factors, but generally the increase in growth rate has ranged from 10 to 30%. A few studies have been reported in which an antibiotic did not improve growth. The only antibiotics that have been studied enough to merit their inclusion in calf feeds are aureomycin and possibly terramycin. Bacitracin and streptomycin appear to have some growth-promoting properties, but insufficient data are available concerning them. Considerably more information is available on the feeding value of aureomycin than of terramycin; consequently, when feeding recommendations are made this difference must be borne in mind. There is seemingly very little difference in the nutritional merits of these two antibiotics; however, as more information becomes available, this statement may need to be qualified.

In this review very little discussion has been given to the economic aspects

of feeding antibiotics to young dairy calves. The question arises whether the added growth advantage can be justified from an economic standpoint. It is the author's opinion that the answer is in the negative when one speaks of added growth alone. Most dairy animals are raised for the sole purpose of herd replacements. The only exception would be calves sold for veal. For herd replacements, available data indicate that any growth advantage produced by antibiotics is small and is of even less importance when the calf grows to maturity. If animals are to be marketed at an early age, less than 6 months, antibiotics would probably then have some economic value strictly from the standpoint of improving body weight gains.

In this discussion considerably more space has been devoted to the effect of antibiotics on the growth rate of calves than to the effect of antibiotics on calfhood diseases, particularly scours. Since most of the reports have stressed the influence of antibiotics on growth more than on scours, this distinction seemed to be justified. Present reports, however, clearly indicate that the true reason for including an antibiotic in a calf feed is not only to improve growth but to reduce the incidence of calf scours. Reports by Ragsdale *et al. (96)*, Wing *(130)*, Weaver *et al. (125)*, Ormiston *(91)*, Davis *(27)* and Lassiter and Seath *(70)* all show that mortality among young dairy calves ranges from 15 to 30% and that calf scours is one of the leading causes of death. This high mortality rate means a tremendous economic loss to dairymen in this country, as well as elsewhere. Since antibiotics apparently reduce the incidence of calf scours, losses can be reduced by their inclusion in calf feeds, thus making an important economic contribution. Based upon foregoing discussions, the effect that antibiotics have on the incidence of scours is the most valid reason for including these products in calf feeds.

Although this field of nutrition has been given considerable attention by research workers, many questions on the feeding of antibiotics to dairy cattle remain unanswered. Information regarding the effect of antibiotics on scours under farm conditions is needed to verify or disprove data collected under experiment station conditions. The long-time experiments in progress need to be continued to study the effects of antibiotics being fed early in the life of the calf or continuously on the later productive life of the cow. Certainly the manner by which antibiotics stimulate growth and reduce scours needs to be clarified.

SUMMARY

An attempt has been made to review the available information on the feeding of antibiotics to dairy cattle as completely and in a manner as unbiased as possible. Many reports used in this review are conflicting; nevertheless, the following conclusions seem to be valid.

1. The antibiotics, aureomycin and terramycin, are the only ones which have been studied sufficiently to warrant valid conclusions. Considerably more research needs to be conducted with terramycin before conclusions concerning its use can be accepted with the same degree of confidence as recommendations for aureomycin.

2. Seemingly no beneficial effects are derived from the feeding of antibiotics to mature dairy cattle.

3. Antibiotics probably improve the growth rate and efficiency of feed utilization of growing dairy animals (over 4 months of age), but unless these animals are to be marketed soon, little economic advantage will result from the feeding of antibiotics.

4. Aureomycin and terramycin stimulate the growth rate of calves from 10 to 30% during the first 16 weeks of age. Most of this growth improvement results before the calves are 8 weeks old. In addition to an improvement in growth, antibiotics appear to reduce the incidence of calf scours, increase feed consumption and feed efficiency, and improve the over-all condition and well-being of the animal.

5. The inclusion of aureomycin or terramycin in the rations of dairy calves seems to be best justified by the beneficial effects of these antibiotics in reducing calf scours and thus calf mortality. Any growth advantage afforded by these antibiotics during the early life of the calf becomes insignificant in a mature animal.

6. Present data indicate that aureomycin should be fed at levels ranging from 15-20 mg. per 100 lb. of body weight daily. There appear to be very few advantages of feeding antibiotics after the calves are 12-16 weeks of age.

7. Two fundamental explanations have been presented regarding the possible mode of action of antibiotics in calves. One of these postulates a stimulation of the pituitary gland and increased production of growth hormone. The other postulation states that antibiotics possibly increase the growth of calves through an alteration in energy metabolism probably involving the microflora of the rumen.

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PROPERTIES OF THE GAMETOKINETIC SUBSTANCE IN THE FECES OF DAIRY CATTLE[1]

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In 1949 Bhaduri and Bardhan *(2)* reported a new pregnancy test for cattle based upon the injection of water extract of feces into the male Indian toad *(Bufo melanostictus)* and subsequent observation of sperm released into the urine of the toad. In further papers by these authors emphasis has been placed upon the importance of establishing the correct feces to water concentration in tests on cattle *(3)* and other ruminant species, such as the buffalo, goat, sheep, and deer *(1)*, in order to correctly distinguish the pregnant animals.

In the work to be reported here, experiments were first undertaken to determine whether the common North American frog *(Rana pipiens)* will respond to the gametokinetic substance discovered by the Indian workers. Inasmuch as little is known about the identity of the active substance, a partial characterization of it has been undertaken.

EXPERIMENTAL PROCEDURE

Urine-free feces were collected from cows or heifers of the Holstein, Jersey, Brown Swiss, and Guernsey breeds in the College Dairy Herd.

Preparation of extracts. In preliminary experiments an aqueous fecal extract was prepared, with slight modifications, by the method of Bhaduri and Bardhan *(3)*. In the procedure adopted after repeated trials with varying feces to water concentrations 20 g. feces was mixed thoroughly with 100 ml. distilled H_2O in a Waring Blendor. The solution was drained through muslin and then filtered through Whatman No. 2 paper. Filtration was very slow, requiring 12 hours to obtain 20 ml. of filtrate. This initial filtrate was clarified by shaking with a small amount of celite and refiltering through No. 40 paper.

Biological assay. Common male leopard frogs *(Rana pipiens)*, weighing 25-45 g., were obtained from a biological supply house. Male frogs can be distinguished from females by the heavy muscular thumb pad in the former. The frogs were stored in the refrigerator at 40° F. and brought into the laboratory 15-30 minutes before use. In each case a specimen of the frog's urine was examined microscopically prior to injection. To date, all specimens examined at this stage have been negative. An initial injection of feces filtrate was made into the dorsal lymph sac with a 22-gauge needle. To avoid leakage, the needle was passed obliquely through the skin and thigh muscle before entering the lymph sac. The frog was then placed in a dry, ventilated jar at room temperature. After 30 minutes a sample of urine was collected, either in a beaker or directly from the

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frog's cloaca by means of a dropping pipette, and examined under a microscope (low power) for the presence of sperm. If no sperm were observed, another 5 ml. of extract was injected and the urine was examined 30 minutes later. If negative at this time, another 5-ml. injection was given and the urine was examined in 30 minutes as before. Exploratory trials on the quantity of extract needed to produce a positive response showed clearly that 10 ml. of extract must be given in the initial dose to get a consistent reaction. When 20 ml. of extract was injected in four equal portions of 5 ml. each, given at 30-minute intervals, only three positive reactions were obtained out of 21 samples of pregnant cow feces that were prepared in a concentration of 20 g. feces to 100 ml. H_2O. The influence of the feces to water concentration on the potency of resulting extracts is shown in Table 1. With 15 g. feces to 100 ml. H_2O no positive responses were obtained

TABLE 1

Influence of feces to water concentration and stage of pregnancy on the gametokinetic response

Gestation period in days	15 g.			20 g.			25 g.		
	No. of tests	No. +[a]	No. −[b]	No. of tests	No. +	No. −	No. of tests	No. +	No. −
35-100				10	9	1			
101-165	3	0	3	15	12	3	2	1	1
166-230	4	0	4	8	5	3	1	1	0
231-275	1	0	1	2	0	2			
Non-pregnant	6	0	6	15	5	10	3	1	2

[a] + Positive response.
[b] − Negative response.

in eight tests from pregnant and six tests from nonpregnant cattle. With 20 g. per 100 ml., 26 tests out of 35 from pregnant cows were positive; in nonpregnant cows, five out of 15 were positive. Only a few samples were tested with a concentration of 25 g. feces per 100 ml. H_2O, but no increase in response was apparent from these results.

In most of the positive reactions a gametokinetic response was observed within 30 minutes after injecting 10 ml. of extract. A few additional responses occurred after each of the added 5-ml. doses. A further examination of Table 1 suggests that the concentration of active substance in feces is the highest during the first 100 days of pregnancy and declines thereafter. However, the data are not extensive enough to clearly establish this point.

Concentration of active filtrates. It was observed that the active substance is readily soluble in mixtures of water and acetone. Such mixtures are filtered and clarified much more easily than an aqueous extract. Two procedures were employed. In the first a positive aqueous extract was obtained as already described and then treated with an equal volume of acetone. In a more rapid procedure, 20 g. of feces was mixed with 100 ml. of distilled water, and 100 ml. of acetone was added. After being thoroughly mixed and allowed to stand for 6 hours, the material was filtered through a Buchner funnel. The acetone was removed by vacuum distillation. The solution remaining was adjusted to 100 ml., mixed with a pinch of celite, and filtered through Whatman No. 40 paper. Such extracts

TABLE 2

Potency of fecal extract from pregnant and open cows at three different concentrations

No. of tests	Extract injected (*ml.*)	Response		Time (*hr.*)	Remarks
		No. +	No. −		
	Initial extract, 100 ml.				
14	10	12	2	½-1½	Pregnant
5	5	1	4	½	Pregnant
8	10	5	3	½	Nonpregnant
7	5	0	7		Nonpregnant
	Conc. to 50 ml.				
10	10	10	0	½-1½	Pregnant
5	5	4	1	1-1½	Pregnant
6	10	5	1	1-1½	Nonpregnant
5	5	1	4	½	Nonpregnant
	Conc. to 25 ml.				
6	5	6	0	½-1	Pregnant
6	2.5	5	1	½-1	Pregnant
4	5	3	1	½-1	Nonpregnant
3	2.5	2	1	½-1	Nonpregnant

were relatively nontoxic to frogs as compared to the earlier extracts. In a similar manner, the extract from 20 g. of feces was concentrated to volumes of 50 and 25 ml., respectively, by vacuum distillation.

Each of the extracts was tested on male frogs as already described. As shown in Table 2, the potency per unit volume of extract increased progressively with decreasing volume. With 10 ml. of initial extract from pregnant cow feces 12 out of 14 tests were positive in ½ to 1½ hours. When this extract was concentrated to 50 and 25 ml., results were obtained with 5.0 and 2.5 ml. of extract, respectively. Somewhat less potency was found in extracts from nonpregnant cows, but the activity was concentrated in the successive preparations just as in the pregnant animals. The fact that little or no loss of activity occurred during the treatments involved indicates that the active principle is quite soluble in water and also comparatively stable.

Solubility in water-acetone mixtures. In view of the high solubility in water of the gametokinetic substance, it was decided to determine whether any of the active substance could be precipitated with increasing concentrations of acetone. To 20 ml. of a potent extract prepared as in the preceding section (threshold dose, 2.5 ml.), acetone was added stepwise in portions of 20, 20, and 40 ml. The precipitate that formed after each addition of acetone was removed by centrifugation, dissolved in distilled water, and tested in a male frog. The precipitates were negative in every test. Finally the acetone was removed by vacuum distillation at 40° C. After filtration of the aqueous solution remaining with the aid of celite, a positive reaction was obtained in the frog with 2.5 ml. In further trials, addition to an aqueous extract of up to 10 volumes of acetone failed to yield an active precipitate. However, when the acetone volume exceeded by more than fivefold the volume of original extract, the activity of the aqueous portion was decreased as much as 50%—by what means is not understood at this time. Al-

though treatment with acetone is helpful in eliminating some foreign matter, no concentration of active substance was obtained.

Solubility in organic solvents. Separate 15-ml. portions of active aqueous extract (threshold dose, 10 ml.) were placed in distillation flasks and evaporated to dryness under vacuum at 40-50° C. In each sample the dried residue was extracted with three portions of the selected solvent (20 ml. total). The solutions were centrifuged, and the undissolved matter was taken up in distilled water for assay (soln. A). Organic solvent was removed from the supernatant solution by vacuum distillation, and the residue was dissolved in distilled water with the aid of a few drops of acetone when necessary to effect complete solution (soln. B). The active substance was found to be soluble in *n*-butanol, but not in diethyl ether, chloroform, acetone, or ethyl alcohol (Table 3). With the four latter solvents the activity was found in the solvent-insoluble residue.

TABLE 3

Solubility of the active dried material in different organic solvents

Solvent	Soln. B Solvent soluble	Soln. A Solvent insoluble
Diethyl ether	−[a]	+
Chloroform	−	+
Acetone	−	+
Ethyl alcohol	−	+
n-Butanol	+[b]	−

[a] − Material inactive, gives negative reaction in frogs.
[b] + Material active, gives positive reaction in frogs.

Heat stability. In the research reported by Bhaduri and Bardhan considerable care was taken to keep feces extracts chilled during processing, although no data were reported on the heat stability or lability of the active material. In experiments undertaken to test this point, 20-ml. samples of active extract were heated at different temperatures for varying periods of time (Table 4). All extracts, even when heated to boiling for 1 hour, retained sufficient activity to give a positive response in frogs. This illustrates clearly that the substance is comparatively stable to heat.

Oven drying. As a further check on its heat stability, 20-ml. portions of an active extract (threshold dose, 5 ml.) were evaporated to dryness in an oven at 94° C. The residue was mixed with 20 ml. distilled water, chilled in the refrigerator for 4 hours, and centrifuged. Only the supernatant fluid was injected.

TABLE 4

Heat stability of the active material

Time heated	No. of tests	Temperature, ° C.		
		65	75	99[a]
15	4	+[b]	+	+
30	4	+	+	+
60	5	+	+	+

[a] Boiling point.
[b] + Positive reaction within 1 hour after injecting into the male frog.

Positive results were obtained in all tests with 10 ml. of extract. With a dose of 5 ml. a positive response was obtained in only half the tests, indicating some loss of activity by exposure to dry heat.

Acid hydrolysis. To 20-ml. portions of an active extract (threshold dose, 2.5 ml.) concentrated HCl was added in amounts equal to 10, 15, and 20% of the volume of extract. The solution was refluxed for 15 minutes, cooled, and neutralized to a pH of 5.0-5.2 by adding 5 *N* NaOH. Crystalline salts were removed by filtration, and the solutions were then placed in cellophane casing and dialyzed against running tap water for 1 hour. The dialysis effected the removal of soluble salts and probably other toxic materials that were invariably lethal to frogs in earlier trials with undialyzed hydrolyzates. Some loss of activity occurred with 10% HCl, as shown by the increase in dosage from 5 to 10 ml. of extract needed to give a response (Table 6), compared to 2.5 ml. of the original extract. Complete loss of activity occurred with addition of 15% and 20% by volume of concentrated HCl.

Dry matter required for gametokinetic response. Inasmuch as all earlier extracts were prepared volumetrically, it was of interest to determine the amount of dry matter required for a response and also the amount of nitrogenous matter involved. For these tests, a series of extracts was prepared from a sample of feces from a pregnant cow as follows:

Extract 1. 20 g. feces in 100 ml. distilled water; prepared as described previously.

Extract 2. 20 g. feces + 100 ml. distilled water + 100 ml. acetone; filtered, acetone distilled off, filtered again, and volume adjusted to 50 ml.

Extract 3. 700 g. feces + 3,500 ml. distilled water + 3,500 ml. acetone; treated as in No. 2. Final volume adjusted to 1,750 ml., boiled for 2 minutes to prevent bacterial growth, and stored in refrigerator for further work.

Extract 4. 800 ml. of extract 3 concentrated under vacuum to 200 ml.; filtered and stored in refrigerator.

Extract 5. 100 ml. of extract 4 mixed with 500 ml. acetone, chilled over night, and centrifuged. The precipitate, being inactive, was discarded. After removal of acetone the volume was adjusted to 100 ml.

The dry matter content of each extract was determined by drying a 2-ml. sample to constant weight at 94° C. Nitrogen content was determined by Nesslerization of a sulfuric acid digest, the final concentration being determined in a Coleman Universal Spectrophotometer. All extracts were tested in male frogs as described previously. The minimum volume of solution needed to stimulate a gametokinetic reaction was determined as shown in Table 6. The gametokinetic action per unit of dry matter administered to the test animals was practically identical in the first four extracts. The small volume of 2.5 ml. of solution required in extract No. 4 is offset by its high dry matter content of 16.0 mg. per milliliter. The final treatment with five volumes of acetone in extract No. 5

TABLE 5
Activity of the substance after evaporating to dryness at 94° C.

No. of tests	Dosage *(ml.)*	No. +[a]	No. −[b]
6	5	3	3
5	10	5	0

[a] + Positive response.
[b] − Negative response.

removed some extraneous matter, as indicated by its reduced dry matter content as compared to No. 4 and also its increased potency on a dry matter basis. The nitrogen content of all preparations was low, ranging from 0.17 to 0.30%. This would represent only 1.1 to 1.9% protein on a dry matter basis even if all of the nitrogen were in protein combination, a point that has not been established.

Tests of extracts in the female rabbit. Inasmuch as all previous tests on the gametokinetic factor in bovine feces were conducted only in amphibia, it was of interest to determine whether a gonadotrophic effect could be produced in a mammal. Since the initial crude extracts were lethal when injected intravenously in rabbits, they could not be tested. However, samples purified as was extract No. 5, described in the preceding section, were well tolerated. In two trials, amounts of 15 and 20 ml. of this extract (six and eight times the effective frog dose) were injected intravenously into isolated virgin doe rabbits. In neither test was there any evidence of ovulation or follicle stimulation 18 to 20 hours after the injection.

Miscellaneous observations. The active substance does not pass through a cellophane membrane. Even when dialyzed against flowing tap water for 48 hours the solution within the dialysis bag has given a positive response. No active substance is precipitated by saturating the solution with ammonium sulfate or by varying the pH. The substance appears to be characterized particularly by its high solubility in water.

DISCUSSION

These results fully confirm the discovery by Bhaduri and Bardhan *(2)* of a gametokinetic substance in bovine feces. Under the conditions of these tests the substance was found in samples from both pregnant and nonpregnant cows, although it was present in highest concentration in the former. Rao and Krish-

TABLE 6
Effect of hydrolysis with varying concentrations of HCl on gametokinetic activity

Vol. per cent of conc. HCl added	Dose *(ml.)*	No. of tests	No. −[a]	No. +[b]
10	5	4	1	3
	10	2	0	2
15	5	3	3	0
	10	4	4	0
20	5	2	2	0
	10	4	4	0

[a] − Negative response.
[b] + Positive response.

TABLE 7

Dry matter and nitrogen content and the minimum quantity of D.M. required for gametokinetic reaction with five different extracts

No. of extract	Dry matter	Nitrogen in D.M.	Dose needed for gametokinetic reaction	
	(mg/ml)	*(%)*	*(ml. soln.)*	*(mg. D.M.)*
1	4.0	0.29	10	40
2	4.1	0.23	10	41
3	4.0	0.30	10	40
4	16.0	0.17	2.5	40
5	10.0	0.23	2.5	25

namurthy *(7)* as well as Bhaduri and Bardhan *(1)* have reported that toads collected in South India (Bangalore) responded to feces extracts from both pregnant and nonpregnant cows. Whether or not there is sufficient difference between pregnant and nonpregnant animals tested under the conditions here described to provide a basis for pregnancy diagnosis cannot be stated from the limited data in this report. A critical test of this question will require more extensive studies, not only at various stages of pregnancy but also during the different stages of the estrous cycle in open cows.

The common male leopard frog *Rana pipiens* responds to the gametokinetic substance as readily as the Indian toad *Bufo melanostictus*. The substance is characterized by its solubility in water and water-acetone mixtures. Its failure to precipitate with acetone or saturated ammonium sulfate, its good heat stability, its resistance to inactivation by HCl, and its failure to induce ovulation in the rabbit differentiate it clearly from the known gonadotrophins.

The active factor also differs from the androgen discovered in bovine feces by Riley and Hammond *(8)* and characterized further by Miller and Turner *(5)*. Fecal androgen is inactivated by drying at temperatures above 45° C. The gametokinetic factor is quite stable to heat in either the moist or dry state. Fecal androgen also appears to differ from the gametokinetic factor in its solubiliy in various solvents. Whereas androgen is extractable from dried feces with methanol, ethanol, butanol, and acetone *(6)*, the gametokinetic factor could be dissolved from the dry state in the present experiments only with butanol or water. However, it is readily extractable from fresh feces by water or water-acetone mixtures. Further work will be required for isolation and complete characterization of the substance involved.

SUMMARY

Studies are reported that confirm the presence in bovine feces of a gametokinetic factor that will induce release of sperm in male amphibia. Male *Rana pipiens* frogs responded in a manner similar to that reported for the Indian toad *Bufo melanostictus*. The active substance was higher in feces of pregnant than of nonpregnant cattle, but its possible validity in pregnancy diagnosis was not evaluated. It is highly soluble in water and water-acetone mixtures. It is also soluble in pure *n*-butanol, but not in diethyl ether, chloroform, acetone, or ethyl

alcohol. It is nondialyzable through a cellophane membrane. Partial inactivation occurred when extracts were dried at 94° C. However, appreciable inactivation did not result from boiling active solutions for as long as 1 hour. Inactivation was complete upon refluxing with 15% and 20% of concentrated HCl added, but was only partial with 10% HCl. Purified extracts failed to induce ovulation when injected into virgin doe rabbits. From the results obtained, it is concluded that the gametokinetic factor differs from the known gonadotrophins and also from the androgen previously described in cattle feces.

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FURTHER STUDIES ON POLYSACCHARIDE PRODUCTION BY BOVINE RUMEN BACTERIA[1]

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In recent years the iodophilic microorganisms of the rumen have attracted the interest of some ruminologists. Oxford *(12)* and Masson and Oxford *(11)* found that holotrich ciliates from the rumen of sheep were active in depositing intracellular polysaccharide granules. These workers have extracted the granules from the cells and found them to be similar to an amylopectin, the main component of which was believed to be glucose. Heald *(9)* has shown that the carbohydrate content of rumen microorganisms increases sharply after feeding, followed by a rapid decline. Elsden *(6)* has demonstrated the same phenomenon after feeding glucose to sheep. Doetsch *et al. (4)* reported that an iodine staining substance, designated ISS, was produced by mixed suspensions of rumen bacteria in vitro from a variety of carbohydrates, including maltose, cellobiose, glucose, and xylose. Finally, Robinson *et al. (17)* have further determined some of the conditions which govern the formation and utilization of ISS using an empirical method of estimation. This group has proposed that ISS formation represents one of several normal processes characteristic to a majority of authentic rumen microorganisms. It was therefore suggested that ISS formation may be used as a criterion for estimating the "normalcy" of the physiological state of the bacterial flora of ruminants. The purpose of the work reported here is to characterize ISS chemically and to note further the effect of some conditions which might occur in the rumen upon the in vitro production of ISS with a more sensitive method of estimation.

