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Contents

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กระทรวงอุตสาหกรรม

ADMINISTRATIVE INFORMATION

TIPS TO SANITARIANS

A BACTERIAL ENZYMATIC METHOD FOR DETERMINING
TYROSINE IN CHEESE

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Free tyrosine determinations of biological materials on a quantitative scale present a complex problem. Important basic difficulties were early recognized by Folin and Ciocalteu *(3)* in their classical studies on protein hydrolyzates and, later, were more extensively discussed by Mogensen *(10)*.

The accurate determination of free tyrosine in cheese requires either prior separation of interfering compounds, such as tryptophan; lower peptides and tyramine, or the introduction of a procedure with specificity for tyrosine. Neither paper chromatography nor microbiological assay has proved entirely satisfactory, as the former method lacks sensitivity for tyrosine and the latter is subject to serious errors *(6)* from peptide interference.

Gale *(4)* has proposed a method for L-tyrosine with tyrosine decarboxylase at pH 5.5 to catalyze the following reaction:

$$\text{L-tyrosine} \rightarrow \text{tyramine} + CO_2$$
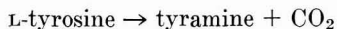
The CO_2 evolved is measured in a Warburg apparatus and equated to tyrosine. Optimal concentrations of tyrosine for this assay were between 0.5 and 2.0 mg. This method could not be applied to dairy products by the present authors because the size of sample possessing the optimum tyrosine concentration was too large for the Warburg apparatus. In addition, CO_2 evolution in recovery experiments bore little relationship to calculated recoveries, evidently because of the complex gas absorptive nature of cheese components.

From the above enzyme reaction a measurement of tyramine instead of CO_2 might serve equally well to determine tyrosine in dairy products. Kosikowski and Dahlberg *(8)* have successfully applied a sensitive analytical procedure for extracting tyramine out of cheese, but a time element of 24-48 hours was required. In the present study, an adaptation of this method, involving multiple ether extractions from a stronger alkaline buffer, gave excellent recoveries of tyramine in a much shorter time and was used to advantage in subsequent determinations.

The proposed method for tyrosine, described in detail here, is based on the following considerations. Free tyrosine present naturally in a cheese suspension serves as substrate for a bacterial decarboxylase added to the suspension in
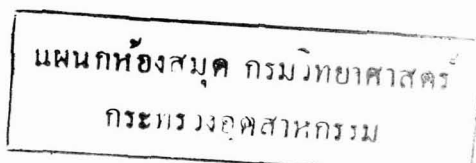
excess. The reaction is brought to completion when all available free tyrosine has been converted to tyramine. In practice two identical cheese suspensions are made from the same cheese; one is subjected to bacterial decarboxylase activity while the other is not so subjected and then each is analyzed for tyramine. From the respective amine values two constants of cheese can be calculated, free tyrosine at time of analysis and total tyrosine liberated from cheese protein during ripening.

REAGENTS AND EQUIPMENT

Reagents:

   a) Acetate buffer pH 5.5 : 8.2 g. anhydrous sodium acetate plus 0.75 ml. glacial acetic acid made up to 100 ml.

   b) Trichloroacetic acid—50% : 50 g. trichloroacetic acid, crystals, made up to 100 ml. in a glass-stoppered volumetric flask.

   c) Phosphate–sulfate buffer *(2)* : 11.4 g. K_3PO_4 anhydrous, 68.4 g. Na_2SO_4 anhydrous dissolved in warm water made up to 250 ml., pH 12.

   d) $\dfrac{M}{50}$ sulfuric acid solution : 1.1 ml. reagent grade concentrated sulfuric acid made up to 1 liter.

   e) Ethyl ether : peroxide-free, reagent grade, anhydrous.

   f) Acetic acid 95%.

   g) Mercuric sulfate–sulfuric acid : 20 g. of $HgSO_4$ dissolved in 190 ml. of water to which 10 ml. of concentrated H_2SO_4 has been added.

   h) Sodium nitrite 1.5%. 750 mg. of $NaNO_2$ dissolved in 50 ml. of water. Use fresh daily.
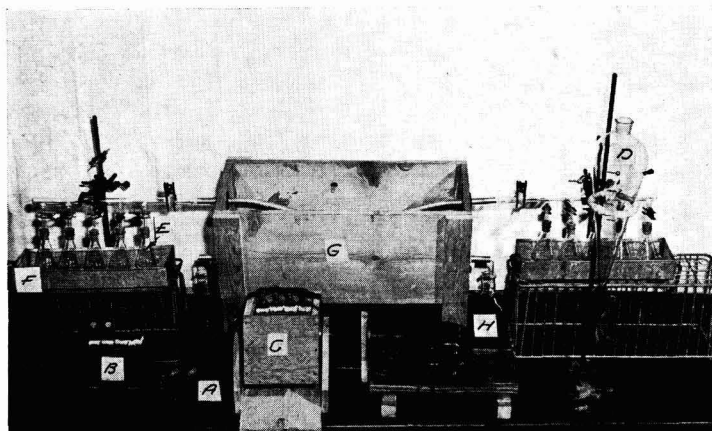
FIG. 1. Ether extraction and evaporation apparatus for tyramine. A = tyramine extraction flask, No. G-3 Mojonnier; B = centrifuge baskets, No. T-66 Mojonnier; C = extraction unit, 10-12 r.p.m.; D = ether dispenser, 2-l. capacity; E = tyramine receiving flask; F = hot water container; G = condenser; H = ether collection flask.