EXPERIMENTAL PROCEDURE

Rumen liquor samples were obtained from a 7-year-old Jersey cow fitted with a permanent rumen fistula. The diet consisted of alfalfa hay and a 16% protein grain mixture. Cell suspensions were prepared by filtering rumen liquor through four layers of cheesecloth, the filtrate being centrifuged for 10 minutes at 771 × G. The resulting supernatant liquor was centrifuged at 12,500 × G for 10 minutes, the clear supernatant liquid decanted off, and the cells suspended in M/15 phosphate (pH 6.9) buffer as previously described *(4)*. This buffered suspension was then centrifuged in 15-ml. conical centrifuge tubes for 2 minutes at 1,200 × G. This last centrifuging removed almost all of the residual plant fragments and protozoa, the bacteria remaining in suspension. This suspension was standardized *(4)*, flushed with nitrogen, and incubated at 39° C. for 1½ hours to obtain utilization of stored ISS which might be present in the cells.

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It has been shown that ISS is produced readily from glucose and other carbohydrates by washed suspensions of rumen bacteria. A pH of 6.9 seems optimum, and an atmosphere of carbon dioxide or nitrogen gives equivalent results *(17)*. Therefore, in all experiments reported here, glucose was used exclusively as a substrate in a final concentration of 1 μM per milliliter, and incubated at 39° C. under an oxygen-free nitrogen atmosphere. Dissimilation tests were performed in 50-ml. Erlenmeyer flasks, 3 ml. of the cell suspension being made up to a volume of 10 ml. with substrate, various factors under investigation, and buffer.

Effect of various factors on ISS production. It is now a well recognized phenomenon that rumen bacteria may utilize urea as a source of nitrogen for the formation of protein *(16)*. Smith and Baker *(18)* have further demonstrated that iodophilic organisms increase in number during protein synthesis from urea. These workers have suggested that iodophilic organisms are responsible for the bulk of protein synthesis. The effect of urea on the production of this polysaccharide was studied with concentrations of 100 μM and 1,000 μM per milliliter. Burroughs *et al. (2)*, using the artificial rumen technique, reported an increased utilization of urea and cellulose when a salt mixture was added. Therefore, the effect of Burroughs' salt mixture on ISS synthesis was noted. In this mixture not all of the salts are in solution, and therefore both the entire mixture and the soluble portion were used, 0.5 ml. of each being added to the dissimilation flasks. Robinson *et al. (17)* reported that a chelating agent, sodium versenate (sodium salt of ethylenediamine tetra acetic acid, EDTA), reduced the amount of ISS produced when using an empirical method of determination. This was repeated with a more sensitive colorimetric technique for ISS measurement. Cobalt has been shown to be required by ruminants for the synthesis of vitamin B_{12} *(3, 8)*. In experiments with cobalt-deficient sheep, it has been reported that 1 mg. of cobalt per day is required. Therefore, the effect of cobalt as $CoCl_2 \cdot 6H_2O$ on ISS production was investigated.

To determine the effect of these various substances on the production of ISS, a series of dissimilation flasks was prepared for each concentration of substance tested. An identical control series of flasks was prepared with the exception that the substance in question was omitted. These flasks were incubated for varying periods of time ranging from ½ to 4 hours. At the end of the designated time, the contents of the flasks were removed and analyzed for ISS and residual glucose.

The amount of ISS was measured colorimetrically by placing 5 ml. of the fermentation mixture in a colorimeter tube and adding 2 ml. of a solution of 0.2% ($^{w}/_{v}$) iodine in 2% ($^{w}/_{v}$) potassium iodide. The tubes were inverted to mix, and the optical density of the suspension was read immediately with a Klett-Summerson (model 800-3) photoelectric colorimeter with a red (640 mμ) filter. A blank was prepared in a similar manner with the exception that the substrate was omitted from the dissimilation flask. The instrument was initially set at 100% transmittance in respect to this blank. To determine the effect of various substances upon ISS production, their optical densities were compared with those of the control series. Residual glucose was determined by removing

the cells from the fermentation liquor by Seitz filtration, the filtrate being analyzed for total carbohydrate by using Dreywood's anthrone reagent *(5)*.

Extraction and identification of ISS. The polysaccharide was extracted from the cells in the following manner: 11 l. of rumen liquor were processed in the manner described above and the resulting unstandardized cell suspension was incubated with 14 g. of glucose dissolved in 100 ml. of buffer for 6 hours at 39° C. At the end of this time the mixture was centrifuged at 12,500 × G for 15 minutes, and the supernatant liquor was decanted off and discarded. The resulting cell paste, which colored intensely purple when tested with iodine, was heated at 80° C. for 10 minutes with constant stirring to inactivate enzymes. It was then frozen over night at –20° C. The paste was thawed and portions were ground with an equal volume of alumina (80-200 MM, Fisher Scientific Co.) and combined. This mixture was extracted with 200 ml. of a calcium chloride solution (sp. gr. $^{20}/_{20} = 1.20$) and 4 ml. of 0.8% ($^{v}/_{v}$) acetic acid at 100° C. for 15 minutes. The resulting mixture was centrifuged at 771 × G for 20 minutes, the supernatant liquor being collected. The residue was extracted in a like manner two additional times with 100-ml. portions of calcium chloride, the supernatant liquors being combined. To this liquor sufficient trichloroacetic acid was added to make the final concentration of trichloroacetic acid 10% ($^{w}/_{v}$). This solution was permitted to stand at 4° C. for several hours to assure complete protein precipitation. The mixture was then centrifuged at 12,500 × G for 15 minutes, and the opalescent supernatant solution was placed in cellophane bags and dialyzed against running tap water over night. Dialysis was continued against several changes of distilled water until the water no longer gave a positive test for chloride ions, as determined with silver nitrate solution. The dialysate was then concentrated by vacuum distillation below 30° C. until the volume was reduced to approximately 75 ml. This solution was highly turbid and colored an intense brownish-purple color when tested with iodine. It was evaporated to dryness over calcium chloride in a desiccator at room temperature under vacuum. The resulting light tan-colored residue was scraped free and powdered by grinding in a mortar. The average yield of extract was 1.5 g. Nitrogen content of the dried extracts was determined by the micro-Kjeldahl method.

Fifteen to 20 mg. of the crude extract was hydrolyzed in 1 ml. of *N* H_2SO_4 in a sealed tube at 100° C. for 3 hours. It was neutralized by the addition of solid barium carbonate and centrifuged, the resulting clear supernatant solution being used for chromatographic analysis and for the determination of the per cent total sugar by using anthrone reagent.

Ascending paper chromatography was employed, and the filter paper sheets were made into cylinders and stapled together. Glass cylinders were used, the solvent was placed in the bottom, and the paper cylinders were permitted to rest in the solvent. Whatman No. 1 filter paper was used throughout. The spots were applied with the end of a fine stirring rod; reference sugars were used along with the hydrolyzate in all cases. The solvents used were butanol, acetic acid, water (4:1:5) *(11)*, butanol, pyridine, water (3:2:1.5) *(10)*, and acetone to which 10% water by volume was added *(7)*. The spots were located by spraying

with the aniline hydrogen phthalate reagent of Partridge *(15)* and heated at 105° C. for 5 minutes to bring out the color. Ketoses were tested for by spraying with napthoresorcinol and trichloroacetic acid *(11)*. Both single and double developments were used, double developments being used to obtain good separation of glucose and galactose with the butanol, pyridine, and water solvent.

RESULTS AND DISCUSSION

The results (Figure 1) show that 100 μM per milliliter of urea do not alter the formation of ISS but hasten its utilization, once produced. Glucose was utilized more rapidly than in the control, and this agrees with the results of Arias *et al. (1)* using the artificial rumen technique, who found that the presence

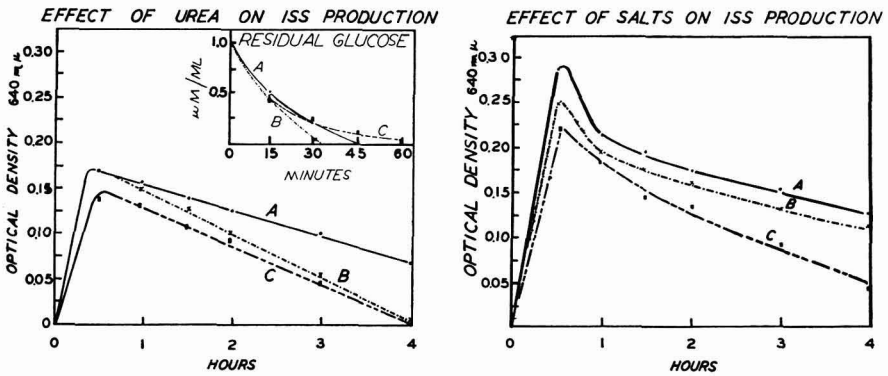

FIG. 1. Effect of urea on ISS production. A—control; B—100 μM urea per ml.; C—1000 μM urea per ml.

FIG. 2 (right). Effect of salts on ISS production. A—control; B—0.5 ml. of entire salt mixture; C—0.5 ml. of soluble portion.

of starch or glucose increased protein synthesis from urea. It appears that a demand for carbohydrate is placed upon the bacteria when converting urea to protein. With 1,000 μM per milliliter of urea, ISS formation was retarded and utilization increased as with the lower urea concentration. The utilization of glucose, however, appeared to be retarded. An explanation of this may be that with high concentrations of urea, cell permeability is affected in some way. However, protein synthesis still requires energy, and the reserve energy available in ISS is called upon as with lower concentrations of urea.

It was found that the presence of Burroughs' salt mixture decreased the amount of polysaccharide produced, the soluble fraction exerting a greater effect than the entire mixture (Figure 2). The utilization of the glucose substrate was found not to be altered. Whether these salts are stimulating the carbohydrate metabolism of the bacteria or are inhibiting enzymatic reactions necessary for ISS formation has not been determined. As the utilization of the polysaccharide itself does not appear to be markedly accelerated, it seems that the latter view is reasonable.

Sodium versenate (EDTA) concentrations of 0.5 μM and 2.0 μM per milliliter reduce ISS synthesis (Figure 3). Corresponding to this decrease, the utilization of the glucose substrate also appears to be retarded. Whether this effect is due solely to removal of essential metal ions or to the toxicity of EDTA per se has not been determined.

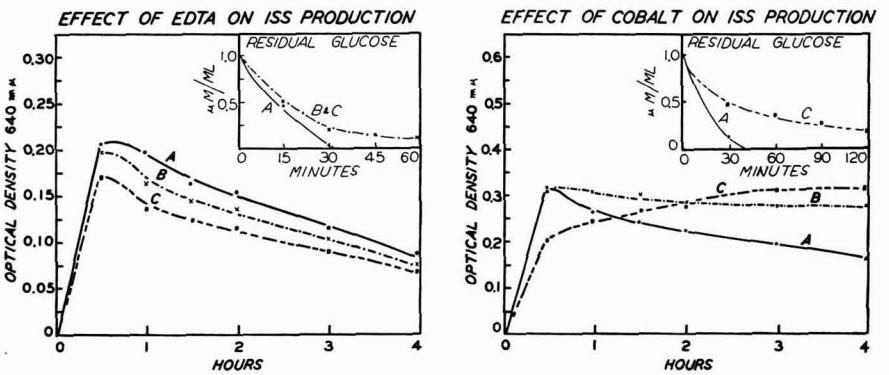

FIG. 3. Effect of EDTA on ISS production. A—control; B—0.5 μM EDTA per ml.; C—2.0 μM EDTA per ml.

FIG. 4 (right). Effect of cobalt on ISS production. A—control; B—0.5 or 2.0 μM cobalt per ml.; C—2.0 μM cobalt per ml. added 30 minutes previous to substrate.

In a concentration of 0.05 μM per milliliter, cobalt did not affect polysaccharide synthesis or glucose utilization. However, in concentrations of 0.5 μM and 2.0 μM, synthesis of ISS equaled that of the control, but utilization was markedly retarded (Figure 4). With these concentrations of cobalt, glucose utilization did not appear to be altered. As the cobalt seemed to exhibit its effect after being in contact with the bacteria for a period of half an hour, cells in a cobalt concentration of 2.0 μM per milliliter were incubated at 39° C. for 30 minutes before the addition of the substrate. In this instance the production of ISS was greatly retarded, as was the utilization of glucose. The level of ISS synthesis did reach that of the control after 4 hours incubation, however. Thus, the presence of cobalt in high concentrations retards polysaccharide synthesis and utilization as well as the utilization of the substrate itself. However, in low concentrations such as might normally be found in the rumen, cobalt was ineffective.

The extracted ISS was found to contain 1.95% nitrogen, and, upon hydrolysis, 78% carbohydrate as glucose. It was sparingly soluble in water and colored brownish-purple when tested with iodine. Chromatography of the hydrolyzate resulted in a single spot. When the *n*-butanol, acetic acid, water solvent with reference sugars was used (Figure 5), the Rf value of this component was found to be that of D-glucose. However, with this solvent good differentiation between D-glucose and D-galactose was not obtained. Therefore, double development employing the *n*-butanol, pyridine, water solvent was used, as suggested by Jeanes *et al. (10)*. ISS is believed to be a polymer composed solely of D-glucose, but

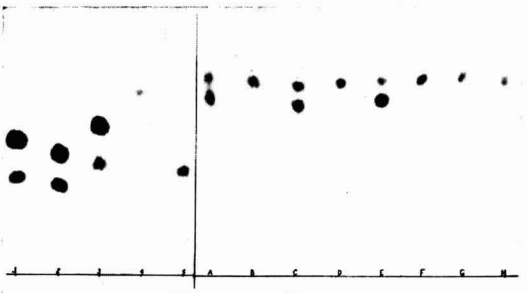

FIG. 5. Chromatograms of ISS hydrolyzates. Known reference carbohydrates: 1—xylose plus glucose; 2—arabinose plus galactose; 3—mannose plus ribose; 4—rhamnose plus fructose (the latter too light to be seen here); 5—ISS hydrolyzate. Solvent system butanol, acetic acid and water. A—glucose plus galactose; B—ISS hydrolyzate (from glucose) plus glucose; C—ISS hydrolyzate (from glucose) plus galactose; D—ISS hydrolyzate (from xylose) plus glucose; E—ISS hydrolyzate (from xylose) plus galactose; F—ISS hydrolyzates from glucose and xylose combined; G—ISS hydrolyzate from glucose; H—ISS hydrolyzate from xylose. Solvent system butanol, pyridine and water.

since the substrate used to produce this polysaccharide was D-glucose, it was considered important to determine what effect a different carbohydrate substrate would have on its composition. Therefore, D-xylose was used and the resulting polysaccharide was extracted as previously described. This extract was found to contain 2.43% nitrogen and 74% carbohydrate determined as glucose. When tested with iodine, it produced a color similar to that of the extract from glucose. Chromatography of the hydrolyzed polysaccharide revealed a single component, which was also identified as D-glucose. Thus, it appears that the same polysaccharide is produced by the bacteria regardless of whether a 5C or 6C carbohydrate is used as substrate. This reaction may be considered one of many which demonstrate the metabolic versatility of rumen bacteria in respect to their ability to utilize a wide variety of substrates and yet produce similar end-products. This is probably the result of the interaction of a large number of metabolic systems such as one finds in the rumen. If this contention is correct, the ISS reaction supports the hypothesis that the rumen microflora is not subject to radical physiological changes by alterations in the animal's diet, unless these are unusual or unnatural.

It is believed that this polysaccharide is of the starch-glycogen class, but further work must be done to prove or disprove this. Oxford *(13)* has extracted a polysaccharide from pure cultures of rumen streptococci and believes it to be of the starch-glycogen class. It would therefore appear that these polysaccharides are identical. Experiments to further purify these extracts and to characterize the physico-chemical properties of this polysaccharide are under way.

The production of ISS may be considered to be a reaction characteristic of a large number of authentic rumen bacteria. It is believed that the reaction may be used for estimating the physiological normalcy of the rumen microflora, as Robinson *et al. (17)* have proposed. The reaction may be significant to the

ruminant in that it enables the microorganisms most active in cellulose digestion and protein synthesis to store a readily available source of energy from the soluble carbohydrate portion of the diet. If it is admitted that the cellulolytic bacteria are among the most important in the rumen, then a reaction which enables them to store a readily available energy source assumes that these organisms will be able to attack cellulose and synthesize protein using ISS when the soluble carbohydrate in the feed is depleted. The further possibility exists that the amount of readily available carbohydrate in a feed may be evaluated by using the degree of polysaccharide synthesis by these organisms as an index. This would provide a rapid in vivo means for comparing various feeds in regard to supplying available carbohydrate to rumen bacteria. Further work along this line should yield profitable results.

SUMMARY

ISS produced by washed suspensions of rumen bacteria has been extracted and found to be a polymer of glucose. Identical polysaccharides were produced when either a pentose or hexose was used as substrate. The influence of urea, Burroughs' salt mixture, a chelating agent, and cobalt upon the synthesis of ISS was studied by using a colorimetric method for determining ISS. Possible significance of this reaction and suggestions for further work are discussed.

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SELECTION INDICES BASED ON MILK AND FAT YIELD, FAT PER CENT, AND TYPE CLASSIFICATION[1]

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Genetic improvement in several traits can be most effectively accomplished if the information about those traits is combined into an index of net merit or expressed as a total score *(7)*. The application of this principle to selection for fat yield and type in dairy cattle was recently demonstrated by Harvey and Lush *(5)*. However, the effectiveness of selection indices which include milk yield, fat yield, fat per cent, and type has not been established. Therefore, the objectives of this study were to obtain the necessary parameters and suitable economic values to construct several selection indices involving milk and fat yield, fat per cent, and type to determine how effectively these indices would indicate the relative breeding worth of individual cows and subsequently to determine the expected genetic improvement per generation of selection.

SOURCE AND DESCRIPTION OF DATA

The data for this study were provided by the American Jersey Cattle Club. They included the Herd Improvement Registry lactation records of all Jersey cows that had completed records some time during the years 1947 through 1950 and the official type classification of the cows that had been rated under the Jersey Herd Classification program. The type classifications were not necessarily made during the years represented by the production data. From the cows that had both a production record and a type classification, 2,810 daughter-dam pairs were obtained from 414 herds. The daughters were the progeny of 756 sires with each sire having at least two daughter-dam pairs. The dam and her daughter(s) were included only if their records were made in the same herd. There were 196 two-or-more sire herds which had 2,146 daughter-dam pairs, with nearly 42% of this total being in herds having ten or more pairs. The sires with four or more daughter-dam pairs made up 58.6% of the total sample size.

The first single lactation records available in these data were used and consisted of milk yield, fat yield, and fat per cent with the milk and fat yield adjusted to a mature-equivalent, 305-day, twice-a-day milking basis. The fat per cents were converted from percentage units into decimal fractions. For example, a fat per cent of 5.3 is equal to 0.053 as a decimal fraction. No record of less than 270 days or more than 305 days was included.

The type ratings were converted to a numerical score by assigning numbers to the ratings beginning with Fair = 2, Good = 3, Good Plus = 4, Very Good =

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[1] The data in this paper are from a thesis presented by the senior author in partial fulfillment of the requirements of the Ph.D. degree, University of Illinois.

[2] Agent of the Dairy Husbandry Research Branch, USDA.

5, and Excellent = 6. In these data there were no cows with "Poor" ratings as the registration is automatically canceled on such animals.

The means of the milk yield, fat yield, fat per cent, and type of the dams and their daughters are given in Table 1. The means for the dams and daughters were nearly equal in all cases. The intra-sire standard deviations in Table 1 indicate that the daughters were less variable in their production than the dams even though the dams are presumed to be a more selected group.

TABLE 1

Means and intra-sire standard deviations of milk yield, fat yield, fat per cent, and type of the dams and their daughters

	Means		Standard deviations	
	Dams	Daughters	Dams	Daughters
Milk yield *(lb.)*	8,733	8,600	1,585	1,419
Fat yield *(lb.)*	462	461	83	72
Fat *(%)*	5.31	5.38	0.45	0.43
Type *(scores)*	4.43	4.18	0.66	0.66

ANALYSIS AND DISCUSSION OF DATA AND RESULTS

Heritability. The procedure of doubling the intra-sire regression of daughter on dam was used to estimate the heritabilities. The heritable differences in single records of milk yield, fat per cent, and fat yield were 0.25, 0.56, and 0.20, respectively. The estimates for milk and fat yield are smaller than those reported by Tyler and Hyatt *(13)*, but they are consistent in that milk yield has a higher heritability than fat yield. The heritability of fat yield of 0.20 is the same as the intra-herd estimate, also for Jerseys, recently reported by Legates and Lush *(11)*.

The intra-sire heritability of type classification was 0.25. Harvey and Lush *(5)* obtained an intra-herd heritability of type of 0.14. This difference of 0.11 has a standard error of 0.064. The probability that such a difference is due to sampling error is less than 0.10 and is approaching significance. Harvey and Lush suggested that their estimate might have been an indication of the importance of judge differences since in the herds classified more than once, there were some pairs in which one judge classified the daughter and a different judge at a different time had classified the dam. On the other hand, Tyler and Hyatt *(14)* found a heritability of 0.28 in Ayrshire data from an intra-sire regression of daughter on dam where all pairs within any sire had been classified by the same judge on the same day.

Phenotypic and genetic correlations. The intra-sire phenotypic and genetic correlations between characters on the same individual are listed in Table 2 for milk yield, fat yield, fat per cent, and type. The phenotypic correlations were estimated from the daughters' records. All of the phenotypic correlations except the one between fat per cent and type are highly significant. The standard errors of all of these phenotypic correlations are of the order of 0.026, or less.

The phenotypic correlation between fat yield and type is approximately the same as that found by Harvey and Lush *(5)* but somewhat smaller than that

TABLE 2
Intra-sire phenotypic and genetic correlations between milk yield, fat per cent, fat yield, and type

	Phenotypic	Genetic
Milk yield and fat per cent	−0.36	−0.50
Milk yield and fat yield	0.88	0.72
Milk yield and type	0.08	0.07
Fat per cent and fat yield	0.15	0.20
Fat per cent and type	0.05	−0.01
Fat yield and type	0.11	0.08

reported by Tyler and Hyatt *(14)*. The small size of the phenotypic correlation between fat per cent and type indicates that these traits are virtually independent.

When comparing the phenotypic correlations found in the present study with those found in other studies, the largest differences are those involving the correlation between milk yield and fat per cent and the correlation between fat yield and fat per cent. In studies involving samples of records from several breeds, investigators *(4, 12)* reported a correlation between milk yield and fat per cent ranging from −0.114 to −0.198 in the Holstein and Ayrshire breeds and from −0.295 to −0.354 in the Jersey and Guernsey breeds. It could be that the difference between the estimates of the correlation between milk yield and fat per cent obtained in the present analysis and the −0.14 reported by Tyler and Hyatt *(13)* is an inherent breed difference and not necessarily a consequence of sampling error.

The method of calculating genetic correlations as outlined by Hazel *(6)* was followed in estimating those in this study. The genetic correlations between milk yield and type and between fat yield and type in Table 2 indicate that milk yield and fat yield have about the same genetic relationship to type. The intra-herd estimates of these two genetic correlations were both 0.14, which is nearly twice as large as the intra-sire ones. The differences could be sampling error, but they also could indicate that the breeders tended to stratify the dams according to their type and then mated the dams in each of these groups to a different sire, as a large number of the herds in this study were using two or more sires concurrently. Another possibility is that the ages at which the dams and daughters were rated may have differed considerably. Many more of the dams than daughters would have had a chance to have their classification raised and, secondly, the judges are likely to be more conservative when classifying young cows. The intra-herd genetic correlation of 0.14 between fat yield and type is comparable to the 0.18 obtained by Harvey and Lush *(5)*.

The genetic correlations obtained in the present study are in rather close agreement with those of Tyler and Hyatt *(13)* except for the genetic correlation between milk yield and fat per cent of −0.50, which is considerably larger than the −0.20 they reported. The reason for this difference is not readily apparent, but they studied data from the Ayrshire breed, whereas the present data came from the Jersey breed. Farthing and Legates *(2)* found a genetic correlation

between milk yield and fat per cent of -0.57 for Guernseys and -0.38 for Holsteins, which brackets the corresponding corrrlation of –0.50 found in this studv. Thus, the more recent results indicate that the genetic correlation betwecn l1Iiik yield and fat per cent may be higher negatively than was reported earlier by Tyler ancl Hyatt *(13)*. The work of Farthing and Legates with Guernsey and Holsteill data appears to confirm the earlier contention of Gaines (3) that the higher-testing breeds should have a higher negative relationship between milk yield and fat pel' cent. This does not appear to be true within a breed, for when Farthing and Legates stt'atified their data for each breed into ten groups 011 the basis of the average fat per cent of the herd there was no clear indication of any trend in the negative relationship of the genetic co)']'elation as the herd level of fat pel' cent increased.

Relative economic I'n/IIcs. The economic values to be assigned to milk yield, fat prl' ernt, fat yield, and type elassificatioll in a selection index are likely to vary with clitfel'rnt herds of cattle. Commercial dairymen will depend almost wholly on the receipts from the sale of milk 01' butterfat, whereas some breeders receive an additional income fl'om tbe sale of their cattle fol' breeding purposes and from show ring advertisement aml prizes.

An estimate of the relative values of milk and butterfat was obtained from the 1945-52 averages of \·early pl'ices paid to milk producers in the Chicago fluid milk marketing area and the Boston cream marketing area. The average blend price per pound of Grade A–Grade B milk testing 5.3% \\'as $0.049. The fat test of 5.3% is the average fat per cent of the Jersey milk ill these data. Since there were no figures available for the price of butterfat marketrd as cream on the Chicago market, the Boston price of $0.869 per pound of butterfat was used. This is based on the price of 40% cream which was intended for butter less the cost of refrigerated transpOJ·tation from Chicago to Boston. Fl'om these prices the a.verage 11umber of pounds of 5.3% milk necessary to equal the price of a pound of butterfat was 17.57. Fat per cent was given an economic value of zero in all of the indices.

The economic value of a.n increase of one type rlassification will depend largely upon the individual dairyman; however, the two extremes of giving type a value of zero anel the cquivalent of a standard deviation in milk or fat will illustrate the influence that paying attrntion to type has on the rate of exprcted progress in increasing milk and fat yield. The assumed economic value for type for the latter extreme, in pounds of milk reqnire(l to equal a change of one grade in type, was 2,]39.3 as determined from the ratio of the intra-sire stanllar(l deviations of milk and type.

The il/dex. The prinriple of selection by means of an index as developed b.v Hazel (6) was followed in deriving the indicrs. Some mOllifications of this method as suggested by Henderson *(8)* and includrd in recent papers by Karam *et al. (10)* and Bernard *et nl*. (1) were also used.

The aggrrgate genotype (H) of an individual is designatr(l here as

$$H = a_M G_M + a_f G_f + a_F G_F + a_T G_T$$

where G_M, G_f, G_F, and G_T are the genic values for milk yield, fat per cent, fat yield, and type classification, respectively, and the a_i's are the relative economic values of these four traits.

Henderson *(8)* has proposed that an index of each genotype be calculated from the phenotypic measures of all of the traits under consideration. On this basis, the best estimate of the breeding value of an individual may be defined in this study as

$$I = a_M I_M + a_f I_f + a_F I_F + a_T I_T$$

where I_M, I_f, I_F, and I_T are indices for estimating G_M, G_f, G_F, and G_T from the phenotypic measurements, X_M, X_f, X_F, and X_T of the four traits, milk yield, fat per cent, fat yield, and type classification, respectively.

I_M, I_f, I_F, and I_T are further defined to be:

$$\begin{aligned}
I_M &= b_{11} X_M + b_{12} X_f + b_{13} X_F + b_{14} X_T \\
I_f &= b_{21} X_M + b_{22} X_f + b_{23} X_F + b_{24} X_T \\
I_F &= b_{31} X_M + b_{32} X_f + b_{33} X_F + b_{34} X_T \\
I_T &= b_{41} X_M + b_{42} X_f + b_{43} X_F + b_{44} X_T
\end{aligned}$$

where the b's are regression coefficients which were derived by maximizing $B\sigma I_i$ of the following equation:

$$E(G_i - \overline{G_i}) = B(I_i - \overline{I_i}) = z/p\ B\sigma I_i$$

$E(G_i - \overline{G_i})$ is the expected genetic gain in the i-th trait, B is the regression of the i-th genic value G_i on the i-th index I_i. The height of the ordinate on the normal curve at the point of truncation is represented by z, and p is the fraction of the population saved. Calculating the b's in this manner maximizes the correlation between G_i and I_i as well as the correlation between H and I *(9)*.

The system of equations from which b_{11}, b_{12}, b_{13}, and b_{14} were obtained is

$$\begin{aligned}
b_{11}\sigma^2 X_M &+ b_{12}\sigma X_M X_f + b_{13}\sigma X_M X_F + b_{14}\sigma X_M X_T = \sigma^2 G_M \\
b_{11}\sigma X_f X_M &+ b_{12}\sigma^2 X_f + b_{13}\sigma X_f X_F + b_{14}\sigma X_f X_T = \sigma G_f G_M \\
b_{11}\sigma X_F X_M &+ b_{12}\sigma X_F X_f + b_{13}\sigma^2 X_F + b_{14}\sigma X_F X_T = \sigma G_F G_M \\
b_{11}\sigma X_T X_M &+ b_{12}\sigma X_T X_f + b_{13}\sigma X_T X_F + b_{14}\sigma^2 X_T = \sigma G_T G_M
\end{aligned}$$

The phenotypic variances and covariances on the left represent those of the records on the daughters as presented in Table 3 and are the same for estimating all four sets of the b's. The right members of this system of equations are the appropriate genetic variance and covariances given in Table 4.

TABLE 3

Phenotypic variances and covariances of milk yield, fat per cent, fat yield, and type classification

	Milk yield	Fat per cent	Fat yield	Type
Milk yield	2,014,770.0	−2.19827	90,003.3	78.103
Fat per cent		0.0000183	0.04665	0.00014
Fat yield			5246.2	5.325
Type				.440

TABLE 4
Genetic variances and covariances of milk yield, fat per cent, fat yield, and type classification

	Milk yield	Fat per cent	Fat yield	Type
Milk yield	495,968.7	−1.13228	16,439.0	17.081
Fat per cent		0.0000102	0.02037	−0.00001
Fat yield			1,051.656	0.828
Type				0.111

The amount of genetic progress to be expected from the selections based on a particular index is proportional to r_{IH}. When selecting for one trait, the correlation, r_{IH}, serves as a basis for determining whether or not there is any additional information provided by traits correlated with the desired trait. When selection is practiced on more than one trait, r_{IH} provides a basis for choosing the index which is expected to provide the greatest amount of total genetic progress toward the desired aggregate genotype.

When selecting according to indices the expected genetic change for each of the different traits can be determined for each index. The expected genetic change in any one of the traits included in the index is

$$E(G_i - \overline{G}_i) = \frac{z/p \; \Sigma_j c_j \, \sigma G_i G_j}{\sigma I}$$

where z is the height of the ordinate of the normal curve at the point of truncation and p represents the fraction of the population saved for breeding. The c_j's are regression coefficients obtained by collecting the appropriate $a_i b_{ij}$ terms. The $\sigma G_i G_j$'s are the genetic covariances between the i-th trait and each of the j-th traits except when $i = j$, in which case $\sigma G_i G_j$ is the genetic variance of the i-th trait. σI is the standard deviation of the estimated breeding value. The above formula is the same as that used by Karam *(9)*.

Several selection indices were calculated from the phenotypic and genetic variances and covariances in Tables 3 and 4. Five of these are listed in Table 5 along with r_{IH} and the expected genetic changes per generation of female selection.

The first two indices, I_1 and I_2, are designed to estimate the genotype for milk yield. In I_1 the genotype for milk yield is estimated from milk yield whereas in I_2 it is estimated from milk and fat yield. The correlation, $r_{IH} = 0.496$ for I_1 indicates that if milk yield alone is used as an index the genic value of the milk yield of an individual can be estimated about half as accurately as it would be if the genetic constitution of each animal were completely known. This can be increased by slightly more than 10% with the addition of fat yield to the index as demonstrated by I_2 in Table 5. An increase of 13 lb. in the expected genetic improvement of milk results in a corresponding reduction of 1.7 lb. in fat yield. An index in which milk yield, fat yield, and fat per cent were included was calculated, but the resulting linear dependence reduced considerably the predictive value of the index. On the other hand, an index which included only milk yield and fat per cent gave about the same results as I_2.

TABLE 5

The indices, the r_{IH}'s, and the expected genetic changes per generation of female selection in milk, fat per cent, fat, and type

Index	r_{IH}	Aggregate genotype	Expected genetic change[a]				Milk equivalent[b]
			Milk	%	Fat	Type	
$I_1 = X_M$	0.496	*M*	122	−0.03	4.1	0.004	194.2
$I_2 = 0.46X_M - 4.7X_F$	0.548	*M*	135	−0.06	2.4	0.004	177.2
$I_3 = X_F$	0.448	*F*	79	0.01	5.1	0.004	168.8
$I_4 = 0.42X_M - 0.81X_F + 546X_T$	0.477	*MFT*	106	−0.02	3.4	0.040	165.8
$I_5 = 0.39X_M + 542X_T$	0.476	*MFT*	103	−0.02	3.5	0.040	165.8

[a] Assuming that 20% of the females are culled on the basis of their index value.
[b] Milk + 17.6 fat.

In I_3 fat yield is used to estimate the genotype for fat yield. A comparison of the r_{IH}'s for I_2 and I_3 shows that the breeding value of a cow for milk yield can be estimated 22% more accurately than her breeding value for fat yield. The expected genetic change in fat yield for I_3 is 5.1 lb. as compared with 4.1 lb. for I_1. However, the increase of 1 lb. in the expected genetic gain for fat yield is accompanied by a reduction of 43 lb. in the expected genetic gain in milk yield. These results indicate that Jersey breeders who select their cows on the basis of milk yield alone are improving the genic value of these cows for fat yield 80% as fast as they would if they paid attention only to fat yield, but in the reverse situation, those breeders who pay attention only to fat yield as a basis for selection are improving the genic value of milk yield only 65% as fast as they would if they were selecting their cows on the basis of milk yield. Whether or not the relative differences in the expected genetic changes for milk and fat yield in I_1 and I_3 are typical of the other breeds, particularly the lower testing breeds, is a matter to be investigated.

If milk yield, fat yield, and type are being selected for and are considered to be of equal economic importance, index I_5, which includes only the phenotypic measures of milk yield (X_M) and type (X_T), is sufficient to depict the merit of such an aggregate genotype. This is evident from the r_{IH}'s and the expected genetic gains for I_4 and I_5. The use of either index I_4 or I_5 may be expected to result in a slight additional increase in the genetic improvement of type but at a sacrifice of 15% in the genetic change of milk and fat yield. Therefore, these values indicate that very little genetic improvement in type can be expected from female selection without at the same time reducing the rate of improvement in production.

The magnitude of the expected genetic change in fat per cent was slight in all of the indices and tended to be in the negative direction. There was an increase of 0.01 in I_3 where emphasis was on fat yield.

In order to compare the rate of improvement in production for the different indices, the expected genetic change in fat was converted to a milk equivalent by multiplying it by 17.6 and adding it to that of milk. The factor of 17.6 is the economic equivalent in pounds of 5.3% milk of a pound of butterfat. These milk equivalents for the indices are given in the last column of Table 5.

The expected genetic changes in Table 5 are those that would come about if

selection were on the female side only. Selection among the sires can be more intense and this makes it possible to increase the level of performance considerably. For example, if only sons of the 5% of the dams with the highest index values are saved for breeding purposes, the contribution of the male selection to the net rate of improvement expected per generation would be 408 lb. milk and 17.1 lb. fat.

The indices in Table 5 were based on parameters derived from single records, and their use is limited to the information provided by a single record. The increased accuracy of prediction of an index when provisions are made for taking into account the variation in the number of fat records available on the individual being indexed as well as various close relatives has been demonstrated by Harvey and Lush *(5)* and by Legates and Lush *(11)*. Thus, the indices which they developed for selecting for fat yield may be more useful than those presented in Table 5. However, it appears from the results of the present study that milk yield is a better criterion of selection for the genetic improvement of production than is fat yield.

SUMMARY

The necessary statistics for deriving several selection indices were obtained from the lactation and type records of 2,810 daughters and mates of 756 sires from 414 Jersey herds. The heritability of differences in single records of milk yield, fat yield, fat per cent, and type were 0.25, 0.20, 0.56, and 0.25, respectively. The intra-sire phenotypic correlations were –0.36 between milk yield and fat per cent, 0.88 between milk yield and fat yield, 0.08 between milk yield and type, 0.15 between fat per cent and fat yield, 0.05 between fat per cent and type, and 0.11 between fat yield and type. The corresponding genetic correlations were –0.50, 0.72, 0.07, 0.20, –0.01, and 0.08, respectively.

The genic value for milk yield can be estimated 10% more accurately by also taking into consideration the cow's fat yield, while fat yield alone is a good criterion of selection for improving the genic value of fat yield. The expected genetic improvement of milk and fat yield was greatest when milk yield alone was the basis of selection. Selection for type along with milk and fat yield resulted in a 15% decrease in the expected genetic gain of milk and fat yield.

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CORNCOBS WITH AUREOMYCIN AS ROUGHAGE COMPARED TO HAY FOR DAIRY HEIFERS

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It has been demonstrated previously *(1)* that corncobs can be utilized by beef cattle to supply part of the digestible energy needed for growth and fattening. In view of the similarity in the digestive systems of beef and dairy cattle and the relatively low nutrient requirement for growth compared to lactation one might assume that large amounts of corncobs could be used for feeding dairy heifers. In the corn belt states cobs are generally plentiful. In using cobs for feed the main costs are for hauling and grinding, which may range from $5 to $10 per ton.

Perry *et al. (4)* reported an increased rate of gain in growing steers when aureomycin supplemented a corncob and Purdue supplement A ration. The feed intakes were equalized for the two lots of steers, and one lot received 75 mg. aureomycin daily per steer. The steers receiving aureomycin gained more weight and utilized feed more efficiently than the controls. Neumann *et al. (3)* observed no benefit from feeding 2 mg. of aureomycin per pound of dry feed to yearling heifers. Reid *et al. (5)* in a review on aureomycin state that "aureomycin stimulates growth in calves up to 6 months but it has little or no effect thereafter."

This paper reports the results obtained when corncobs and supplements replaced the hay normally fed to dairy heifers. Aureomycin[1] (Aurofac 2A) was added to the rations to determine whether or not larger and more efficient gains would be obtained.

EXPERIMENTAL PROCEDURE

Twelve growing dairy heifers ranging from 6 to 9 months of age were assigned to four ration groups so that each group was made up of three heifers—two Holstein and one Brown Swiss. The half sisters (sired by one bull) were distributed throughout the groups. There were two sets of four half-sisters, and one sister of each set was assigned to each ration group. The average initial live weights were 542, 505, 524, and 500 lb. for groups 1, 2, 3, and 4, respectively.

Each group of heifers was fed its experimental ration for the duration of the experiment without change-over. Complete individual feed records were maintained. Each heifer was fed as much corncob mixture or hay as she would consume without excessive refusal. The amount of concentrate mixture fed was the same for all heifers. The four rations fed were as follows:

1. Corncob mixture No. 45 and concentrate mixture No. 43.
2. Corncob mixture No. 45 and concentrate mixture No. 43 with 18 mg. aureomycin added per pound.

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[1] Aurofac 2A was supplied by R. F. Elliott, Lederle Laboratories Division, American Cyanamid Company, Pearl River, N. Y.

3. Bromegrass hay and concentrate mixture No. 43 with 18 mg. aureomycin added per pound.
4. Bromegrass hay and concentrate mixture No. 43.

With this experimental design, the six heifers being fed corncobs were compared to the six receiving hay, and the six heifers being fed aureomycin were compared to the six not receiving aureomycin.

The feed mixtures were composed of the following ingredients:

Corncob mixture No. 45	*(lb.)*	*Concentrate mixture No. 43*	*(lb.)*
Ground corncobs	1,500	Corn meal	400
Soybean oil meal	450	Ground oats	400
Steamed bonemeal	30	Soybean oil meal	170
Trace-mineralized salt	15	Trace-mineralized salt	18
Vitamin A-D concentrate	5	Steamed bonemeal	10
		Vitamin A-D concentrate	2
		Aureomycin	[a]

[a] Aurofac 2A was used to supply 18 mg. aureomycin per pound of grain for rations 2 and 3 but was not included for rations 1 and 4.

The average daily intake of aureomycin was 81 mg. per heifer—90 mg. during the first 12 weeks and 72 mg. during the last 12 weeks. The vitamin A concentrate supplied more than 75,000 I. U. of vitamin A daily per heifer.

The corncob mixture was formulated so that it would be essentially equivalent to bromegrass hay in protein content and would supply adequate amounts of calcium and phosphorus, according to Morrison's *(2)* feed composition values. This procedure equalizes the nutritive values for purposes of making economic comparisons of the rations.

The bromegrass hay for rations 3 and 4 contained a very small proportion of alfalfa. It was harvested at a medium stage of maturity and was medium in quality with only slight weather damage.

The heifers were housed in a good barn with stanchions and individual feed mangers. Wheat straw bedding was used. The heifers were permitted to exercise in a dry lot daily except during inclement weather.

The live weight, heart girth, and height of withers were recorded on three successive days initially and at the end of the 168-day feeding trial. In addition, single measurements were recorded every 4 weeks. The differences in gains were used to measure the relative feeding values of the rations.

RESULTS

The average amount of air-dry feed consumed was 14.61, 14.02, 16.22, and 15.63 lb. per heifer daily for rations 1, 2, 3, and 4, respectively (Table 1). The heifers receiving corncobs consumed about 10% less feed than did the others.

In rations 1 and 2, corncobs made up 51% of the total feed consumed. The concentrates consumed in the corncob mixture amounted to 2.5 and 2.4 lb. per

TABLE 1

Average daily feed consumption per heifer, air-dry basis (lb.)

Ration	Roughage	Concentrates	Total
1. Cobs, grain	10.11[a]	4.5	14.61
2. Cobs, grain, aureo.	9.52[a]	4.5	14.02
3. Hay, grain, aureo.	11.72	4.5	16.22
4. Hay, grain	11.13	4.5	15.63

[a] Corncob mixture No. 45 (75% cobs).

day for rations 1 and 2, respectively, in addition to the regular concentrate allowance.

Palatability of the cobs when mixed with soybean meal was not a serious problem. When the heifers were changed from standard rations to the experimental rations at the start of the experiment, the corncob mixture was consumed in relatively large amounts.

The average live weight gain for each ration group was normal or above, based on Morrison's *(2)* normal growth values for Holstein heifers (Table 2). The average daily gain in live weight was slightly higher for rations containing cobs (1.59 lb.) than for the hay rations (1.46 lb.). The increase in heart girth was 9.2 in. for cob rations compared to 7.7 in. for hay rations. The increase in height of withers was 4.85 in. compared to 5.15 in. in favor of the hay rations. These differences are not statistically significant. The analysis of variance for gain in live weight is given in Table 3.

The addition of aureomycin to rations 2 and 3 did not increase the live weight gains. The average daily gain for the aureomycin rations was 1.50 lb. compared to 1.55 lb. for the non-aureomycin rations. Based on a comparison of gains for rations within each kind of roughage, no beneficial effect of aureomycin was evident.

DISCUSSION

The live weight gains from the corncob rations demonstrated that cobs furnished a large amount of energy for growth. In planning the rations, protein concentrate was added to the cobs to make a mixture equivalent to the hay in protein and digestible energy, based on average feed composition values. The cob mixture had a calculated TDN value of 51.3% and should have replaced the

TABLE 2

Average gains in body measurements per heifer (24 weeks)

Ration	Live weight		Heart girth	Height at withers
	(lb.)	*(Av/day)*	*(in.)*	*(in.)*
1. Cobs, grain	270	1.61	9.0	4.5
2. Cobs, grain, aureo.	264	1.57	9.4	5.2
3. Hay, grain, aureo.	240	1.43	7.5	4.9
4. Hay, grain	248	1.48	7.9	5.4
Aureomycin rations	252	1.50	8.5	5.1
Other rations	259	1.55	8.5	5.0

TABLE 3
Analysis of variance for individual live weight gains

Source	Degrees of freedom	Mean squares	F value
Total	35	—	—
Periods	2	2,016	12.07**
Heifers	11	339	2.03*
Rations	3	194	1.16
Within rations	8	394	2.36*
Error	22	117	—

* Significant at 5% level.
** Highly significant at 1% level.

hay on an equal weight basis. However, the observed hay replacement value of the corncob mixture was equivalent to 63.3% TDN.

If the extra protein needed in the cob rations had been supplied by increasing the protein percentage in the concentrate mixture, less energy would have been supplied by concentrates and lower gains would have been expected. However, it was deemed desirable to add sufficient supplements to the cobs to make a mixture equivalent to the hay in nutritive value. There were several reasons for this: The cob mixture was more palatable than the cobs alone; the supplements needed to correct the deficiencies of the cobs were consumed in proportion to the cobs consumed; and a simple economic comparison can be made between the costs of the two rations.

In practice, rations fed to heifers may contain more than the minimum amount of protein required. Good quality legume roughage usually supplies more than the minimum allowances and, also, it is not practical to feed the exact minimum. Thus, the substitution of corncobs for part of the high-protein roughage, thereby reducing the surplus protein intake, may appear to show an economic advantage for corncobs. The critical economic test should be made with rations of equal or similar nutritive values.

The cost of the corncob mixture, which supplied a liberal amount of protein, was $33.80 per ton with corncobs priced at $6.00 per ton. Thus, there was no economic advantage for feeding cobs in place of hay at normal prices of $25 to $30 per ton. If hay is unusually high in price, it might be economical to feed cobs.