Equipment—See Figure 1.

a) Extraction—A revolving type, 12 r.p.m. geared drive, agitation box. Tyramine extraction flask seated in metal holders and held in place by rubber straps.

b) Evaporation—Tyramine receiving 125 ml. Erlenmeyer flasks in circulating hot water (65°-70° C.) secured to glass manifold. Latter connected to aluminum condensing tube and cooled by cold tap or ice water.

c) Colorimeter—Bausch and Lomb "Monochromatic," selected ½ "dia. test tubes or Coleman model"—13 mm. square cuvettes.

Tyrosine decarboxylase preparation (1, 5). Streptococcus faecalis, strain R, is inoculated daily into broth A and incubated at 30°-33° C. for 24 hours four times prior to use. Ten ml. of this actively growing culture is introduced into 2 l. of broth B and incubated at 30°-33° C. for another 18 hours. The cells are harvested by centrifugation and washed twice in distilled water. A thick slurry of the cells is dispersed into five volumes of cold (0°-5° C.) acetone and refrigerated at 5° C. for 30 minutes. The resultant cell suspension is filtered onto a small Buchner funnel and successively washed with acetone, a mixture containing equal parts of acetone and anhydrous ethyl ether, and finally with only anhydrous ethyl ether. The greyish-white powder is spread on a watch glass and allowed to dry until no ether fumes are detectable. This product, if stored in a desiccator over $CaCl_2$ at 0°-5° C., is usually stable for at least 1 month.[2]

Broth A	*Broth B*
1% yeast extract	1% yeast extract
1% tryptone	1% tryptone
0.1% glucose	1% glucose
0.5% K_2HPO_4	0.5% K_2HPO_4
pH 7.0	pH 7.0

THE METHOD

Preparation of samples and decarboxylation of tyrosine. Two g. of cheese (±0.01 g.) are ground to a paste in a mortar with a small portion of 50° C. distilled water. With small volumes of warm water this cheese suspension is quantitatively transferred to a 100-ml. volumetric flask until the flask is two-thirds full.

The flask is placed in a bath of boiling water for 10 minutes, then cooled to 33° C. Five ml. of acetate buffer pH 5.5 is added, followed by 1 ml. of a suspension containing 25 mg. of active tyrosine decarboxylase powder per millimeter, and the whole is thoroughly mixed. The suspension at pH 5.5 is incubated in the water bath at 32°-34° C. for 30 minutes with periodic agitation. Then 10 ml. of 50% trichloroacetic acid is pipetted into the flask, and distilled water is added to

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[2] Prior to use, test for activity. Pipette 1 ml. of standard tyrosine solution, containing 200 γ L-tyrosine per ml., into a tyramine extraction flask. Adjust to pH 5.5 by adding 4 ml. of acetate buffer. Add approximately 2 mg. of decarboxylase preparation and incubate for 30 minutes at 32°-34° C. Then add 16 ml. phosphate–sulfate buffer and extract and develop color as for regular tyrosine analysis. Recovery of at least 45 γ of tyramine indicates an active preparation.

the 100-ml. mark. The contents are mixed and after standing for 5-10 minutes are filtered through Whatman No. 42 filter paper.

Tyrosine analysis–extraction of tyramine. Five ml. of the filtrate is pipetted into a tyramine extraction flask. Then 16 ml. of the tripotassium phosphate–sodium sulfate buffer (pH 12) is added, followed by 40 ml. of peroxide-free ether.

The extraction flask is stoppered by a cork, preferably collodion covered, and the contents are agitated for 3 minutes in the extraction unit (Figure 1-C). Upon standing the two-phase system quickly separates and the ether layer is decanted into a 125-ml. Erlenmeyer flask containing 2 ml. of $\frac{M}{50}$ sulfuric acid. The ether is evaporated from this receiving flask in the evaporating apparatus described in Figure 1, the tyramine being trapped by the dilute sulfuric acid. This extraction and evaporation is repeated on the same 5-ml. filtrate an additional four times with 40-ml. quantities of fresh ether.