SUMMARY

Twelve dairy heifers were fed experimental rations for a 24-week period to compare corncobs with bromegrass hay as the source of roughage. Aureomycin was fed to half of the heifers in each roughage group. Protein, mineral, and vitamin supplements were mixed with the corncobs to make the mixture equivalent to the hay in nutritive values.

The rations containing cobs and extra supplements compared favorably with bromegrass hay rations. The average daily gains were 1.59 lb. for the cob rations and 1.46 lb. for the hay rations. The addition of aureomycin did not increase the live weight gains. Differences in gains were not statistically significant.

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TECHNICAL NOTES

THE DESTRUCTION OF OXALATES BY THE RUMEN CONTENTS OF COWS

It has been known for many years that ruminants can consume large quantities of plant oxalates without suffering any apparent ill effects due to oxalate poisoning. This immunity to oxalate poisoning, which is not exhibited by nonruminants, has been explained generally on the basis that oxalic acid is broken down by the rumen. Another explanation that is sometimes offered is based on the postulate that an appreciable portion of the oxalates pass through the digestive tract as inert calcium oxalate. These explanations, one or both of which can be found in most textbooks and reviews on ruminant nutrition, have been offered with reservations because no direct evidence has been available to support them *(2)*. Talapatra recently demonstrated in a qualitative manner that oxalates are destroyed in the rumen *(4)*.

During a recent feeding trial in which an oxalate-rich diet was fed to dairy cows, it was observed that no more than trace amounts of oxalates appeared in the blood, urine, and feces. These observations were in agreement with those of Talapatra *(4)* and strongly indicated that the oxalates had been broken down in the rumen. However, this evidence was not considered to be unequivocal because it did not eliminate the possibility that the oxalates had been absorbed and rapidly detoxified.

The present study was made for the purpose of collecting quantitative data on the destruction in vitro of oxalates by the rumen contents of cows.

The techniques used in maintaining conditions as regards to pH, temperature, and anaerobic conditions were essentially those developed and discussed in detail by Maynard and coworkers *(1)*.

Through the cooperation of the local slaughter house, an unlimited supply of fresh rumen ingesta was available. Samples of ingesta were collected under anaerobic conditions by forcing it out the cardiac orifice into glass containers maintained at 38° ± 1° C. All samples were collected immediately after the cows had been slaughtered. A sample weighing approximately 500 g. was found to be a convenient size for this study. An aqueous solution of sodium oxalate, previously warmed to 38° C. and adjusted to the proper pH, was added to each sample. These mixtures were then incubated at 38° ± 1° C. Aliquots were removed after 1, 2, 3, 4, 8, 16, and 24 hours, and the oxalate concentration was determined by the method of Moir *(3)*.

The results obtained during several initial incubation trials were very erratic. However, more consistent results were obtained when ingesta samples were taken from cows that had grazed during the morning and were slaughtered during the early afternoon. Table 1 shows some of the data collected after incubating samples from 35 different cows. These results demonstrate conclusively that a full rumen is able to destroy the oxalate ion. Further, the quantity that can be broken down in a given period far exceeds that which is apt to be consumed by a cow while grazing any of the commonly encountered forage grasses. Certain weeds and herbs which may contain extremely high concentrations of oxalates are not meant to be included in this discussion.

TABLE 1

The destruction in vitro of oxalic acid by the rumen contents of cows (incubated under anaerobic conditions at 38° ± 1° C.)

Cow No.	Weight of oxalate-free ingesta	Oxalic acid added as sodium oxalate	Oxalic acid remaining after 1 hr.	Rate of destruction for 100-lb. rumen
	(g.)	*(mg.)*	*(mg.)*	*(g/day)*
3	485	625	505	265
5	516	625	554	150
24	492	625	487	306
25	489	625	472	342
31	505	625	481	311
32	495	625	543	180
			Average	259

It should be pointed out with emphasis that this study does not demonstrate that the consumption of oxalate-rich grasses is not detrimental to cattle. This study merely shows that any ill effects resulting from the consumption of oxalate-rich grasses could not be due to the oxalate ion, per se. The decalcification of ruminants maintained on certain oxalate-rich forage is not due to the precipitation and excretion of calcium oxalate, as may be true with nonruminants, such as the rat. This decalcification of ruminants appears to be due to, or closely related to, the high concentrations of certain cations which remain after the destruction of the oxalate ion. This latter possibility is being investigated.

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PEOPLE and EVENTS
in the Dairy Science World

Pioneers in the Dairy Industry

CLAWSON YOUNG CANNON has a long and distinguished record in the dairy field. Born at Salt Lake City, Utah, he graduated from Utah Agricultural College in 1913 and remained as instructor in dairy husbandry until 1914. From then until 1920 he was an instructor in agriculture at Boise, Idaho, High School. Smith-Hughes work and the development of vocational agriculture courses in high schools were first introduced in Idaho during that time.

C. Y. Cannon

From 1920 to 1930, Cannon was a member of the animal husbandry staff at Brigham Young University. It was during this time, as owner of a farm and a Jersey herd, that he personally bred, fed, developed, and milked a Jersey cow which made a world's production record, as well as another which took twelfth place.

Spurred by an interest in educational work, Cannon obtained leave for advanced study and obtained the M.S. degree in 1923 and the Ph.D. degree in 1927 from Iowa State College. This was the first Ph.D. degree in dairy husbandry granted at that institution.

In addition to his duties on the animal husbandry staff at Brigham Young University, Dr. Cannon served as dean of the Summer School from 1927 to 1930. A tough decision was faced when he was invited in 1930 to succeed Earl Weaver as head of Dairy Husbandry at Iowa State College. To accept would mean leaving his beloved Utah, his farm, and his dairy herd. It was characteristic of C. Y. Cannon that he accepted the challenge and cast his lot with the development of dairying in Iowa. His wife, Winifred Morell Cannon, was a pioneer, too, and did not hold him to familiar ties.

Cannon was a stimulating and enthusiastic teacher and his inquiring mind led him into many interesting research areas related to dairy production. He is the author or co-author of more than 60 publications on nutrition and management. In addition, he has written many popular articles for agricultural magazines. When he reached the age of retirement from administrative duties in 1951, his former students, who are connected with many universities and other organizations all over this country and in foreign lands, presented him with a portfolio of letters commemorating his leadership.

His early publication of investigations on water consumption of dairy cows has become a "landmark." Soybeans were new in Iowa and Cannon directed his efforts to research on the use of soybeans and soybean by-products in the rations of dairy cows. Construction on the first milking parlor in Iowa was started at Iowa State College under his direction.

In spite of a busy schedule, Cannon always found time for his family. His three sons and a daughter were graduated from Iowa State College. Robert Cannon is on the dairy staff at Alabama Polytechnic Institute. Clawson is in the Music Department at Brigham Young University at Salt Lake City. The daughter, Mrs. Winifred Jardine, has continued to use her journalism training as Food Editor of the Deseret News in Salt Lake City.

Cannon served on many committees of the A.D.S.A. and was on the board of directors. He was a member of Phi Kappa Phi, Sigma Xi, Gamma Sigma Delta, the American Society of Animal Production and the A.A.A.S.

In November, 1951, Cannon was granted leave of absence to accept a position under the Office of Foreign Agricultural Relations in Beirut, Lebanon, where he has remained as agriculturist and research advisor in animal husbandry, helping develop the dairy program of that country. Dr. and Mrs. Cannon returned to the United States to visit friends and relatives in the summer of 1954 but went back to Lebanon to assist in programs of livestock improvement which he had helped get started.

C. Y. Cannon is recognized as a pioneer in dairying but is best thought of as father, friend, and teacher. A. H. PORTER

National Intercollegiate Dairy Cattle Judging Contest

The Production Section of the A.D.S.A. in the business meeting at the 50th Annual Meeting of the Association voted to reestablish its authority over the National Intercollegiate Dairy Cattle Judging Contest and officially authorize the rules under which it is conducted.

It then voted to recommend to the Association that the published rules and regulations for the conduct of the 1955 National Intercollegiate Dairy Cattle Judging Contest, except for time and place of the contest, become the official

rules of its conduct subject to the changes later approved. The official rules and regulations as adopted are as follows:

Teams and Entries

The teams shall consist of three members representing the agricultural college in which they are regularly enrolled.

Eligibility of Contestants

Each contestant must be a student of a Land Grant agricultural college in the United States or of an agricultural college of corresponding rank in the Dominion of Canada or of an institution offering a full degree course in agriculture with a full major in dairy production in a division of animal husbandry or dairy husbandry, whose application is approved by the membership of the American Dairy Science Association at their annual meeting and accepted by the management of the National Intercollegiate Dairy Cattle Judging Contest.

The student must be enrolled in a 4-year or longer course leading to a degree in agriculture, veterinary medicine, or agricultural education and must have completed not less than 36 weeks of college work. Any student who has been a member of a team competing in the National Intercollegiate Dairy Cattle Judging Contest or has acted as an official judge of dairy cattle at a fair or show or has at any time served as a teacher of dairy cattle judging in an agricultural college or secondary school or has completed a 4-year course in college work or has competed in more than two intercollegiate dairy cattle judging contests is not eligible for entry in this contest.

If the eligibility of any student is protested, such protest may be made in writing at the arena side on the day of the contest before the contest begins. The protest must be accompanied by a check for $25, which will be returned if the protest is sustained. If the protest is not sustained, the $25 shall be forfeited.

Superintendent

The superintendent shall have complete charge of the contest. He shall see that all rules and regulations governing the contest are duly carried out and that the contest is conducted with fairness and justice to all concerned. The superintendent shall also decide all questions which may arise in connection with the interpretation of the rules.

Judging System

A committee of judges shall place all classes and hear and grade reasons. If the number of teams entered indicates that additional associate judges are necessary to expedite and make more efficient the hearing and grading of reasons, the superintendent shall have the authority to appoint such judges.

Method of Conduct

Neither a member of any team nor the coach shall be allowed in the cattle barns or have the privilege of inspecting any cattle on the grounds previous to the contest.

Inspection of cattle. Team members shall be divided into a sufficient number of groups to provide the best opportunity to view and judge the animals; one assistant superintendent shall have charge of each group. Contestants shall not at any time place their hands on any animal.

Reasons. Contestants shall give oral reasons, 2 minutes in length, on one class of each breed, and of these not less than three nor more than four cow classes shall be included. The superintendent shall notify the contestants before the contest starts, the classes on which reasons will be required in order that contestants may take notes. Reasons will be given on the afternoon of the day of the contest.

Materials. The superintendent shall supply placing cards and any other necessary forms for conducting the contest. No contestant shall be allowed to take any book, notes, or writing paper into the ring except such materials as are furnished by the superintendent.

Time allowed. Contestants will be allowed 15 minutes to place each class and will be notified at the end of 12 minutes.

Classes

Ten classes of four individuals each shall be judged. These shall consist of five classes of cows, two classes of bulls, and three classes of yearling heifers in the Holstein, Guernsey, Jersey, Brown Swiss, and Ayrshire breeds. The animals shall be held in a careful manner so that all contestants may have a fair chance to examine them. The animals shall be lined up consecutively in single file and numbered 1 to 4, inclusive.

Determination of Ratings

Ratings shall be based upon a possible score of 50 points for each ring placed and 50 points for each set of reasons. Since there are ten classes to be placed and five sets of reasons, there will be a possible total score of 750 points per contestant.

The final rank of each contestant in each breed shall be determined by totaling his grades on placing and reasons for that breed. The individual rank for all breeds shall be determined by adding his grades for each of the five breeds.

The team ranking for each breed shall be determined by totaling the grades on placings and reasons of each of the three team members for that breed. In like manner the team ranking for all breeds shall be determined by combining the grades of the three team members for each of the five breeds in the contest.

In case of a tie, the individual or the team ranking highest in placing shall be awarded the prize in question.

Is Your Department 100 Per Cent?

LYMAN RICH, chairman of the A.D.S.A. membership committee, has reported that all members of the Dairy Dept. of the Utah Agricultural College are now members of our Association. It is Lyman's hope to make this achievement universal throughout the country. He has organized a committee to cover all sections to solicit members from industry as well as universities. Every member should feel obligated to help. Why not give memberships in A.D.S.A. as Christmas gifts this year?

Ohio News

The Dairy Technology Dept. of the Ohio State Univ. assisted in conducting the dairy products phase of the Ohio State Fair. In the dairy products building there were several exhibits, including a full size cow sculptured in butter and a full size figure of Davy Crockett, also in butter. The sculpturing was done by J. E. WALLACE, who has been appearing at the Ohio State Fair since 1914. Other exhibits included a model market milk plant operation, a moving slide story of the dairy industry of Ohio, and a series of color transparencies depicting ice cream manufacturing operations.

A unique feature of the building was the sale of prize Swiss cheese, prize Cheddar cheese, Swiss cheese and rye bread sandwiches, high quality ice cream, and plain and chocolate milk. Approximately $20,000 worth of these products were sold. Contests were held for Swiss cheese in block, Swiss cheese in wheels, Cheddar cheese under 3 months of age, Cheddar cheese over 3 months of age, cottage cheese, and butter workmanship, wherein the contestants were awarded prizes for good consistent effort in making good butter. Cash prizes and ribbons are awarded for winners in each of these classes.

Another feature of the dairy products phase of the Fair were the judging contests. These contests were open to anyone, including students in dairy technology who may later be on the judging teams. There were four contests: milk judging, ice cream, butter, and cheese. Sixty dollars was awarded in prizes among the top five contestants for each contest.

Manufacturers Conference at MSU

The 16th annual Michigan State Univ. Dairy Manufacturers Conference will be held on Wednesday and Thursday, Nov. 2 and 3. The Wednesday afternoon program will feature products clinics on milk, ice cream, and butter. The third annual MSU Dairy Manufacturers Award will be presented to an outstanding member of the dairy processing industry at the Wednesday evening banquet. On Thursday the nearly completed MSU Dairy Manufacturing Center will be opened for inspection.

Kentucky Events

The faculty of the Univ. of Kentucky has approved a rather extensive revision of curricula of the College of Agriculture and Home Economics, including those of dairy production and dairy manufacturing. Both of the dairy curricula have been redesigned to give the student a stronger background in basic sciences and an opportunity to include more cultural courses in his program. For the freshman year the curricula are identical. In the following years the curriculum in dairy production will include more training in nutrition, breeding, and management.

In the dairy manufacturing curriculum all individual products courses have been dropped. Basic instruction and theoretical work concerning the products have been combined on a utility basis. Practical work and management will be taught on this basis also. At the beginning of their junior year, students will be required to choose one of the following options: general dairy manufacturing, laboratory research, or administration.

LINVILLE J. BUSH, field agent in dairying, has resigned in order that he may continue study for a Ph.D. degree at Iowa State.

Representatives of six western Kentucky dairy firms met with members of the Dairy Section at Bowling Green to organize the Western Kentucky Manufacturers Milk Improvement Assoc. Purpose of the organization is to further improve the milk quality in that area of the state and to meet and discuss other problems of the dairy industry. J. O. BARKMAN, associate professor of dairy manufactures, and W. L. KING, field agent in dairying of the University, will work with the group in development of a program.

South Carolina News

The Agricultural Center of Clemson College, consisting of two new buildings, was dedicated on Aug. 17, 1955. The Plant and Animal Science Building contains 175,000 sq. ft. of floor space. Completely air-conditioned, its rooms furnish classrooms, laboratory facilities, and office space for dairy teaching, research, and extension personnel. The Food Industries Laboratory, adjacent to the Plant and Animal Science Building, provides facilities for processing dairy, poultry, livestock, and horticultural products. This building contains 75,000 sq. ft. of floor space and will be used for teaching and research. The dairy section features a quality control laboratory and modern equipment for processing milk, ice cream, cheese, and other dairy products. The Clemson Blue cheese project is provided with facilities in a special space for establishing control conditions.

A graduate program leading to a master of science degree in dairy industries was initiated at Clemson College for the fall semester. The graduate program in dairy production has been in effect for several years.

News from the Illinois Campus

G. W. SALISBURY, head of the Dept. of Dairy Science at the Univ. of Illinois, left Sept. 1 for sabbatical leave in The Netherlands. Dr. Salisbury will study and teach at the Wageningen Agricultural College during his stay there.

Prior to his return in August, 1956, he will attend the IIIrd International Congress on Animal Reproduction at Cambridge University. During Dr. Salisbury's absence K. E. GARDNER will serve as acting head.

C. S. RHODE, who has been in charge of dairy extension at the Univ. of Illinois for a number of years, retired August 1. He is devoting full time to the selection of bulls for the Northern Illinois Artificial Breeders' Assoc. at Hampshire, Ill. He has been succeeded at the University by J. G. CASH, who has been assistant to Professor Rhode for a number of years.

Illinois to Hold Disaster Conference

A conference on "Practical Planning for Emergencies in the Dairy and Food Industries" is to be given under the sponsorship of the Department of Food Technology at the Univ. of Illinois, Dec. 7 and 8. Registration will be limited to 75 and is open only to key men in the food industries. A series of related conferences will be held throughout the state at a later period to acquaint workers with proper procedures to follow in time of a disaster. Government and state officials will cooperate in presenting the material. Disaster areas to be covered include atomic explosions, tornadoes, floods, and biological and chemical contamination. P. H. TRACY, professor of Dairy Technology at the Univ. of Illinois, is chairman of the planning committee.

Virginia Polytechnic Institute News

The name of the Dept. of Dairy Husbandry at V.P.I. has been changed to Dept. of Dairy Science. An attractive, printed brochure describing the department and the opportunities in dairying in Virginia for graduates in dairy science has been prepared. The brochure, entitled "Your Future and the Dairy Industry," will be used in the recruitment of students of dairy science. The brochure was sponsored by the Virginia Dairy Products Assoc. and prepared by the Dept. of Dairy Science.

H. B. HENDERSON, Univ. of Georgia, was one of the featured speakers at the general sessions of the Institute of Rural Affairs on the V.P.I. campus on July 28. His topic was "Milk and You." The Dept. of Dairy Science sponsored two sectional meetings during the Institute. One program dealt with the use of dairy products in the home and was attended by farm women; the other dealt with green forage feeding of dairy cattle, dairy research in progress, and the V.P.I. dairy herd.

A Rogers spray milk dryer with a capacity of approximately 50 lb. of dried product per hour is being installed in the dairy building.

D.H.I.A. Records Point the Way

The following table, taken from a recent Dairy Herd Improvement Assoc. letter, shows the production of the Association cows since 1906.

Year	Cows on test	Average production	
		Milk	Butterfat
	(No.)	*(lb.)*	*(lb.)*
1906	239	5,300	215
1910	25,000[a]	5,730[a]	227[a]
1920	203,472	6,175	247
1930	507,549	7,642	303
1940	676,141	8,133	331
1950	1,088,872	9,172	370
1951	1,186,615	9,195	370
1952	1,166,297	9,192	366
1953	1,226,588	9,253	368
1954	1,311,698	9,363	372

[a] Estimated.

According to the letter, the fundamental principles that have been followed by the association members throughout this period are:

1. Cull the unprofitable cows from the herd.
2. Feed according to individual producing ability.
3. Select the best animals for breeding stock.

A simple yet effective program such as this should not be difficult to sell the dairymen of our country.

Business Notes

The Tastee Freez Corp. plans to establish 12 new stores in Trinidad, British West Indies. The first two stores will be built in Port of Spain. By the end of 1956 the Corporation expects to have 2,000 stores.

The Continental Can Co. has added the "Bondware" line, a nested paper service for the dairy and food industries.

Beatrice Foods Co., Chicago, has added further diversification to its line of dairy and food products by the purchase of the D. L. Clark Co., Pittsburgh candy manufacturers.

Fairmont Foods Co. of Omaha has added to its increasing list of dairy plants the Beach Milk Co. of Denver.

The Cherry-Burrell Corp. recently announced the addition of the Westfalia clarifier and separator to its line of dairy plant equipment. These machines are made in Germany by the Westfalia Separator Ag., a company founded in 1893. The machines are built in a wide range of capacities with either the liquid or hermetical seal. Either white enamel or stainless steel finish can be obtained.

Minute Maid Corp., New York City, has consented to dispose of the Snow Crop Division of Clinton Foods (Iowa), which it recently purchased as a result of suit brought against them by Attorney General Herbert Brownell for violation of the Clayton Act. Minute Maid, one of the largest processors of frozen citrus juice, purchased the Snow Crop plants at Dunedin and Frostproof, Florida, in which state they have extensive holdings.

Completed Theses

M.S. degree:

D. L. MacFadden—The determination of body composition by the use of antipyrine in aureomycin fed dairy calves, noting their rate of gain and feed efficiency. Univ. of Delaware.

J. M. Treece—The inheritance of head patterns in spotted breeds of cattle as represented by Holstein-Friesians. The Ohio State Univ.

Ph.D. degree:

H. E. Affsprung—The use of ion exchange resin membrane electrodes in the study of the inorganic equilibria of milk. Univ. of Missouri.

June Marshall Baker—Studies on the inorganic equilibria in milk by an ion exchange resin contact time method. Univ. of Missouri.

The Dairy Industry Today

A Guest Editorial

A. H. Bayer

Our industry today differs considerably from that of 25 years ago in respect to methods of production, distribution, regulations, and outlets. All of these have changed the philosophy of business in respect to the present and the future. A look at the production phase of our industry is revealing.

There have been changes in milk processing as follows:

Short-time high-temperature, continuous pasteurization of milk.
Vitamin enrichment of milk.
Homogenization of milk.
Bulk handling of producer's milk.
Paper containers.
Continuous in-place cleaning of sanitary lines and other equipment.
Elimination of 40-qt. cans in processing of by-products.
Equipment of better sanitary and mechanical design.
Reduction in bacterial standards for products—raw and finished.
Improved housekeeping and sanitary plant conditions.
Improved plant design.
Mechanization of operations.
Automation to a limited degree.
The 40-hour 5-day week.
The addition of new items, such as the 2-qt. and 1-gal. container and special by-products containers; orange juice and flavored drinks.
A greater and better utilization of refrigeration in processing and delivery.
Warehouse delivery.

In our ice cream operations, in addition to some mentioned above, the following practices are now common:

Continuous freezers.
Automatic packaging.
A complete change from bulk to a large variety of smaller units of sale.
Mechanization of operations with some automation.
Mechanization in product handling and transportation.

All of these changes have resulted in better products with longer shelf life and a much higher productivity per man-hour.

However, new problems now confront management, such as:

Need for better trained men in supervisory and junior executive positions.
Greater variety of items needed to care for sales.
Changing trends in types of outlets and private labeling.
The short work week.
The attitude of labor.
The maintenance problem resulting from mechanization.

In the early days of our industry, many scientists were interested in the improvement of dairy products and devoted their efforts in that direction. These were the men who did the most in developing the industry to its present state. They usually received recognition but not always a monetary reward. Today there seem to be fewer of these men in our industrial operations. Perhaps this is because management may have lost sight of the need for such men. If true, this is an unfortunate situation which may eventually retard the progress of our industry. The need for scientifically trained men is greater than ever before. Management should encourage them by providing the opportunities and recognition necessary to keep them satisfied to remain in the industry.

The emphasis in recent years has been placed on mechanization, automation, methods improvement, and productivity. All of this is important, but it should not be forgotten that there is a need for men with knowledge and appreciation of the science of dairy technology if we are to do the best job with these new tools. There is an ever increasing need for men with initiative and imagination and a desire to make a career out of the science of dairy plant management.

Currently more and more is being done in the management of our business by junior or mid-line executives in respect to planning, organization within the plant, conference supervision, employee problem solving, labor handling and training. The junior executive is not the over-all plant manager but the specialized

manager of production, selling, accounting, or engineering.

Many plant operators or superintendents grew into the job without scientific, engineering, or management-training background. Yesterday's background of "growing up in the business" does not provide the tools for today's job. The need now is for men with the proper background to learn a specialized phase of the business. Today's job in plant operations requires more time for thinking, planning, and organizing, than for muscle. The industry has now grown to man size but with too few men capable of coping with today's complexities.

It is impossible to obtain a ready-made man, college trained in dairy technology, accounting, engineering, and management, who has the skills necessary for an over-all job of production plant management. This is not an indictment of the colleges and universities. In this age of specialization in industry, as well as in education, it perhaps is not to be expected that an all-around, broadly-trained man would be available from any school.

It was once customary to call the man in charge of operations a superintendent. Truly he is a production manager required not only to get out the number of units desired but to do it efficiently and under the most exacting conditions of housekeeping and sanitation. He must plan smoothly, hire effectively, deliver and transport efficiently, and direct labor relations and safety programs. He should be trained for these responsibilities and should be paid a salary commensurate with the level of his responsibilities.

Where can industry get such men? They can take graduates trained in dairying, engineering, management, and accounting. After the men have been properly screened and appraised, they will then need to be extensively trained in the specific requirements of the job. They must learn the scientific and engineering backgrounds and be given experience in work simplification methods, management and human relations problems, and the accounting system used. Skills in these areas cannot all be obtained in college, but they can be acquired in industry if a properly outlined program is set up and followed.

Such a program is expensive and time-consuming, requiring application and patience on the part of both the employer and the employee. Industry may be reluctant to embark on such a program because of the cost and time involved. However, those who do not will be behind the times in respect to methods, costs, morale, quality and efficiency in their plant operations. The program to train an individual may take from 3 to 5 years. It may require enrollment in special schools of instruction (now available) from 3 to 6 weeks each year, in management, in work simplification, and in other phases of the business.

In the past, a plant manager had to be a salesman, an accountant, a production man, an engineer—a jack of all trades—without the background and qualifications needed. Today, because of the complexity of business and the growing importance of each phase of the business, the organization of operations with a thoroughly trained and competent production manager in charge is necessary.

Industry today, in addition to being complex, employing new ideas and mechanization, and being involved in human relations, is at a turning point in respect to the immediate future. What happens will depend on its ability to attract the right caliber of men and to train and develop that man power. Attracting the right men involves paying them a satisfactory wage and providing opportunity for advancement. Industry has been slow to recognize that good men should be paid good salaries and given opportunity to develop and grow.

Production is only one angle of the change in industry. There has been a similar change in accounting, transportation and delivery, selling, and other phases of business. In all phases there is a common problem—it pertains to the selection, training, and rewarding of men for leadership. The solution of this problem is up to top management in industry.

A. H. Bayer
National Dairy Corporation

OUR ASSOCIATION

Credits and Debits—and a Job to Do

Dear Fellow Members:

Can we not imagine what would happen to our Association—or to any association—if the members were mainly of the "getting" and not the "giving" kind? The dismal and rapid demise of such an organization is clear to foresee. And how pleasant is the task of the president in an organization where the members "pitch in," where they accept committee appointments willingly and eagerly, and where there is the common spirit of getting the job done. Such a response by the members has been mine to enjoy these past few months in the process of organizing the Association committees, and it brings to me—as I am certain it did to my predecessors—a glow of pride to be even a small part of a group where members are so willing to play the game 100%—to "carry the ball" when their "numbers are called." Surely, this cooperative spirit bodes well for the Association and insures for it the grand GOLDEN JUBILEE YEAR we are hoping for and the bright future we all envision. My heartfelt personal thanks to all who have accepted these various assignments and who are already hard at work for the Association.

And NOW!—will you mark November 1 on your calendar—and fix it in your mind—because this is the kick-off date for the most ambitious membership and Journal subscription campaign in the history of our Association.

Our membership is now 1,750 and a minimum goal of 2,000 has been set for this year. Lyman Rich of Utah and his enthusiastic membership committee have the campaign organization established at the state level—and each state has a quota—to increase members by about 14%. They have lighted the torch and started the parade and are anxious that you march with them. We all know that membership selling is most effective when conducted on a personal basis; therefore, will you not each become a part of the membership committee and do your share to solicit new members for the Association and new subscribers for the Journal?

Not only will the getting of new members and subscribers result in a stronger Association capable of rendering greater service, but, in terms of dollars and cents, it is just good business on the part of the present membership. We cannot ignore the fact that it takes money to run our Association, and the requirements have increased greatly in recent years mainly because of a normal rise in printing costs of the Journal. Last year (1954). $6,000 worth of government bonds, held in reserve, were cashed in order to balance the budget. Also, increases in reprint charges and in membership, subscription, and advertising rates were made in order to achieve reasonable balancing of the 1955 budget. Financially, our Association is now in a better condition. However, costs are continuing upward, and, in addition, continuing expansion and improvement of the Journal and an enlargement of the Association activities are desirable and inevitable—all of which will result in a further cost burden. It stands to reason that the simplest means to greater Association income, and the one which is least costly to the present membership, is by increasing the number of members and subscribers. Indirectly, too, this will help through greater advertising revenue.

The selling of our Association is not difficult. We have a great deal to offer and positive sales points are before us. To be a member of the leading scientific and educational organization in the dairy field would seem to be a professional necessity for any aggressive and progressive qualified person, or at least a matter of professional pride. To have the opportunity to speak through the Journal, to attend the annual meetings, to elect officers, and to participate directly in Association affairs are privileges that should be desired by every person in the dairy field having technical background and holding a position of responsibility. All those who are staff members of dairy departments of educational institutions may be expected to belong to the Association—and to wield their influence in guiding the dairy students into the Affiliate Membership fold.

From the cost standpoint, the membership and subscription rates are low in comparison to those of other organizations of similar stature; from the quality standpoint, the Journal is the outstanding publication of its kind. When someone offers sales resistance by saying, "I don't understand the Journal," or "It contains too many articles on subjects I'm not concerned with," etc., the opportunity is ours to point out that the Journal is a publication of ideas—ideas which come from original research—and that, page for page, issue after issue, there is no other publication which can match its idea value! And what is an idea worth? Certainly many, many times the membership or subscription fee! The Abstract section and the Industry and Events sections are other logical points to emphasize, especially in contacting persons in the commercial field.

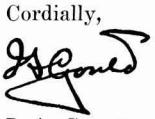
Surely, we need feel nothing but pride and confidence in embarking upon this sales campaign. The goal is high but is within easy reach. Can we not make the 4-month period of Nov. 1 to March 1 outstandingly productive in new members and new subscribers?

Just a word about another important committee that is hard at work: the special Education Committee headed by E. L. (GENE) JACK of California. This committee has the big task of finding ways and means by which the Association may effect improvement in the teaching ability of its members. The time is long overdue when we should look critically at the teaching

methods and materials being used at the undergraduate, graduate, and adult education levels. The committee will be planning for a special session on teaching at Connecticut and will welcome your suggestions. Also, with your help the Committee must reach a decision as to whether or not our Association should establish a new section on Dairy Education.

In conclusion, I invite you each to serve as eyes and ears of our Association and to convey to us, your officers, the things you see and hear which will make for a better organization. Free communication and open discussion are essentials of a strong Association.

Cordially,



I. A. GOULD
President

THE AMERICAN DAIRY SCIENCE ASSOCIATION CONSTITUTION AND BY-LAWS

Revised 1955

Article I — Name

Section 1. The name of this organization shall be The American Dairy Science Association.

Article II — Object

Section 1. The object of the Association is to promote the welfare of the dairy industry by stimulating scientific research, improving educational methods, encouraging worthy intra-industry and inter-industry cooperative endeavors, and by publishing the Journal of Dairy Science and other official periodicals.

Article III — Membership

Section 1. Any person shall be eligible to membership who has had College training in dairying or who is in a position of responsibility that requires a technical knowledge of dairy science.

Section 2. Any person shall be eligible to nonvoting membership as a student affiliate who is a regularly enrolled College student and who does not hold a rank of instructor or higher or the equivalent thereof.

Article IV — Officers

Section 1. The officers of the Association shall be President, Vice-President, Secretary-Treasurer, Journal Editor, and seven Directors, one of whom shall be the immediate Past-President.

The Vice-President shall be elected by the vote of the membership for a term of one year beginning at the time of his installation during the first annual meeting of the Association following his election. If an annual meeting cannot be held, his term of office shall begin on July first. On the completion of his term as Vice-President, he shall automatically become President for one year beginning at the time of his installation during the annual meeting, or, in the absence of such a meeting, on July first.

The Secretary-Treasurer and the Journal Editor shall be elected annually by the Executive Board.

Two directors shall be elected by the membership each year to hold office for a term of three years beginning either at the time of the installation of officers at the first annual meeting following their election or on July first if an annual meeting is not held.

Section 2. The Executive Board shall consist of the President, Vice-President, seven Directors, and with the Secretary-Treasurer and the Journal Editor as ex-officio members. The Board shall be responsible for the business of the Association.

Article V — Meetings

Section 1. Meetings of the Association shall be held at least once during each calendar year. The exact date and place of each meeting shall be fixed by the Executive Board. Notice of the time and place of meetings of the Association shall be given to all members not less than four weeks prior to the date of the meeting. In an emergency the annual meeting may be canceled by action of the Executive Board.

Article VI — Amendments

Section 1. The Constitution and By-Laws may be amended at any meeting of the Association by an affirmative vote of three-fourths of those members present, provided not less than five per cent (5%) of the voting membership is present at the meeting. All amendments must be submitted for approval only after they have been presented in writing to the membership at the previous regular business meeting or have been published in the Journal of Dairy Science at least 30 days before the regular meeting at which the amendments are offered for approval. All amendments must have been acted upon by the Executive Board prior to final action by the Association.

Section 2. The Executive Board may submit proposed amendments, approved by the Board, to the members of the Association for vote by mail. In such a case, a minimum of twenty-five per cent (25%) of the membership must vote on the proposed amendment and an affirmative vote by two-thirds of all voting shall be necessary for its approval.

BY-LAWS

Article I — Duties of Officers

Section 1. The President of the Association shall preside at all meetings of the Association and the meetings of the Executive Board, and

shall perform such other duties as pertain to that office. The President shall call meetings of the Executive Board, and notices of such meetings shall be sent to each member of the Board not less than ten days before the meetings. As chairman of the Executive Board, the President shall submit to the Executive Board for approval his nominations of members to fill vacancies that may occur among elective offices of the Association. The President shall appoint without the approval of the Executive Board the standing, nonelective committees of the Association.

Section 2. The Vice-President shall perform the duties of the President in the absence, illness, resignation, or death of the President.

Section 3(a). The Secretary-Treasurer shall manage the business of the Association in accordance with the policies established by the Executive Board.

(b). He shall have custody of the books and records of the Association, keep the minutes of all meetings of the Association and the Executive Board, maintain a list of all members and subscribers, keep the funds of the Association, maintain in current condition an official Procedural Handbook of the Association, submit an annual budget for consideration by the Executive Board, make disbursements as authorized in the budget approved by the Executive Board, and cause an annual audit of the books to be made by a certified public accountant.

(c). He shall receive applications for membership which are submitted in writing and endorsed by one member. He shall refer to the Executive Board applications of doubtful eligibility. He shall, upon receiving the annual dues of the successful applicant, enroll him as a member of the Association, and shall place his name upon the list of those to receive the Journal of Dairy Science.

(d). He shall remove from the roll of members those individuals who have failed to pay the annual dues on or before January first. He shall restore these individuals to membership upon receiving their payment of current dues.

Section 4. The Editor of the Journal of Dairy Science shall have direct charge of all editorial details of the Journal and of other official regular publications of the Association under the general supervision of the Journal Management Committee and shall assume such other management responsibilities as may be designated by the Executive Board upon the recommendation or with the approval of the Journal Management Committee.

Section 5(a). The Executive Board shall have full control of the business of the Association and shall report its official actions to the members of the Association at the annual business meeting, or, if such a meeting is not held, through the Journal of Dairy Science.

(b). The Executive Board shall hold the title to all property and funds of the Association and shall have all the rights and powers vested in the Association by the laws of the District of Columbia under which it was incorporated.

(c). The Executive Board shall pass upon all applications for the establishment of divisions, sections, and student branches of the Association.

(d). The Executive Board shall fix the amount of annual dues to be paid by members and student affiliate members, and the amount to be paid by nonmember subscribers to the Journal of Dairy Science.

(e). The Executive Board shall adopt the annual budget under which expenditures of Association funds will be authorized by the Board.

(f). The Executive Board annually shall elect the Secretary-Treasurer and the Editor of the Journal of Dairy Science.

(g). The Executive Board shall elect three members of the Association, who, with the Secretary-Treasurer and Editor of the Journal of Dairy Science as ex-officio members, shall constitute the Journal Management Committee. This Committee shall be responsible to the Executive Board and shall have general supervision of the Journal of Dairy Science and other official periodicals of the Association. The Executive Board shall elect to the Journal Management Committee one member each year for a term of three years. Members may be elected to succeed themselves for one term only. The elected member having seniority of service shall be the chairman of the Journal Management Committee. A member reelected for a second term shall become the junior member of the committee.

(h). The Executive Board may appoint or cause to be appointed such committees of the Association as it deems necessary.

(i). The Executive Board shall have authority to approve or disapprove nominations made by the President to fill vacancies in unexpired terms of office that may occur among the elective officers of the Association.

(j). The Executive Board shall have the authority to present to the Association a resolution asking the expulsion of any member whose conduct has been shown to be damaging to the Association, or its reputation, or to the objects of the Association after: (1) the individual has appeared before the Board; (2) the individual has heard reasons for his expulsion presented by his accuser; and (3) the individual has had opportunity to present his witnesses and to plead his case, with or without the benefit of counsel, who must be a member of the Association.

(k). The Executive Board shall have authority to define and establish awards and to grant honorary and life memberships according to conditions described in Article III of the By-Laws.

Article II — Election of Officers

Section 1. Nominations for the offices to be filled by membership voting shall be by a Nominating Committee appointed by the President.

Section 2. The Nominating Committee shall have five members including the immediate Past-President, one representative each from the extension, manufacturing, and production sections, and one representing commercial interests. The names of the Committee shall appear in the January issue of the Journal of Dairy Science to permit members to offer suggestions to the Committee. Such suggestions should be in the hands of the Committee not later than April 1.

Section 3. The Nominating Committee shall select two candidates for each office to be filled. The selections shall be made preferably to permit yearly alternation of the office of Vice-President between the two broad fields of interests of production and processing of milk. Selection of candidates for the offices of Directors shall be chosen to provide equal representation of the production, manufacturing, and extension sections.

Section 4. The official ballot, containing the nominations of the Nominating Committee and pertinent biographical information regarding each candidate, shall be mailed by the Secretary-Treasurer to each member of the Association on or before May 1. Ballots shall be returned to the office of the Secretary-Treasurer by the members voting.

Section 5. The ballots shall be opened and counted by a three-member Balloting Committee appointed by the President, one of whom may be the Secretary-Treasurer, and the results certified to the President. A tie vote shall be broken by the Executive Board.

Section 6. The elected officers shall be so informed by the President prior to the annual meeting at which the election results will be announced publicly. New officers will begin their term of office on July first if the annual meeting is not held.

Article III — Awards and Recognitions

Section 1(a). An award of Honorary Membership may be bestowed by the Association upon any person who has been a member of the Association for not less than ten years, who has participated in the activities of the Association, and who has rendered meritorious service to the dairy industry in research, academic teaching, extension, administration, or industry.

(b). Honorary Members shall be entitled during their lifetime to all the rights and privileges of membership and shall receive the Journal of Dairy Science without the payment of annual dues.

(c). The citation and award of Honorary Member shall be presented at a suitable function of the Association during the annual convention.

(d). No more than one honorary membership may be awarded during any one year.

Section 2. The Association, upon the recommendation of the Executive Board, may grant life membership to any individual who has been a member of The American Dairy Science Association for 25 years or more and who, upon retirement, advises the Executive Board in writing of his eligibility. The status of life membership shall entitle the holder to all the rights and privileges of regular members without the payment of annual dues.

Section 3. The American Dairy Science Association may honor chosen members for distinguished services or achievements. The Executive Board shall be charged with the responsibility of defining and establishing, or causing to be defined and established, suitable awards or recognitions to accomplish this purpose. The Executive Board may accept the cooperation, financial or otherwise, of organizations or individuals who may wish to participate in honoring the man chosen to receive such awards or recognitions.

Article IV — Organization of Divisions, Sections, and Student Branches

Section 1(a). Divisions of the Association, organized by members of the Association on the basis of geographical location, may be authorized by the Executive Board upon petition by members of the Association.

(b). Membership in Divisions is open only to those who are members of the Association.

(c). The Divisions shall select officers, one of whom will be the chairman, and shall govern themselves in a manner consistent with the Charter, the Constitution, and the By-Laws of the Association.

(d). Divisions may collect funds and/or dues to be expended for their own purposes.

Section 2(a). Professional groups organized by members within the Association on the basis of specialized interests and to be known as sections, may be authorized by the Executive Board upon petition of not less than ten members.

(b). The Sections shall elect their own officers and make rules for their own guidance consistent with the Constitution and By-Laws of the Association.

(c). Sections shall conduct their official business during the annual meeting of the Association.

Section 3. Student Branches of the Association, organized by college and university groups with interests in the dairy industry, may be authorized by the Executive Board on petition from a majority of the local group's members and on recommendation by two faculty representatives who are regular members of the Association. Annually, the secretary of each Student Branch shall submit a brief report of its activities to the Secretary-Treasurer of the Association.

OUR INDUSTRY TODAY

Brief Reviews of Current Topics

Evaporation of Water from Milk by Spray Drying[1,2]

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The distinguishing characteristic of spray drying is the large surface area presented to the drying air. Because of the large surface the drying rate is rapid and the drying time very short. Mathematical relationships have been developed for air drying under static conditions such as in tray drying. The general theory of air drying can be applied to spray drying, but adequate data are difficult to obtain because of the shortness of the drying time. Mathematical treatment of the drying data is complicated because the conditions of drying, such as the humidity and temperature of the air, the moisture content and diameter of the particle, and the relative motion between the particle and the air stream, are constantly changing.

Owing to the lack of adequate data on which to base design, spray dryers generally have been built on an empirical basis. Widely different designs have been evolved. The effect of variation in operating conditions on powder characteristics may not be the same in all dryers, and the complexities of dryer operation tend to obscure these relationships even in the same dryer.

Spray drying usually is visualized as involving three operations: *(a)* atomization, *(b)* evaporation from drops, and *(c)* flow and mixing of gases and particles. This paper will include a brief discussion of pertinent aspects of drying theory and a consideration of operational and design factors influencing milk powder quality. Commercial operators are primarily concerned about meeting quality standards at a reasonable production cost. The necessity of minimizing heat damage and at the same time attaining low moisture levels in a heat sensitive, hygroscopic material such as dry milk is at times difficult. Factors which influence heat damage, therefore, such as the temperature and time of drying, and those determining the moisture content of the powder are of primary importance.

Drying usually occurs in two more or less distinct stages. The first is a period of surface evaporation known as the constant rate period, and the second, a falling rate period, during which the rate of evaporation continually decreases. The evaporation of pure liquid drops would involve only the first or constant rate period. Duffie and Marshall *(1)* have shown that the time for complete evaporation of pure liquid drops increases as the square of the diameter of the drops. At moderate temperatures particles 20 to 30μ in diameter may dry in a fraction of a second. Larger particles may require several seconds. Kitzes *(2)* secured data in spray drying milk products showing that there is no clearly defined constant rate period, but at about 80% solids the drying rate begins to fall off sharply and at the lower moisture levels may be very slow, particularly from the larger particles.

During the early stages of drying, water moves freely to the surface to maintain the surface completely wetted. Even while the surface is completely wetted, the drying rate falls because of changes in a number of factors, including the rate of movement of the air in relation to the movement of the particle, the size of the particle, and the humidity of the air. As the concentration increases, the rate of water movement becomes so slow that the surface is no longer maintained completely wetted. Rate of water movement then becomes a limiting factor and this decreases with further increase in concentration. With some materials the zone of evaporation may recede from the surface so that diffusion of water vapor also becomes a limiting factor. Microscopic examination of spray dried milk reveals no distortion of the surface, as would be expected if expansion of water vapor occurred within the particle, which seems to be evidence that evaporation occurs at the surface of the particle. Probably the time of drying during the falling rate period also increases as the square of the droplet diameter, although adequate data on this point are lacking.

One effect of the relationship between particle size and time of drying is that the larger particles will have a higher moisture content at any given stage in the drying operation. As shown by the data in Table 1, this may persist for several days after the powder is removed from the drying chamber, although equilibra-

[1] Presented at 50th annual meeting of American Dairy Science Association, East Lansing, Michigan, June 20-23, 1955.

[2] Paper No. 897, Miscellaneous Journal Series, Minnesota Agricultural Experiment Station.

TABLE 1

Moisture content of different sizes of particles sieved from nonfat dry milk solids

Size of mesh		Sample 1[a]			Sample 2[a]	
Passed through	Retained on	Initially	After 24 hr.	After 72 hr.	Initially	After 5 days
		(%)	(%)	(%)	(%)	(%)
	80	3.7	3.2	3.2	3.4	3.05
80	100	3.4	3.3	2.8	3.2	2.85
100	140	3.1	3.0	3.2	3.05	2.85
140	160	3.0	3.1	3.2	3.0	2.8
160	180	2.9	3.1	3.0	2.7	2.8
180		2.3	2.9	2.8	2.5	2.75

[a] Moisture content on original composite 3.0%.

tion of the moisture among the particles in mass gradually occurs. The difference in the time required to dry various sized particles must be considered not only in the design of dryers, but also as a factor in dryer operation, since heat damage during drying is determined by moisture content as well as temperature of the particle and the time of exposure.

The temperature of a droplet of water evaporating in a spray dryer would correspond to the wet-bulb temperature of the air. This, of course, is determined by the dry-bulb temperature of the air and its humidity. A droplet of milk would be at a somewhat higher temperature since materials in solution and suspension in milk lower the vapor pressure. This effect is greater than might be expected, as Ranz and Marshall *(3)* have shown that a droplet evaporates with a surface temperature corresponding to that of the saturated solution even though the droplet concentration is less than saturation. Nevertheless, during the initial stages of drying and under maximum conditions of temperature and humidity, the temperature of the drop will be nearly that of the wet-bulb thermometer, which will not exceed 130° to 140° F.