Color development—with Millon reagent.[3] When all the ether has been boiled off after the fifth and final extraction, the tyramine receiving flasks are removed and placed for 10 minutes in the air current from an electric fan. Following this, 3 ml. of 95% acetic acid and 2 ml. of mercuric sulfate–sulfuric acid solution are added. After mixing, the cooled ether-free solution is poured from the receiving flask directly into a Bausch and Lomb selected ½-in. diameter test tube with a 5-second pause after the last drop.

The tube is placed in boiling water for 3 minutes, after which the mixture is cooled to about 25° C. An initial transmission reading (G) for turbidity, prior to color development, is made at 505 mμ against a reagent blank set at G = 100. Then 0.2 ml. of fresh sodium nitrite solution is added, and after 12 minutes of color development at room temperature per cent transmission (G) of the red color complex is made at 505 mμ against a reagent blank set at G = 100. After turbidity value, if any, has been taken into account, L = 2 – log. G.

Initial tyramine analysis. A 2-g. sample of the same cheese is ground and suspended in approximately 60 ml. of 50° C. distilled water in a 100-ml. volumetric flask. After placing this suspension in boiling water for 10 minutes, 10 ml. of 50% trichloroacetic acid is added. The contents are mixed and brought to 100 ml. volume with distilled water and allowed to stand for an additional 10 minutes at room temperature before being filtered (Whatman No. 42). No acetate or decarboxylase preparation is added. Multiple extraction of 5 ml. of this filtrate and color development of the free tyramine proceeds as described above.

Preparation of standard curve and calculations for tyrosine. Solutions con-

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[3] Alternate use of Folin-Ciocalteu reagent for color development. After removal of the final ether and 10-minute air blowing, 10 ml. of a 15% sodium carbonate–2% Quadrafos solution *(7)* are pipetted into the tyramine receiving flask followed by 3 ml. of diluted Folin–Ciocalteu phenol reagent (1 part reagent + 2 parts water). In 10 minutes 0.1 g. Celite 535 filter aid are added and the solution is filtered through Whatman No. 12 fluted filter paper. Per cent transmission, G, of the blue color complex is read at 650 mμ against a reagent blank set at G = 100. No reading for turbidity is required with this reagent. A specific standard curve must be prepared for this alternate step.
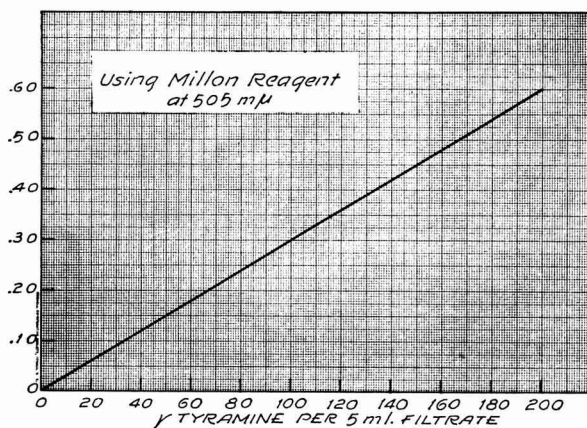
FIG. 2. Standard curve for tyramine in bacterial enzymatic method for tyrosine using either Bausch and Lomb ''Monochromatic'' colorimeter ½-in. selected tubes, or Coleman Model 11 spectrophotometer, 13 mm. square cuvettes against a reagent blank.

taining 10, 20, 30, 50, 100, 150, and 200 γ tyramine in 5 ml. of 5% trichloroacetic acid are extracted and subjected to colorimetry with Millon's or Folin-Ciocalteu's reagent at 505 and 650 mμ, respectively, with identical pH and similar size of tubes and equipment as in actual testing.

A resulting standard curve, based on Millon's reagent (Figure 2), makes it possible to obtain free tyramine values of cheese before and after the addition of the decarboxylase preparation. Values from this standard curve are expressed as gamma tyramine per 5 ml. of filtrate. Insertion of these values in formulas listed below gives two tyrosine constants of cheese.

(1)   Free tyrosine content of cheese, γ/g cheese $= \dfrac{T_{TA} - T_A}{0.076}$

(2)   Total tyrosine liberated from cheese protein, γ/g cheese $= \dfrac{T_{TA}}{0.076}$

T_{TA} = total free tyramine in cheese after decarboxylation, $\gamma/5$ ml. filtrate.

T_A = initial free tyramine in cheese, $\gamma/5$ ml. filtrate.

0.076 = conversion factor tyramine to tyrosine.[4]

[4] The dilution factor for converting values of tyramine in $\gamma/5$ ml. to γ/g of cheese is 10. The equivalent weight necessary for converting tyramine values to their corresponding tyrosine values is 0.76. Therefore, a divisor of 0.076 is employed for expressing results as γ/g of tyrosine.