The temperature of the particle increases rapidly during the falling rate period. In strictly cocurrent flow dryers the maximum temperature of the particle will be essentially that of the dry-bulb temperature of the exit air with which it is in contact. If flow were strictly counter-current, the temperature of the nearly dry particle would approach that of the dry-bulb temperature of the hot inlet air. No spray dryers are strictly counter-current, although some involve at least partial counter-current flow. In these dryers the particles during the falling rate period come into contact with the hot incoming air. Thus, the temperature of the particles during the later stages of drying may be considerably higher than in cocurrent flow dryers. Recycling of the partially dried particles back into the hot gases even in cocurrent dryers also must be considered in evaluating the extent of heat exposure.

The driving force for the drying action is p_w-p_a where p_w and p_a are the vapor pressure of the water associated with the milk particles and air respectively. In cocurrent flow dryers p_w should approach p_a as the powder particles are removed from the exit air. Only the surface of the particles, however, ever attains approximate equilibrium with the drying air. Kitzes *(2)* by actual vapor pressure measurements has shown that the vapor pressure of the water associated with the milk powder particles increases on storage so that after holding for 2 weeks to permit moisture equilibration, the vapor pressure is considerably higher than that of the exit air from the dryer. This difference would be greater as particle size increases since, as previously pointed out, large particles have a higher moisture content at any given stage during drying than small particles.

Logarithmic plots of the vapor pressure of the water above dry milk against the vapor pressure of pure water at the same temperature give straight lines as shown in Figure 1, taken from Kitzes *(2)*. Dryer operators commonly regulate the moisture content of powder by varying the exit air temperature. The water-holding capacity of air as measured by the vapor pressure of water in air over water increases semi-logarithmically with increase in temperature, whereas the partial pressure of water vapor itself increases only in proportion to the absolute temperature. Thus, p_w-p_a, under any given set of conditions, increases with increase in temperature, resulting in a lower moisture powder.

Kitzes *(2)* has shown that in a cocurrent flow dryer the moisture content of the powder increases with increase in the exit air humidity under any given set of drying conditions. This relationship is shown in Figure 2. Since the exit air humidity is attributable to the inlet air humidity plus the water evaporated from the milk, plots of the inlet humidity and moisture content of the powder show a similar relationship (Figure 3). The differences in moisture content attributable to air humidity as shown on these charts are probably greater than would be observed in commercial practice. Commercial units would rarely, if ever, operate over such a wide humidity range, and any change in operating conditions affecting particle size

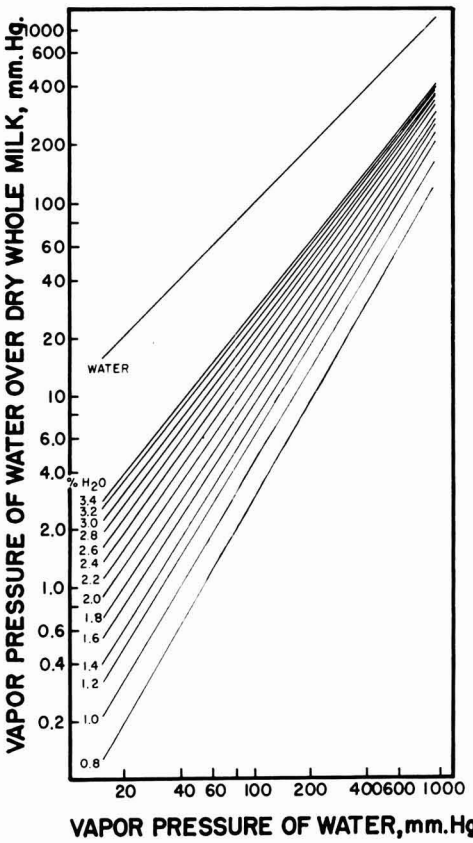

FIG. 1. Relationship between vapor pressure of water over dry whole milk and the vapor pressure of water.

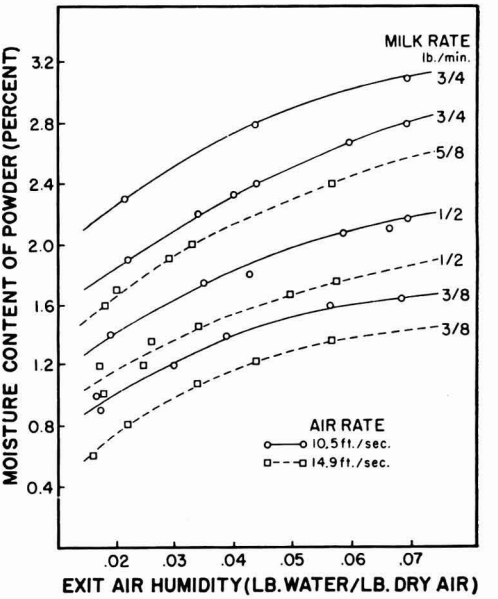

FIG. 2. Effect of exit air humidity on the moisture content of dry whole milk.

would tend to obscure the effect of humidity. The use of a redryer would further obscure the relationship.

One of the major advantages of spray drying is that heat damage may be held at a low level. Heat damage in spray drying is evaluated largely in terms of increased solubility index but certainly is not limited to casein destabilization. Wright *(5)* has shown that the time required to precipitate 50% of the proteins in heating milk decreases rapidly with increase in concentration up to about 88% solids. A further increase in concentration, however, increases the stability of the proteins, and in milk powder of normal moisture content they are quite heat stable. This fact is of importance in the design and operation of dryers. Heat damage will be lowest under conditions which provide most rapid drying at the lowest temperature, particularly during the later stages of drying. Townley *(4)* has shown (Figure 4) that in cocurrent flow dryers the solubility index increases semi-logarithmically with increase in exit air temperature above a certain critical temperature. The critical temperature varies, depending upon the design of the dryer; the operating conditions, particularly with respect to those factors influencing particle size; and also with the heat stability of the milk. In dryers in which air-spray flow is partially counter-current, the inlet air temperature also would be a factor, since in these dryers the temperature of the particle as it passes through the concentration range of maximum heat lability will be higher than in a strictly cocurrent flow dryer. The time required for drying through the critical concentration range increases, of course, with increase in particle size.

Factors in Design

Spray dryers may be classified according to method of atomizing and as to arrangements for mixing the drying gas and the sprayed particles. Because of limitations of space, methods of atomizing will not be considered in this article. With respect to arrangements for mixing, dryers may be classified in the following categories:

1. Cocurrent or counter-current.
2. Updraft, downdraft, or horizontal.
3. Straight line or rotary flow.

Dryers have been designed to utilize most possible combinations. There are few objective data available for the evaluation of various design arrangements. Counter-current air-spray flow would impose severe limitations on the temperature of the inlet air in drying heat labile materials such as milk. Nevertheless,

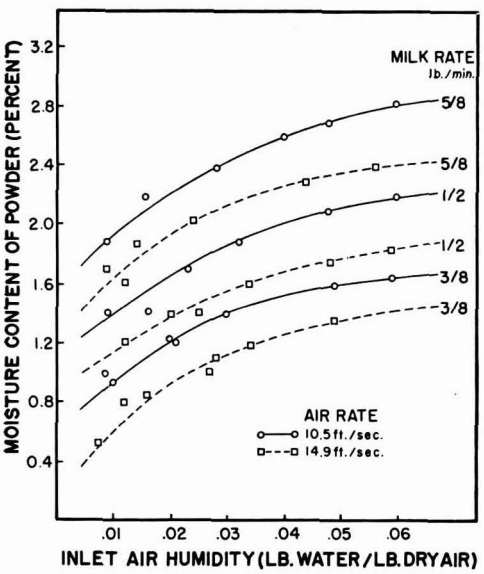

FIG. 3. Effect of inlet air humidity on the moisture content of dry whole milk.

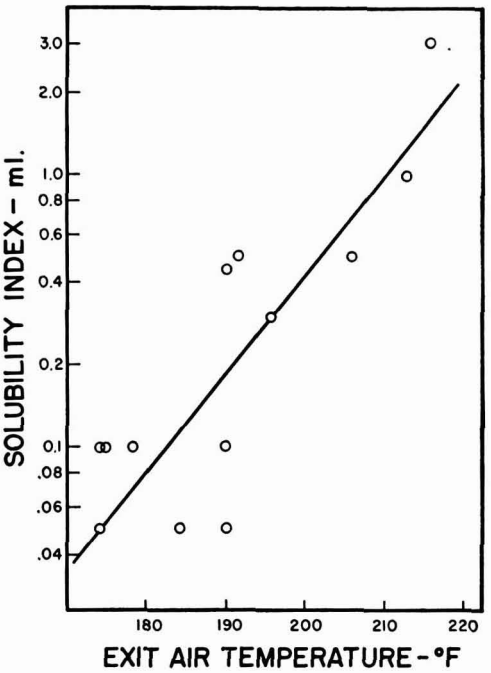

FIG. 4. The effect of exit air temperature on the solubility index.

dryers utilizing partial counter-current flow have been widely used in drying milk. The size of the powder particle has a material influence on the time the particle must remain in contact with the drying air. For lack of precise information this usually is evaluated primarily on a "cut and try" basis. The fact that widely different designs are successfully used is evidence that no one arrangement has outstanding advantages in all respects.

Operational Factors

The operator has the problem of producing the best possible product at the lowest cost with the equipment available. Quality as influenced by dryer operation is evaluated in terms of moisture content and heat damage (solubility index and presence of burned specks). The operator may regulate the inlet and outlet air temperatures and the particle size. The humidity of the inlet air may vary with weather, plant conditions, leaky coils, etc., but is rarely changed at the will of the operator.

Reasons of economy dictate the use of the highest possible inlet air temperature and the lowest possible outlet temperature. The inlet air temperature may be limited by the method used for heating or by the design of the dryer. Direct fire gas burners are becoming popular, since they permit heating to considerably higher temperatures than are practical with steam coils. Any design which provides partial counter-current air-spray flow or permits recycling of the partially dried material back into the hot air stream reduces the maximum air temperature which can be used without causing excessive heat damage. Some dryers are now operating at inlet temperatures of about 500° F. for drying milk and at much higher temperatures with less heat-sensitive materials. The difference in temperature of the air entering and leaving the dryer is primarily the result of moisture evaporation; therefore, other factors remaining constant, the higher the inlet temperature the higher the humidity of the exit air. Since the exit air humidity influences the moisture content of the powder in cocurrent flow dryers, there is a practical limit to the inlet air temperature even though heat damage of the product is not a factor. This limit would vary with the design of the dryer and the product being dried.

Dryer operation commonly is controlled to secure an exit air temperature which will yield the desired moisture content without exceeding permissible limits for heat damage. Liquid feed may be altered to secure the desired exit air temperature at an essentially constant inlet air temperature, or the inlet temperature may be varied to adjust for minor changes in liquid feed or concentration. The latter method is most practical with direct fire in dryers in which the inlet temperature is not a critical factor in powder quality.

There has been a demand for products of lower moisture content in recent years in recognition of the inverse relationship between moisture content and storage stability. This has

added to the difficulty of dryer operation, particularly in drying highly heat-sensitive materials, such as whole milk at times when the air humidity is high. When exit air temperature cannot be raised sufficiently to bring the moisture content within permissible limits without exceeding the critical temperature for heat damage, the simplest recourse is to reduce particle size. Reduction in particle size lowers the moisture content, since under any given set of drying conditions the moisture content decreases with decrease in particle size. It also reduces heat damage since the drying time is presumed to be proportional to the square of the diameter of the particle. First change in reducing particle size may be the usual manipulation of spraying pressure and orifice diameter. Under severe conditions and with other types of atomizers, it may be necessary to reduce the viscosity of the condensed milk at the time of atomizing. This may be effected by increase in temperature or reduction in concentration.

Obviously, an increase in temperature may not be possible as heat exposure may already be approaching critical limits. Reduction in concentration very effectively decreases particle size even at the lower spraying pressure which may be necessary with fixed inlet air temperature to compensate for the increased water content.

Quality requirements may be expected to become still more stringent if dry milk products are to enjoy increased usage, particularly at the household level. The ideal is a reconstituted product which is indistinguishable flavorwise from the original. Heat damage is difficult to avoid, even in spray drying. Minimum moisture content is desired for maximum storage stability. Attainment of low moisture without heat damage is the goal of dryer design and operation. Progress still is to be made. Research on the fundamentals of spray drying may point the way.

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Streptococcus Lactis and the Streptococci of the So-called Lactic Group

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From the nature of the problem, any attempt to divide bacteria into more or less homogeneous aggregates that may be designated as "species" must of course be a rather arbitrary procedure. Be that as it may, every bacteriologist is engaged in the classification of bacteria; that is, he must deal with biological divisions containing closely related organisms, whether these entities be designated as species and varieties, or as groups and types.

However, I wish to make it clear at the outset that I have no fixed ideas on the subject of species in the lactic group of streptococci. I have no quarrel with those who wish to consider the group as consisting of one species, *Streptococcus lactis*, with its several varieties. Since, on the other hand, I spent much time twenty years ago helping to "prove" that Orla-Jensen's *S. cremoris* is a distinct species, I naturally cannot disagree very violently with those who wish to recognize that and other types as being entitled to rank as species.

But before we take our views on this subject too seriously, it might be well to recall the oft-quoted statement of Duclaux:

> The species name has only the value of a name on a package; we must always be ready to change it or to do away with it.

However clear and secure our species may have looked 15 years ago, in view of the present findings in the field of microbial genetics with respect to mutations, sexual reproduction, and other mechanisms of genetic recombination, one can scarcely be so sure today. On the other hand, whatever may happen in the future to our taxonomic criteria, the better work of the past 35 years or more on the physiological and biochemical aspects of these organisms will retain its value. It is especially gratifying to note that the work of the past 15 years on the newer aspects of bacteriology has on the whole extended and confirmed the work of the earlier years.

Although students of *S. lactis* and its relatives among the so-called lactic streptococci have sometimes reported results that vary slightly in detail, these variations in results in the vast majority of cases are accounted for by slight differences in technique, as Abd-El-Malek

and Gibson *(1)* have indicated, the basic facts being verified by practically all investigators. Far from seeing any great disparity in the results of most workers, I am particularly impressed with the fact that the same basic conclusions have been reached among the majority of students of the streptococci based upon works that involved extensive physiological and biochemical reactions, nutritive requirements, metabolic processes, and serological properties. Whatever the approach, these diverse studies, from various parts of the world, have supplemented and confirmed one another.

Streptococcus lactis

S. lactis, which was isolated from milk and studied by Lord Lister and reported by him in 1873 and 1878, is the first streptococcus that was described. The fact that he gave it the name of *Bacterium lactis* is entirely logical in that the generic name *Streptococcus* had not been proposed at that time. Even though the species could not be differentiated from some other streptococci by the methods he used, there can be little doubt that Lister really isolated and studied *S. lactis.*

Some early investigators identified *S. lactis* by its ability to reduce litmus in litmus milk cultures before the milk was curdled. It is now known that this property is correlated with the production of a low oxidation-reduction potential, or, in other words, a strong reducing action. On the other hand, reduction of litmus after the milk is curdled is of little differential value, as such reduction is in considerable part caused by the reducing action of the milk itself after the elimination of convection currents following the formation of the curd.

This simple test, on the whole, served very well the few bacteriologists who used it on streptococci from milk and sour milk products. They were wrong, however, in thinking that *S. lactis* is the only streptococcus that has this characteristic, which is shared by a majority of strains of *S. faecalis* and some other varieties of the enterococcus group. There can be little doubt, in view of present knowledge, that many cultures that were isolated from cheese, especially later in the ripening process, that were identified as *S. lactis* were in fact *S. faecalis.*

In 1918, the lactic-acid streptococcus was defined as having, in addition to other characteristics, a strong reducing action, a minimum temperature of growth below 10° C., and a maximum growth temperature below 45° C. This combination of characteristics still appears to be unique to the *S. lactis* group, though of course much more is now known, including nutritive requirements, serology, and other physiological properties.

In America, there has been no confusion about the identity of *S. lactis* and its differentiation from other groups of streptococci for more than 20 years, and as new techniques and criteria have become available the differences between the "lactic" streptococci and other groups and species within the genus have become more clearly and completely defined. On this occasion there is not time to discuss in detail the characteristics of *S. lactis,* nor is this necessary, as these details have been reviewed and published many times during the past two decades.

Perfectly correlated with the physiological characteristics are the serological findings. That *S. lactis* has a group-specific antigen and that the so-called lactic streptococci constitute a distinct serological group was established independently in England *(8, 12),* in Germany *(11),* and in America *(14),* but our English colleagues are the ones who have done the most extensive and thorough work on this subject.

In 1955, as opposed to 15 years ago, a full consideration of the species should take into account what is known about the nutrition and metabolism of the organisms, the antibiotics they produce, and bacteriophage phenomena, but these subjects are reserved for others on this program.

In a consideration of a group of microorganisms and the species and varieties that it contains, attention should be given to the occurrence of these organisms in nature and especially their probable natural habitats. In work done in our own laboratory years ago, authentic strains of *S. lactis* were isolated from various plant materials and consistently from a number of these that were tested over and over, but it was not possible at that time to demonstrate these organisms in large numbers from such sources. Since this group of organisms has not been shown to be animal parasites or consistently associated with any animal source, the habitat associated with vegetation appears to be the most logical one based on present knowledge. This is especially true of *S. cremoris,* which has a very low maximum temperature of growth, a large proportion of strains failing to grow at the body temperatures of higher animals. In this connection, also, it should be noted that the species or variety that has been named *Streptococcus diacetilactis* was first isolated from fermenting vegetable materials and, on the basis of present information, should probably be considered as being associated with plants in nature.

Although many of us may be strongly inclined to the point of view that the lactic-acid streptococci had evolved to their approximate present status long before there was any organized dairy industry to furnish milk as a more or less continuing natural habitat, Hirsch *(5)* suggests that the evolution of these organisms may be so recent that milk may be considered their true habitat. In fact, Hirsch considers *S. lactis* and *S. cremoris* as probably being not only quite distinct species, but as perhaps having different lines of descent in their courses of evolution.

As an illustration of the fact that we must

keep our ideas somewhat flexible with respect to what we think we know about bacteria is the possibility that the lactic-acid streptococci may be very rarely associated with infections. It is now known that the early reports of such pathogenicity were based upon inadequate identification of the organisms used. More thorough studies with authentic cultures of *S. lactis* in later years showed that virulence could not be easily developed in cultures of this organism by serial animal inoculations, as had been claimed by a few earlier workers. Note should therefore be made that Wagner *(16)* has reported two instances of *S. lactis* being associated with human infections, and Wood *et al. (17)* have reported the recovery of *S. lactis* from the blood of a patient with subacute bacterial endocarditis. In both of these cases, it is stated by the authors that the organisms recovered agreed with the description of *S. lactis* on the basis of a large number of physiological and biochemical reactions, and in each case extracts of the organisms were found to react with group serum prepared against *S. lactis*. In view of what we know about the rare occurrence of many ordinarily nonpathogenic bacteria in human and animal infections, this should not come as too much of a shock, however distasteful it may be to some of us to think that *S. lactis* should ever invade the human body.

Streptococcus cremoris

Whether considered as a species or a variety, the *S. cremoris* of Orla-Jensen *(10)* deserves special consideration. As opposed to the typical *S. lactis, S. cremoris* fails to grow at 40° C., and many strains do not grow at 37° C. *(13, 18)*. It does not hydrolyze arginine or does so very feebly *(1, 9)*. It is also inhibited at lower concentrations of sodium chloride, alkali, and other inhibitory substances than is *S. lactis*. As grown in the laboratory, it is in general a more delicate organism than is the typical *S. lactis* and ferments fewer substances, most strains failing to ferment maltose. Orla-Jensen considered the inability to ferment maltose and dextrin as being especially valuable criteria in the differentiation and identification of *S. cremoris*. On the morphological side, strains of *S. cremoris* in general form longer chains and are inclined to have somewhat larger cells than does *S. lactis*.

It should be noted that those who have studied the nutrition and metabolism of the lactic group have detected slight differences between *S. lactis* and *S. cremoris*, and the English investigators have shown that these two organisms are also different on the basis of the antibiotics they produce.

Despite the apparently imposing list of differences between *S. cremoris* and *S. lactis*, on the basis of serological grouping their relationship is close. All of those who have produced group-specific sera against *S. lactis* have obtained presumptive evidence that *S. cremoris* belongs to the same group, based on the finding that extracts of *S. cremoris* reacted with anti-*lactis* group sera. However, it appears to have remained for Briggs and Newland *(3)* to produce a "clean" anti-*cremoris* grouping serum, which proved to be specific for the whole lactic group. These organisms are, therefore, exceedingly close relatives from the serological point of view, just as they are with respect to their nutritive requirements and most of their more basic physiological characteristics.

Most cultures of *S. cremoris* have been isolated from commercial starters, and the view that it is therefore an adaptive mutant within the lactic group is a natural and attractive one. On the other hand, strains of *S. cremoris* have been isolated by several workers directly from raw milk in such widely separated areas of the world as the eastern part of the United States, the western part of Canada, the British Isles, and the mountains of France.

It was the opinion of Orla-Jensen that *S. cremoris* is superior to the more typical *S. lactis* as a starter organism, but this view has not been shared by some workers in America and elsewhere. On purely physiological grounds, because of its low tolerance to salt and a low maximum temperature of growth, *S. cremoris* would not appear to be a suitable organism for use as a starter in the making of cheese. However, opposing opinions have been expressed on this question.

Citrate-Fermenting Lactic Streptococci

Innumerable variant types of lactic streptococci have been reported during the past 35 years, and many names of both species and variety rank have been applied to such strains. If an additional species were to be recognized at the present time, probably the best case could be made for that variety, or group of varieties, which is characterized by the ability to ferment citrates actively with the production of carbon dioxide, volatile acids, and other volatile products. Not to be confused with the heterofermentative cocci of the *Leuconostoc* group, these organisms are homofermentative lactic-acid streptococci in their action on glucose and resemble *S. lactis* in their general physiological characteristics.

Krishnaswamy and Babel *(7)* have made an excellent contribution to this subject, and they are to be commended for not having given us another species name with which to contend. Sensibly, I think, they considered the cultures they studied as being entitled to rank only as a variety within the lactic group, and they suggested the name of *S. lactis* var. *aromaticus*.

Swartling *(15)* has concluded that many of the species and variety names that have been applied to streptococci of this type are in fact synonyms, and in his opinion *S. diacetilactis* is the name that has the best claim for retention on the bases of priority and validity, if a

species is to be recognized. Swartling has isolated a number of strains of this organism from milk and milk products and has found it to be a homofermentative streptococcus in the fermentation of sugars, which produces the *dextro*-rotatory form of lactic acid. However, it produces carbon dioxide, volatile acids, acetylmethylcarbinol, and diacetyl from citrates. This organism does not appear to show any relationship to the enterococci, on the one hand, nor to the heterofermentative *Leuconostoc* group on the other.

Swartling *(15)* and Briggs *(2)* have shown *S. diacetilactis* to agree with *S. lactis* in its general physiological reactions and to belong to the same serological group as do *S. lactis* and *S. cremoris,* the group N of the English workers.

Similar work has been done by Kneteman *(6)* on the *S. citrophilus* of van Beynum and Pette, which appears to be the same as the *S. diacetilactis* of Matuszewski *et al.*, as Swartling has shown. Of especial interest is the finding of Kneteman that variants were obtained that were unable to ferment citrate and were like "ordinary lactic acid streptococci from which they cannot be distinguished . . ." In other words, mutations occurred that gave rise to apparently typical strains of *S. lactis.*

Whether *S. diacetilactis* (and its relatives) should be considered a distinct species or as a variety in the general *S. lactis* group is a matter of opinion at the present time. This writer does not feel competent to express an authoritative opinion and is quite willing to abide by the decision of those better qualified to judge. However, the mutations reported by Kneteman and previous workers would indicate that definite conclusions might well be delayed for the present. Such caution appears to be further supported by the reports of variant and intermediate types by several other investigators *(4, 7, 19).*

In conclusion, an explanation and an apology should be offered. No specific citations have been made to the literature of more than 20 years ago, with the exception of one to the work of Orla-Jensen. This has excluded such outstanding and pioneering contributions to the lactic streptococci as those made by Hammer and by the late S. Henry Ayers and their associates, who recognized a number of varieties of *S. lactis,* as well as making many other important contributions to this and other groups of streptococci. And, to keep this short discussion from being a mere listing of references, the review of the more recent literature is "spotty" and incomplete.

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ABSTRACTS OF LITERATURE

W. O. Nelson, Abstract Editor

ANIMAL DISEASES

667. Oxytetracycline in bovine mastitis. I. Treatment of mastitis. L. E. BARNES, Iowa State Coll., Ames. Am. J. Vet. Research, **16**, 60: 386. 1955.

Cows in the Iowa station herd were routinely tested by strip cup, Hotis test, pH of milk, catalase test, Whiteside test and bacteriological culturing to determine cases of clinical and subclinical mastitis and the causative organisms. Both types of cases were treated by udder infusions of 400 mg. of oxytetracycline (terramycin) every 24 hr. for 3 days. 82.7% of *S. uberis* infections were eliminated for three weeks or more following treatment, compared to 33.3% of *S. dysgalactiae,* 25% of *M. pyogenes* var. *aureus,* 69.2% of coliforms, and none of *P. aeruginosa.* Micrococcus cases of more than one mo. duration were not as effectively cured as recently acquired infections, but the duration of the *S. uberis* infections didn't alter recovery efficiency. In bacteriostatic and bactericidal *in vitro* tests it was found that *S. uberis* and *M. pyogenes* var. *aureus* were both inhibited by 1 to 3 μg. per ml., but were not killed. *S. uberis* was killed by 10 to 20 μg. per ml. but *M. pyogenes* var. *aureus* required over 200 μg. per ml. In some cows the treatments produced definite irritation and inflammation of the glands. Nearly all acute cases showed clinical improvement after treatment even when the infection persisted. E. W. Swanson

668. Congenital porphyria in swine and cattle in Denmark. S. K. JORGENSEN and T. K. WITH. Nature, **176**, 4473: 156. 1955.

This condition, found at times in pigs and cattle, impairs the quality of meat for market purposes. Teeth, bones, etc. are discolored because of the presence of porphyrin. The health of animals subject to this condition is not seriously impaired. The heredity of the condition is obscure but is being studied in Thisted County in Jutland, where it is found in both swine and cattle. Animals having the condition may be detected by examination of the color of the teeth and analyzing for porphyrin in the urine and faeces. R. Whitaker

669. An immunogenic agent for the protection of cattle against Leptospira pomona. W. G. HOAG and W. B. BELL, Va. Agr. Expt. Sta., Blacksburg. Am. J. Vet. Research, **16**, 60: 381. 1955.

A soluble antigen was prepared by acid-heat extraction of cultures of *Leptospira pomona,* preserved with merthiolate and suspended in mineral oil. Its immunogenic efficiency was tested on calves which were later challenged. All control calves developed typical symptoms of leptospirosis and had positive kidneys up to 53 days later. None of the vaccinated calves developed disease symptoms and all had normal, negative kidneys 53 to 55 days after challenge, as well as continuous negative urine and blood tests. The effectiveness of the vaccine against other strains of leptospira has not been checked. E. W. Swanson

670. Parasitological significance of bovine grazing behavior. J. F. MICHEL. Nature, **175**, 4468: 1088. 1955.

As a preliminary step in a study of the habit of cows not eating grass growing in the vicinity of bovine faeces, it was shown that cows grazing on pasture infested with lungworm *(Dictyocaulus viviparus)* larvae were highly selective in grass eaten from a parasitological point of view. This behavior was shown to be due to something other than the difference in grass size caused by increased fertility of the earth adjacent to droppings. R. Whitaker

BUTTER

671. The detection and quantitative determination of coloring materials in butter and margarine. H. M. ESPOY and H. M. BARNETT, Barnett Lab., Long Beach, Calif. Food Technol., **9**, 8: 367. 1955.

A procedure was developed for the separation, identification, and quantitative estimation of the principal pigments found in commercial butters and margarines without saponifying the fats. Carotene and annatto extract were obtained by chromatographic separation while the "certified" coal tar dyes were removed by acid extraction. The recovered pigments were dissolved in suitable solvents, spectrographic readings were made on the solutions and the concentration of the individual pigments in the butter or margarine was calculated from these readings. The concentration of the different pigments found in 47 samples of commercial butter and 8 samples of margarine, analyzed by this method, is shown. It is postulated that this procedure probably is applicable also to other foods containing oil soluble pigments. E. R. Garrison

672. Combined butter dish and slicer. G. LERNER. U. S. Patent 2,716,814. 5 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 24. 1955.

A butter dish for holding a quarter-pound print and having a cover equipped with strips which slice the butter when the cover is pushed down over the print. R. Whitaker

CHEESE

673. Factors involved in the control of gelatinous and defects of cottage cheese. E. B. COLLINS, Dept. Dairy Ind., Univ. of Calif., Davis. J. Milk Food Technol., **18**, 8: 189. 1955.

Chlorine was effective in destroying cultures of *P. fragi, P. viscosa, A. metalcaligenes,* and *E. coli* in concentrations ranging from 3 to 5 p.p.m. of residual chlorine and at pH 6.0. Low temperature and high pH decreased the effectiveness of chlorine. *P. fragi* was more resistant than the other organisms studied. Doubling the concentration of residual chlorine decreased the time for destruction of *P. fragi* by 50%. H. H. Weiser

674. Method and apparatus for continuous cheese manufacture. B. T. HENSGEN, J. C. VANDEN BOSCH, A. M. LEDERER, and P. M. WOOD (assignors to Swift & Co.). U. S. Patent 2,717,212. 7 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 123. 1955.

Coagulated milk is centrifuged to remove the whey. The curd is then held in the rotating bowl at reduced speed to permit cheddaring of the curd. R. Whitaker

675. Adjustable tension wire type cheese cutter. J. P. MULHALL (assignor to Irving Rubin). U. S. Patent 2,714,251. 7 claims. August 2, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 1: 19-20. 1955.

A hand-operated cutter for slicing cheese for table use. R. Whitaker

CONDENSED AND DRIED MILKS; BY-PRODUCTS

676. Attachment for punching and for clearing pouring openings in can tops. C. C. HART. U. S. Patent 2,716,808. 3 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 23. 1955.

A device for punching a pouring hole in evaporated milk cans, consisting of a frame which fits over the top of the can and a conical shaped punch mounted on a pivoted spring which may be rotated around the circumference of the can top. R. Whitaker

677. Baby food. H. A. CRANSTON (assignor to Carl S. Miner). U. S. Patent 2,717,211. 15 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 123. 1955.

A baby food containing whole milk solids and concentrated pathogen-free mammary gland phosphatase. R. Whitaker

678. Food whipping agent. B. A. PATTERSON. U. S. Patent 2,716,606. 8 claims. Aug. 30, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 5: 652. 1955.

An alkali metal caseinate, lactose, and phosphate blend at pH 6.8 to 7.2 is used as a whipping agent for cakes, pies, meringues, etc. R. Whitaker

679. Casein contact printing emulsion. J. M. LUPO, JR. (assignor to Direct Reproduction Corp.). U. S. Patent 2,716,061. 2 claims. Aug. 23, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 4: 505. 1955.

Details are given for the formulation, manufacture, and application of a casein base photographic emulsion to glass and plastic sheeting. R. Whitaker

680. Hog food. P. B. SHEARER. U. S. Patent 2,716,063. 9 claims. Aug. 23, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 4: 505. 1955.

A feed for pigs consisting of cheese whey neutralized with an ingestible reactive calcium compound. R. Whitaker

681. Comestible and comestible base and method of making the same. H. M. LEVIN. U. S. Patent 2,715,068. 9 claims. Aug. 9, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 2: 234. 1955.

Dried nonfat milk solids, reconstituted, acidified, and pasteurized, is blended with mayonnaise to make a salad dressing–like material. R. Whitaker

682. Diacasein and process for its manufacture. F. W. BERNHART, E. R. ECKHARDT, M. H. JANSON, and F. H. TINKLER (assignors to American Home Products Corp.). U. S. Patent 2,714,068. 10 claims. July 26, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 4: 522. 1955.

A dry, neutral, high protein, low ash, dispersible, slow settling phosphoprotein is made from skimmilk by coagulating with rennet, cutting the curd, draining the whey, pressing the curd, then acidifying, washing, neutralizing, heating, agitating, drying, and finally grinding to a powder. R. Whitaker

683. Ice box dessert. G. H. STUART, J. T. WATSON (assignors to the Borden Co.). U. S. Patent 2,714,069. 4 claims. July 26, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 4: 523. 1955.

A casein lactalbumin blend prepared in a soluble nonheat-denatured form to make a cheese-like product of high water absorptive properties, is suspended in an aqueous solution of gelatin. A whipping agent is added and the mixture whipped. The mixture sets on cooling in a refrigerator as a result of the absorptive action of the curd particles for the moisture in the blend and forms a dessert having a consistency suitable for eating with a fork. R. Whitaker

DAIRY BACTERIOLOGY

684. The isolation and classification of proteolytic bacteria from the rumen of the sheep. J. C. APPLEBY, Rowett Research Inst., Bucksburn, Aberdeen. J. Gen. Microbiol., **12**, 3: 526. 1955.

Proteolytic bacteria were obtained from the rumens of three fistulated sheep using a mineral salts–yeast extract or rumen fluid–casein medium containing a variety of additional supplements. On this solid medium proteolytic organisms were considered to be those which produced a zone of clearing. Total viable counts obtained from the rumen contents appeared to be somewhat low, of the order of 10^5 to 10^7 per milliliter of rumen fluid. Predominating organisms consisted of gram positive cocci with smaller numbers of bacilli; however, some gram negative rods were observed when medium variations were introduced. A number of cultures of *Bacillus* species, *Clostridium* species, *Flavobacterium*, *Proteus* and micrococci were isolated and described. J. J. Jezeski

685. The preservation of lactobacilli by freeze drying. M. BRIGGS, G. TULL, L. G. M. NEWLAND, and C. A. E. BRIGGS, N.I.R.D., Univ. of Reading. J. Gen. Microbiol., **12**, 3: 503. 1955.

Cultures were grown anaerobically on tomato-glucose-Tween agar and suspended in several media for freezing. After freezing, these were dried in a desiccator over P_2O_5. On the basis of percentage survival rate, the main factor influencing survival was the suspending medium and best results were obtained in horse serum plus 8% added glucose. With the majority of the 44 cultures tested, the survival rates were in excess of 50% immediately after drying with 25% survival after 6 mo. storage. Several cultures survived as long as two years under these experimental conditions.

J. J. Jezeski

686. The physiological and serological characters of freeze dried lactobacilli. M. E. SHARP and D. M. WHEATER, N.I.R.D., Univ. of Reading. J. Gen. Microbiol., **12**, 3: 513. 1955.

A study of 41 strains of lactobacilli showed that the physiological and serological characteristics of these organisms did not change during the course of freeze drying or after storage for 6 mo. J. J. Jezeski

687. Sur l'emploi de différents indicateurs d'oxydo-réduction pour l'étude des bactériophages des streptocoques lactiques (Use of different oxidation-reduction indicators for the study of bacteriophages of Streptococcus lactis). E. VALLES. Le Lait, **35**, 242. 1955.

A quantitative method of determining the effect of bacteriophages is described. The method makes use of serial dilutions and the oxidation-reduction indicators litmus, methylene blue and resazurin. The method is used in a study of concentration and purification of bacteriophages and their specificity for *S. lactis*. Comparisons are made to the other methods.

A. W. Rudnick, Jr.

688. Influences respectives de la propreté des utensiles et du refroidissement après la traite sur la qualité bactériologique du lait cru (Respective influence of cleanliness of utensils and refrigeration after milking on the bacteriological quality of raw milk.) A. PORTMAN, Central Microbiol. and Dairy Research Sta. Le Lait, **35**, 132. 1955.

Evidence is presented to show that clean utensils and immediate cooling are necessary to reduce bacterial growth in milk until the product is delivered to the processing plant. Initial bacterial counts were high according to United States standards. An attempt is made to calculate the amount of cooling water needed to adequately cool milk in several types of coolers. The article is obviously written for producer and fieldman use.

A. W. Rudnick, Jr.

689. The nature, significance and control of psychrophilic bacteria in dairy products. J. C. OLSON, R. B. PARKER, and W. S. MUELLER, Univ. of Minn., St. Paul; Ore. State Coll., Corvallis; and Univ. of Mass., Amherst. J. Milk Food Technol., **18**, 8: 200. 1955.

The authors use the term psychrophile to indicate any bacterial species capable of growth at temperatures ranging from 35° to 45° F. They emphasize the importance of these organisms as they affect the keeping quality of dairy products stored at low temperatures.

These organisms can be controlled by proper pasteurization, proper cleaning and sanitizing of all equipment, and chlorination of water supplies to be used in the manufacture of butter and cottage cheese. Usually 5 to 10 p.p.m. of available chlorine is adequate.

H. H. Weiser

DAIRY CHEMISTRY

690. Occurrence of different beta-lactoglobulins in cows' milk. R. ASCHAFFENBURG and J. DREWRY. Nature, **176**, 4474: 218. 1955.

In a study made of the milk of 70 cows, it was found that two kinds of lactalbumin are secreted in the milk. These have been designated as B_1 and B_2, and when both are found, $B_{1,2}$. The type is characteristic for a given cow. Thirty of the cows gave mlik containing $B_{1,2}$; 7 gave B_1; and 33 gave B_2. The $B_{1,2}$ and B_2 were found in the milk of Shorthorn, Friesian, Ayrshire, and Guernsey cows, whereas B_1 was found only in the Shorthorn and Friesian milk. The milk from one-egg twin heifers was alike in the four sets studied, and all three types of lactalbumin were found. B_1 and B_2 lactoglobulin differ in several respects: crystalline form, isoelectric point, and solubility, for example. The two forms were identified electro-

phoretically. When $B_{1,2}$ milks were encountered, in most instances more B_1 was found than B_2. The existence of the two distinct types of lactalbumin has been overlooked in the past as workers have used mixed milk but have been puzzled by the heterogeneity of this "pure" protein. R. Whitaker

691. Lactase enzyme preparation. E. R. MORGAN (assignor to National Dairy Research Laboratories, Inc.). U. S. Patent 2,715,601. 4 claims. Aug. 16, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 3: 379. 1955.

A lactase-active, zymase-inactive product is prepared from a lactase-active yeast by treating the yeast with 1.5 to 3.6 times its weight of ethyl alcohol to kill the yeast cells and then drying to a powder. R. Whitaker

692. Difficulty in the crystallization of rennin. N. J. BERRIDGE, N.I.R.D., Univ. of Reading. Milchwissenschaft, **10**, 195. 1955.

Rennin in certain cases was found to be contaminated with impurities which were difficult to remove. This is particularly true when the enzyme is prepared from fresh calf stomachs. It was found, in the course of this study that electrophoresis may be used to advantage in purifying the enzyme. In M/10 phosphate buffer at pH 6 the fast moving component was pure rennin which formed crystals at 2° C. after the solution was acidified to pH 5.4 and 40 mg. NaCl was added per ml. J. Tobias

693. Études sur la caséine. II L'homogénéité de la casein. (Study of casein. II Homogeneity of casein.) M. BEAU. Le Lait **35**, 258. 1955.

The author suggests that the variations in composition of α, β, and γ casein reported in the literature may be caused by the methods of extraction of casein. It is suggested that extraction with HCl and NaOH is too drastic and that investigators should use physical or mechanical means of extraction.

A. W. Rudnick, Jr.

DAIRY ENGINEERING

694. The control of stainless corrosion. Staff Editors. Milk Plant Monthly, **44**, 8: 37. 1955.

The causes of failure in stainless steel are: (1) Pitting caused by mechanical damage, dirt, porous welds, or chlorides, (2) Crevice corosion caused by incomplete weld penetration, (3) Inter-granular corrosion at the grain boundaries, (4) Erosion-corrosion, a combination of mechanical wear and corrosion, and (5) Stress-corrosion, a cracking of stainless steel under stress. Practices recommended for the control of stainless steel corrosion are discussed.

C. J. Babcock

695. Symposium on plastics as materials of construction. Food processing plants. L. J. TURNEY, the H. W. Madison Co., Medina, Ohio. Ind. and Eng. Chem., **47**, 7: 1366. 1955.

The food industry is urgently in need of superior materials of construction. Coolers, mixing tanks, agitators, piping, and all materials of construction are subjected to damp, highly corrosive conditions, and it is believed that plastics in some forms, because of their resistance to corrosion and sweating, may serve well in these applications. Careful consideration must be given to the possibility of toxicity and contamination from plastic materials. A review of the properties of the presently available plastics indicates where they may be used with hope for success. Better machining methods as well as more complete handling knowledge are essential before a successful widespread use of plastics may be realized. The potential field for these materials is tremendous but suppliers must be conscious of the dangers of misuse, especially where toxicological data are incomplete. Such errors may do much harm to the food manufacturer as well as to the plastic suppliers. B. H. Webb

696. L'upérisation (Uperisation). P. LANIESSE, Natl. Inst. of Agron. Le Lait, **35**, 151. 1955.

A diagrammatic description of a Swiss high-temperature short-time sterilization process.

A. W. Rudnick, Jr.

697. Container traversing mechanism for food processing apparatus. J. H. BLAIR, JR. (assignor to Food Machinery & Chemical Corp.). U. S. Patent 2,717,548. 5 claims. Sept. 13, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 2: 216. 1955.

A method of continuously rotating and moving cans of liquid food products in a continuous-type of sterilizer. R. Whitaker

698. Electrical fluid treating apparatus. R. J. HERBOLD (assignor to Winger Dairy Products Processing & Manufacturing Corp.). U. S. Patent 2,713,818. 5 claims. July 26, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 4: 459. 1955.

Milk to be pasteurized, sterilized, or otherwise heated, is passed through a tube. The temperature is raised due to the resistance offered to an electric current which flows through the milk in the tube. Instead of electrodes in contact with the milk, the current is introduced through osmotic diaphragms which separate the milk from small reservoirs of electrolyte in which the electrodes are installed.

R. Whitaker

699. Automatic doors. ANON. Milk Dealer, **44**, 11: 39. 1955.

The installation of automatic cold storage doors at H. C. Christians Co., Chicago, Ill., and at the Supplee-Wills-Jones Milk Co. in Philadelphia is discussed. C. J. Babcock

700. Bubbles that do work. ANON. Milk Dealer, **44**, 11: 43. 1955.

Air agitation of milk is being used more in many milk pasteurizing plants. The advantages

are that milk in holding tanks can be mixed down to the last four inches, and that it is a quick process, functioning just as well whether the holding tank is long or short. Compressed air, oil-free, also may be used in other plant operations. C. J. Babcock

DAIRY PLANT MANAGEMENT AND ECONOMICS

701. Packaging's "private eye." ANON. Milk Prod. J., **46**, 8: 12. 1955.

The hidden camera has been used in today's modern supermarket to register consumer reaction where it counts most—at the point of sale. This technique has been used by a leader in the packaging field for the last four years to determine how and why people buy what they do.

Thus far, three hidden camera studies on bread, bacon, and frozen foods have been completed. The results of and conclusions from these studies are presented and discussed.

J. J. Janzen

702. Developing executives you can depend on. O. K. MCMAHON, Rohrer, Hibler & Replogle, Atlanta, Ga. S. Dairy Prod. J., **58**, 3: 32. 1955.

Failures in executive positions, in general, are much more often due to shortcomings in the executive's psychological qualifications than to lack of technical job know-how. Many who have been successful as individual technical workers or managers of small groups have been unable to cope with purely managerial duties in charge of large groups, a job for which they were not trained. The increase in size of businesses and in complexity of operation requires a type of executive personnel for which there is a real shortage. Special training to develop such men is needed.

The executive of today must be a person who can get a job done through others, is selfless rather than selfish, takes pride in what his organization does rather than in his personal accomplishments; is able to analyze himself and change his habits when necessary, intellectually competent, able to make up his mind and stick to it, emotionally stable and able to make fair decisions; sets good examples in behavior; has insight to organize and direct, ability to delegate and helps to develop and inspire his men as individuals.

F. W. Bennett

703. Sorge builds multiple quart sales with twin hanger. ANON. Milk Dealer, **44**, 11: 40. 1955.

Faced with aggressive paper ½ gal. competition, the Sorge Ice Cream and Dairy of Manitowoc, Wis., developed its own "twin hanger" which fastens two single paper qt. into a ½ gal. unit. The "twin hanger" operation functions as an integral part of the filling line. It is made of heavy paper and is specially treated to prevent the absorption of moisture. It measures 2⅝″ by 5⅜″, and folds in half to an approximate square, 2⅝″ by 2⅝″. It is centered between the two qt. containers and stapled to the cartons. A handy finger grip is perforated in the handle. Between the filling and stapling operations a turntable turns every other quart container to prevent one carton from having its "pitcher-pour" side clipped shut. C. J. Babcock

704. Milk consumption can be increased. H. SMITH and F. MADASKI, Mich. State Coll., East Lansing. Milk Dealer, **44**, 11: 49. 1955.

A recent study showed that when a choice of beverage was offered to people, 11% chose milk. Only three people drank milk when coffee was served to them. When people were served milk with their meals, 84% drank it. It was found that 25% of the agricultural people drank milk with their meal when given a choice as compared to 6% in the non-agricultural groups. Men drank more milk with their meals than did women. When agricultural and non-agricultural groups were served milk with their meal, 93% and 75%, respectively, drank it. There was no charge for the additional milk consumed. The study further showed that only 77% drank milk with their dinner, while 97% drank it with their lunch. Data are presented to show the variations in milk consumption with different methods of service, the level of milk consumption by men, women, and mixed groups, milk consumption by agricultural and non-agricultural groups, and milk consumption at lunch and dinner. C. J. Babcock

705. Training your clean-up personnel. L. G. DORMUTH, Penn. Salt Mfg. Co., Philadelphia. Milk Plant Monthly, **44**, 8: 31. 1955.

To prevent off-flavors, high bacteria counts and other quality defects, milk solids, butterfat, and milk stone must be completely removed from all milk contact surfaces. In addition, foreign matter such as rust, scale, and oil may not be allowed to accumulate on heat transfer surfaces. Experience indicates that quick and thorough cleaning improves the efficiency of the processing operation and extends the life of costly equipment. The importance of cleaning and sanitizing is, therefore, rarely questioned. This highly important job, however, is often relegated to an organization's least experienced employees with little training and no understanding of what is expected of them. The results are lower quality and higher processing costs for the company, and job dissatisfaction for the employee. The personal qualifications of the clean-up crew must satisfy high standards of personal cleanliness, intelligence, and reliability. Only through skillful training and guidance can a supervisor expect to develop these qualifications and have a satisfied crew of technically skilled clean-up men who know what they are doing and why. C. J. Babcock

FEEDS AND FEEDING

706. Economic efficiency in pasture production and improvement in southern Iowa. E. O. HEADY, R. O. OLSON, and J. M. SCHOLL. Iowa Agr. Expt. Sta., Ames Research Bull. 419. 1954.

Much of the data included in this study was obtained by interviewing numerous farmers (122 in study #1, and 200 in study #2) in southern Iowa, as well as various contractors who were acquainted with costs of clearing land. All costs were based on 1951 prices.

Birdsfoot trefoil-orchardgrass and the lespedeza systems have a life expectancy of 20 years. Therefore, on a yearly cost basis these two systems cost $3.92 and $2.94 per acre respectively. The shorter term grasses cost as follows: alfalfa-bromegrass-ladino mixture $7.09, reed canarygrass-ladino mixture $6.55, and for the clipping and fertilization system $9.54. When costs are discounted at a 5% rate (market rate, which is applicable to farmers with unlimited capital), the long term grasses still give the lowest costs but the differences are smaller; birdsfoot trefoil-orchardgrass $2.34, alfalfa-bromegrass-ladino $2.38, reed canarygrass-ladino $2.49, and lespedeza $1.36. Clearing costs per acre ranged from $25 for light brush to several hundred dollars for timbered land. Lack of capital and insufficient stock were the two main reasons for not improving pastures. R. W. Hunt

707. Grazing and grass-silage feeding studies at the Northern Great Plains dairy station. R. F. GAALAAS and G. A. ROGLER. U.S.D.A., Washington, D. C. Tech. Bull. 1115. 1955.

The results of rotational grazing systems in which Holstein cows were pastured on crested wheatgrass, a grass-alfalfa mixture, sudangrass, and native grasses, are reported for the years 1949-1953. Also reported are results of feeding trials in which oat-pea silage, corn silage, and wildrye-sweetclover silage were fed to heifers and cows. The heifers were fed grass hay, and the cows grass hay and a concentrate along with the silage.

Use of adapted perennial and annual grasses, along with native grasses, in a rotational system lengthened the grazing season and improved the quality of the roughage. Milk production per acre from the rotationally grazed tame grasses was 50-100% more than from native grasses alone. The grazing season was 50-100% longer on the rotationally grazed tame grasses than the native pastures. Oats and Canada field peas grown together and stored as silage made excellent winter forage for milking cows and heifers. The yield per acre of oats and peas was equal to that of corn. A mixture of oats and Canada peas as a crop had several advantages as compared to corn. Wildrye-sweetclover would make good silage if the sweetclover could be maintained. In these experiments the yield of wildrye alone was too low to be satisfactory. R. W. Hunt

708. Use of distillers' grain solubles in calf starters. C. A. LASSITER et al. Ky. Agr. Expt. Sta., Lexington. Bull. 623. 1955.

Forty-four Jersey male calves were used to evaluate the use of distillers' dried solubles in calf starters. It was indicated that corn or milo distillers' dried solubles could replace an equal amount of dried skimmilk in the calves' rations. Rye distillers' solubles were only 85% as effective as the other solubles in promoting growth. None of the distillers' solubles, particularly rye, were considered as palatable as skimmilk. R. W. Hunt

709. Antibiotic studies in young dairy calves. C. A. LASSITER et al. Ky. Agr. Expt. Sta., Lexington. Bull. 624. 1955.

Part A of this study included the use of 24 Jersey and Holstein calves who were fed aureomycin plus a basal ration of corn distillers dried solubles (plant protein) or a basal ration containing dried skimmilk. Aureomycin appeared to stimulate growth more with the plant protein than when used with the skimmilk. The difference was not significant, however, The antibiotic seemed to increase efficiency of feed utilization but did not reduce scours. The type of protein in the basal diet tended to influence the effects of the aureomycin on daily gain, skeletal growth, incidence of scours, and consumption of starter and hay. Part B was devoted to 32 Jersey and Holstein calves which were raised to 86 days of age on a limited milk-starter-hay system to evaluate the use of a combination of aureomycin and terramycin, as compared to either antibiotic when used individually. Growth rate was stimulated, but not significantly, by either supplement or in combination. R. W. Hunt

710. Fodder for ruminants. J. KAMLET. U. S. Patent 2,715,067. 3 claims. Aug. 9, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 2: 234. 1955.

A roughage-type feed stuff for cattle, comprising, in part, ground-up waste newspapers and other wastepaper products.

R. Whitaker

711. Silage preservative. E. J. RUSSELL (assignor to Trojan Powder Company). U. S. Patent 2,714,067. 2 claims. July 26, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 4: 522. 1955.

Silage is preserved by a mixture of 50-60 parts by weight of sodium formate and 40-50 parts of calcium chloride. R. Whitaker

HERD MANAGEMENT

712. Figuring milk production costs in northern Illinois. R. H. WILCOX. Ill. Agr. Expt. Sta., Urbana. Bull. 583. 1954.

Milk production per cow averaged 9,366 lb/yr

in northwestern Ill. Feed and labor expenses constituted 91 to 95% of the net production cost; of this, feed accounted for 68 to 70% and labor 23 to 25%. The greatest fluctuation in feed consumed occurred with silage and hay. High producing cows have the lowest unit cost for milk produced. Formulas and graphs are presented for use in computing milk production costs in northern Ill. R. W. Hunt

713. Guide in answering basic questions on farm machinery costs. L. E. CHOATE and S. A. WALKER. Idaho Agr. Expt. Sta., Moscow. Bull. 224. 1954.

The following questions are presented with adequate answers to assist a farmer in purchasing machinery. What machinery do I need? What does farm machinery cost? What are the costs of the machinery on my farm? Do I have a farm size that fully utilizes my machinery? If my machinery is not operating at capacity, how much can I afford to pay for additional land? Should I buy new replacement equipment? What does a second tractor cost me? Other additional machinery—should I buy or hire it? Can I reduce my machinery costs by doing custom work? What will it cost me to add another crop? How will acreage controls under price support programs affect my machinery costs? R. W. Hunt

714. Ringing device for cattle. E. W. ROBERTSON. U. S. Patent 2,716,752. 7 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 10. 1955.

A device for inserting a ring in the nostrils of cattle. R. Whitaker

715. Calf feeder nipple and valve assembly. A. F. WILSON and J. J. CLEMENT (assignors to Lisk-Savory Corp.). U. S. Patent 2,717,000. 2 claims. Sept. 6, 1955. Offic. Gaz. U. S. Pat. Office, **698**, 1: 71. 1955.

A nipple for inserting in a hole in the side of a pail, near the bottom, for feeding calves. A built-in ball type valve controls the flow. R. Whitaker

716. Head gate for cattle chute. L. E. HELDENBRAND. U. S. Patent 2,714,872. 6 claims. Aug. 9, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 2: 184. 1955.

A gate-like device for restraining cattle in a chute by holding the animal's head in a frame made of pipe or tubular material. R. Whitaker

717. Teat cup reversing tool. C. E. NELSON. U. S. Patent 2,714,319. 3 claims. August 2, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 1: 37. 1955.

A device for easily turning rubber teat cups inside out. R. Whitaker

718. Calf feeding devices. H. L. VOIGT and R. N. SELLON, JR. (assignors to Geuder, Paeschke & Frey Co.). U. S. Patent 2,714,368. 1 claim. August 2, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 1: 49. 1955.

A nipple for feeding calves is installed in a hole in the side of the pail near the bottom by means of a ferrule. A ball and socket type valve prevents leakage when feeder is not in use. R. Whitaker

719. Electric dehorner. B. B. SMITHEY. U. S. Patent 2,713,717. 1 claim. July 26, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 4: 433. 1955.

An electric motor driven circular saw for dehorning cattle. R. Whitaker

720. Bulk milk cooler. H. E. CANN, SR. and M. B. CANN (assignors to Esco Cabinet Co.). U. S. Patent 2,713,251. 7 claims. July 19, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 3: 303. 1955.

A vat for cooling milk in bulk, wherein a refrigerant is passed through coils adjacent to the outside walls of the vat. Water is continually circulated from a pump through sprays at the top of the vat wall. The cascading water is cooled by the ice forming on the coils and, in turn, cools the milk. R. Whitaker

721. Refrigeration cabinet. H. E. CANN, SR. and M. B. CANN (assignors to Esco Cabinet Co.). U. S. Patent 2,713,248. 9 claims. July 19, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 3: 302. 1955.

A cabinet for cooling milk in cans, wherein a refrigerant is passed through coils surrounding the space where the cans are supported. Water is circulated from a pump through spray nozzles directed over the cans. The water is cooled as it passes over ice which forms on the coils. R. Whitaker

722. Animal chute. I. L. STEPHENSON; B. DUKE, administrator (assignor to Fannie Stephenson; Basil Duke, guardian). U. S. Patent 2,713,326. 6 claims. July 19, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 3: 322. 1955.

A device for holding calves and other farm animals in any position for medical attention. R. Whitaker

ICE CREAM

723. New patterns in modern merchandising of ice cream. D. J. COOK. S. Dairy Prod. J., **58**, 3: 34. 1955.

Radical changes in the merchandising of ice cream during the last 10 years have resulted from conditions which have caused much modification in some businesses and resulted in failure of others. Some of the new developments have been packages and techniques for self-service shopping, lower price spreads in food store outlets, keener competition between kinds of foods, increasing population, abundance of

products on the market, increasing cost of rent and labor and greater competition for outlets. Merchandising methods must change with the times if the ice cream manufacturer is to survive. A program adapted to local conditions is one of the most important factors. Individual and special attention should be given to merchandising methods in food stores, fountains, and restaurants. Ice cream must be made available wherever people choose to eat. Advertising and merchandising should be well coordinated. Complete cooperation should be given to all promotion events of related industries and of national dairy organizations.

F. W. Bennett

724. Automatic dispenser for ice cream balls or the like. R. G. TARR. U. S. Patent 2,716,385. 7 claims. Aug. 30, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 5: 597. 1955.

An automatic dispenser for producing individual portions of ice cream in the shape of spheres. The device which fits into the sleeve of an ice cream cabinet and uses ice cream in bulk containers, dispenses measured portions through a tube by application of pressure on the ice cream. R. Whitaker

725. Ice cream packaging nozzle. G. G. ALEXANDER. U. S. Patent 2,715,484. 3 claims. Aug. 16, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 3: 351. 1955.

An ice cream package filling head, consisting of four parallel tubes so spaced that the ice cream uniformly fills cartons as it is extruded from the nozzles. The tubes are attached to a manifold which in turn is attached to a continuous freezer. R. Whitaker

726. Ice cream dipper. S. GARGANO. U. S. Patent 2,714,862. 2 claims. Aug. 19, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 2: 181. 1955.

A hand dipper designed to serve spherical-shaped individual portions of ice cream and having pointed teeth protruding from the edge of the bowl which can be oscillated by rotating the handle of the dipper. R. Whitaker

727. Ice cream spoon. J. H. JACOBSON. U. S. Patent 2,715,175. 2 claims. Aug. 9, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 2: 253. 1955.

An ice cream dipper having an adjustable electrical heating unit in the handle.

R. Whitaker

728. Apparatus for making ice cream. E. F. CHANDLER (assignor to Peter Fries, Jr.). U. S. Patent 2,713,253. 1 claim. July 19, 1955. Offic. Gaz. U. S. Pat. Office, **696**, 3: 304. 1955.

A small freezer for making frozen desserts at point of sale, consisting of a hopper which feeds mix into the first of two chambers. Here the mix is whipped, agitated, and pressurized by a stream of CO_2 gas which is boiled off from a coil containing liquid CO_2 immersed in the second chamber where the whipped mix is frozen, and then discharged. R. Whitaker

MILK AND CREAM

729. Farm bulk handling in Ohio. F. KOVAL, Dept. of Dairy Technol., Ohio State Univ., Columbus. Milk Prod. J., **46**, 8: 14. 1955.

A survey on farm bulk milk handling in Ohio was made in the fall of 1954. The survey was directed towards:

(1) Licensed dairy plants to determine the number of plants in farm milk pickup; proportion of milk being collected by this means; number and proportion of shippers who were involved by a plant using this procedure; and to inquire as to the future plans of the plant managers relative to bulk milk pickup.

(2) Plants receiving milk from at least one pickup route. Information solicited was for number of routes, number and size of tanks on farms, daily production figures, difference in quality as compared to can handling, frequency of pickup, and methods of financing.

(3) Major county and city health departments to ascertain the policy and thinking of these groups on the various aspects of farm bulk milk handling such as legality of practice, inspection problems and responsibilities, quality of product, and every-other-day pickup of milk.

The 260 replies to (1) (of a total 675 mailed) indicated that 14 were receiving some milk from farm bulk milk pickup. The length of time these plants were involved in this system ranged from 1 mo. to 3 yr. Seven of these 14 plants had less than 25% of their shippers involved in the pickup; 3 reported about 25% of their shippers were involved; 1 indicated 75% of the producers were on the system; 4 reported that all of their producers had converted to this system.

Questionnaire (2) went to 14 plants who indicated at least one pickup route in operation. The replies indicated: there were 24 farm bulk pickup routes in operation with about 232 farm tanks. Practically all of the milk was on every-other-day pickup. The most prevalent size tank was 300 gal., however, there were a few 100 and 500 gal. tanks. The average daily production on farms having tanks was 870 lb. with a range of 500 to 3,000 lb. All plants reporting indicated improvement in the bacteriological quality of the milk over the previous manner of handling.

Of 90 local health departments contacted concerning (3); 60 answered, revealing the following information: 76% of the sanitarians expressed a favorable attitude toward the system of handling milk, 12% had an unfavorable attitude; the remaining ones did not express an opinion. Eighty-five % mentioned that their codes would permit every-other-day pickup; 11% indicated it was contrary to the code interpretation; the others expressed no opinion. Seventy % felt this new development

would increase inspection responsibilities, 11% said the responsibilities would decrease; the remainder did not express an opinion.

The sanitarians felt that farm methods of cleaning equipment, sanitary procedures in tank truck pickup operation, and methods of milk production; sanitary milking techniques, use of strip cup, etc., would require special attention. J. J. Janzen

730. Importance of original leveling noted in bulk tank survey. ANON. Milk Dealer, **44**, 11: 124. 1955.

A survey made by the Wisconsin Dept. of Agr. has shown that the accuracy of milk measurement is dependent upon leveling of the farm bulk milk tank at the time of installation, maintaining this level at all times, and accurate calibration in the tanks and care in reading the bulk tank measuring stick.

C. J. Babcock

731. Untersuchungen zur H_2O_2 Behandlung der Milch (Experiments on H_2O_2 treated milk). H. LÜCK and F. J. JOUBERT, Dairy Ind. Control Board, Windhoek, Southwest Africa. Milchwissenschaft, **10**, 161. 1955.

The subject of milk preservation with chemicals, particularly H_2O_2, is reviewed. The effect of H_2O_2 treatment of milk on proteins was studied by means of electrophoresis. No changes were observed in the casein components. The electrophoretic mobility of β-lactoglobulin was reduced. The peak was flattened and disappeared completely with higher concentration of H_2O_2. The effect of H_2O_2 treatment on the SH groups of proteins also was investigated.

J. Tobias

732. Observations on temperature changes in pasteurized milk during bottling, storage, and distribution. A. RATZLAFF, Marigold Dairies, Inc., Rochester, Minn. J. Milk Food Technol., **18**, 8: 195. 1955.

These studies indicate that storage room temperatures of packaged milk are not reduced appreciably. The best results are obtained when the milk is cooled after pasteurization and before packaging. Open display cabinets showed marked variation in temperatures, especially when the cabinets were over-loaded. This could alter the keeping quality of milk.

H. H. Weiser

733. Paper milk container with cream separating attachment. C. N. BERGSTROM. U. S. Patent 2,716,518. 4 claims. Aug. 30, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 5: 631. 1955.

A hinged wax coated paper gate is attached to the inside wall of one side of a Pure-pak type milk carton, in such a manner that when a certain spot is pressed on the outside while pouring, the gate allows only gravity raised cream to reach the pouring port. Milk can be dispensed by holding the opposite sides of the carton when pouring, which squeezing action manipulates the gate to mix the cream and skimmilk. R. Whitaker

734. Whipping device. A. SCHWALBE. U. S. Patent 2,715,519. 2 claims. Aug. 16, 1955. Offic. Gaz. U. S. Pat. Office, **697**, 3: 360. 1955.

A hand operated cream whipper of the egg beater type. The diameter of the blades is small enough to permit insertion and whipping of the cream in the original glass bottle.

R. Whitaker

MILK SECRETION

735. The fat content of milk. Why it varies. T. R. FREEMAN. Ky. Agr. Expt. Sta., Lexington. Cir. 530. 1955.

Breed is the most important factor influencing the fat content of milk. However, even within a given breed, there is considerable variation. A cow in poor health may produce milk above or below the expected fat content. As the age of the animal increases the fat content of her milk may drop slightly, usually no more than 0.5%. Fat secretion also is influenced by stage of lactation. For about the first two months fat drops slightly, remains fairly constant for two or three months, and then increases slightly till the end of lactation. Strippings are higher in fat than foremilk. Variations in intervals between milking will result in higher testing milk drawn after the shortest period. Types of feed do not influence fat content of the milk unless the ration is deficient in roughage. R. W. Hunt

NUTRITIVE VALUE OF DAIRY PRODUCTS

736. Relative nutritive values of proteins in foods and supplementary value of amino acids in pearled barley and peanut flour. B. SURE, Dept. of Agr. Chem., Univ. of Ark., Fayetteville. J. Agr. and Food Chem., **3**: 789. 1955.

A study was conducted on the relative nutritive values of the proteins in various foods at different levels of intake and during several periods of experimentation. The results are expressed as gains in body weight of the albino rat per g. of protein intake. Data are included on the influence of heat and duration of heating on the nutritive value of proteins in dried nonfat milk solids. Detrimental effects of high heat treatment in baker's grade non-fat dry milk solids are noted. S. Patton

737. Influence du caillage sur l'utilisation digestive et sur la valeur biologique des protides du lait. Essais de supplémentation des protides du lait, du caillé et du Camembert par la cystine. (Influence of coagulation on digestibility and biological value of proteins of milk. Study of supplementing proteins of milk, of curd, and of Camembert cheese with cystine). L. RANDON, C. JOURDUN, and M.

CAUSERET, Natl. Inst. of Agron. Research. Le Lait, **35**, 272. 1955.

Digestibility of the proteins of curd was found to be slightly superior to those of milk in rat feeding tests. The biological value of the curd protein was markedly inferior to milk protein. It was postulated that part of the reduction of biological value was due to loss of soluble proteins in the whey. Results with Camembert cheese corresponded to those of the curd, indicating that subsequent curing of cheese has only a secondary influence on the biological properties of the proteins. Under conditions of the experiments, the addition of cystine did not improve biological value of the products. A. W. Rudnick, Jr.

PHYSIOLOGY AND ENDOCRINOLOGY

738. Physiological studies of the vagal nerve supply to the bovine stomach. I. Comparison of responses in milk-fed and roughage-fed calves, using chronic intrathoracic vagal electrode technique. H. E. DZIUK and A. F. SELLERS, Univ. of Minn., St. Paul. Am. J. Vet. Research, **16**, 60: 411. 1955.

Silver electrodes embedded in a polyethylene shield were attached to the vagal nerve trunks of 17 calves, 11 of which recovered satisfactorily from the operation. These were used to record rumen motility patterns, both normal and when stimulated by electricity applied to the vagal electrodes. Stimulated contractions in all calves were similar to the spontaneous contractions. Rumen contractions in five milk-fed calves were of low amplitude, but those of roughage-fed calves were quite marked, more regular and stronger. Data relative to two calves changed from milk to roughage indicated that it took about 60 days to attain full characteristic roughage type motility.

E. W. Swanson

739. Utilization physiologique comparée du phosphore et du calcium du lait et du yoghourt. (Comparison of physiological utilization of phosphorus and calcium from milk and yoghourt). J. CAUSERET and D. HUGOT. Le Lait, **35**, 129. 1955.

In three rat feeding experiments, it was found that the animals utilized 35%, 9%, and 14% less calcium and 21%, 6%, and 10% less phosphorus when yoghourt was fed than when fresh milk was fed. The wide variations in results were attributed to either the milk used or the conditions under which the yoghourt was prepared. The authors conclude that despite the decrease in physiological utilization of calcium and phosphorus in yoghourt, it is still an equal or better qualitatively and quantitatively source of these elements than are some cheese. A. W. Rudnick, Jr.

SANITATION AND CLEANSING

740. Plant equipment cleaned-in-place. G. A. SMITH and A. A. ROTH, Wyandotte Chemicals Corp., Wyandotte, Mich. Milk Plant Monthly, **44**, 8: 27. 1955.

The methods of cleaning lines in place apply to other pieces of equipment through which solutions may be pumped. The following points, however, must be considered: (1) Cold milk lines may be effectively cleaned by a single circulation of a chlorinated cleaner. An acid cleaner at times may be necessary, (2) Double circulation is required with hot milk lines. Sometimes it is possible to save time and money by circulating hot and cold lines separately, (3) Separators and clarifiers cannot be cleaned by in-place methods. They can, however, be thoroughly rinsed while the machine is in operation. Power brushes are most commonly used, (4) Holding tanks, standardizing and processing vats, and other large pieces of equipment are cleaned by spray jets, and (5) Spray jets, and more recently, steam jets are successfully used in cleaning vacuum pans and evaporators.

Cleaning equipment in-place by mechanical means creates a problem of corrosion. It is necessary to prevent the flow of electric current from one piece of metal to another or from one spot to another spot on the same piece of equipment. It is important that all metal in any C-I-P circuit be of the same type and all motors, liquid level controls, and other electrical equipment be grounded away from stainless steel. C. J. Babcock

741. Engineering for C-I-P. D. A. SEIBERLING, Dept. of Dairy Technol., Ohio State Univ., Columbus. Milk Plant Monthly, **44**, 8: 13. 1955.

Cleaning-in-place refers specifically to the cleaning and sanitizing of dairy processing equipment and piping in its assembled condition. The following steps necessary in developing a C-I-P program are discussed: (1) Developing the circuit, (2) Selecting the materials, (3) Completing the installation, and (4) Planning a cleaning program capable of producing the desired results.

It is pointed out that the general operation of a C-I-P system involves the following procedure: (1) Flush system free from product residues, (2) Clean valves, caps, and other manually cleaned items, install by-passes, and connect return lines to complete the circuit, (3) Wash by recirculating acid or alkaline detergents under conditions of time and temperature which will produce the desired results, (4) Rinse and cool the system; then drain completely, and (5) Reassemble for processing and sanitize with hot water or chemical sanitizers. In addition to economical advantages, C-I-P can assure improved sanitations, more attractive piping layouts, and better employee morale. C. J. Babcock

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