RECOVERY EXPERIMENTS AND DISCUSSION

To test the accuracy of the method, known amounts of pure tyramine and tyrosine were added to ripened Cheddar cheese, and recovery experiments were conducted. The procedure proposed here for measuring the tyrosine and tyramine concentration in cheese is shown to be quantitative (Tables 1 and 2) with an experimental error of 5% or less. Analysis of a large number of cheese samples indicates that at very low concentrations slightly less sensitivity may be

TABLE 1

Recovery experiments on 2% solutions of Cheddar cheese that had been ripened for 3 months at 60° F. and contained added amounts of tyramine

Tyramine added	Tyramine recovered	Recovery
(γ/g)	(γ/g)	$(\%)$
0	200	—
125	325	100
250	455	101
500	737	105

TABLE 2

The recovery of added L-tyrosine from 2% solutions of Cheddar cheese that had been ripened at 60° F. for the period indicated

Tyrosine added	Tyrosine recovered	Recovery
(γ/g)	(γ/g)	$(\%)$
	2 months	
0	214	—
500	714	100
625	839	100
100	312	99
200	421	102
500	734	103
	6 months	
0	376	—
250	612	98
500	854	97

expected, which can be increased either by the use of a more concentrated cheese solution or by adjustment of the extraction pH to 10. The simultaneous presence of comparatively large concentrations of tyrosine and tyramine in cheese will not interfere with the determination of either.

Values which determined the standard curve (Figure 2) cannot be obtained directly from standard solutions but only by subjecting standard solutions to

TABLE 3

The partition coefficients of tyramine between ether and an aqueous phosphate–sulfate buffer

Apparent pH	K
8.5	0.04
9.0	0.14
9.5	0.23
10.0	0.29
10.5	0.23
11.0	0.14
11.5	0.06

$$\text{K (partition coefficient)} = \frac{\dfrac{\text{conc. tyramine in ether}}{\text{volume of ether}}}{\dfrac{\text{conc. tyramine in aqueous buffer sol.}}{\text{volume of aqueous buffer sol.}}}$$

the extraction step. The partition coefficient of tyramine between ether and the aqueous phase is low, in the order of 0.2 (Table 3); consequently, after five extractions approximately 80% of the original concentration of tyramine is obtained. This figure is sufficiently constant for quantitative purposes, less than 5% variation, so that in actual practice it is possible to show values approaching

100% recovery. After five extractions approximately 1.7 ml. of $\dfrac{M}{50}$ H_2SO_4

is retained in the tyramine flask from the original 2 ml., but this loss, being constant, does not significantly affect the results if standard curves are prepared properly.

The present method possesses the advantage of a very high degree of specificity for free tyrosine and total tyrosine liberated. Tyrosine decarboxylase is capable only of decarboxylating the biologically active levo form of tyrosine at significant rates (4, 9, 11). Such specificity is of significance, as the presence of interfering compounds in the past has invariably led to higher results than true values. Recovery experiments of tyrosine from cheese suspensions gave no evidence of the presence of possible active peptidase systems in the acetone enzyme preparation which might otherwise have distorted tyrosine values. There is every reason to believe that this bacterial enzymatic method can also be applied to milk and other dairy products.

With the Folin-Ciocalteu reagent specificity will be somewhat lessened if tryptamine, indole, or skatole is present in the cheese, as such compounds are ether-extractable and form blue complexes with this reagent and would be recorded with the initial tyramine values. The danger of this interference is potentially not very great, as indole and skatole do not appear to be present in significant concentrations in ripened Cheddar cheese, and tryptophan is not normally considered as one of the six amino acids capable of being decarboxylated by bacteria (4, 5) to tryptamine. Surface mold and bacterial ripened cheese, possessing a high pH, however, might have significant quantities of indole or skatole. The highest degree of specificity is obtained with the Millon reagent, and its use is preferred.

The pH of the extraction solution is important. Table 3 shows the variation in partition coefficients obtained by extraction at apparently different pH levels. The final pH of the aqueous phase, because of the mixture of 5% trichloroacetic acid and the buffer of pH 12, is slightly less than 10.5. This pH was chosen because it appears to be a range of maximum buffer action for the system and is applicable to the analysis of many other dairy products. Kosikowski and Dahlberg (8) employed a dilute carbonate buffer at about pH 9, and for this reason continuous extraction over an extended length of time was necessary. At a pH of 10.5 with an almost saturated buffer solution, which contributes to the "salting out" effect, it is possible to obtain more efficient extraction (most efficient is at pH 10) and thus curtail drastically the time required. Satisfactory results can be obtained between the pH range of 9.5 to 10.5, but the standard curve must always be obtained at the pH selected for actual testing.

PRECAUTIONS

The Folin-Ciocalteu reagent will react with a variety of agents. Peroxide formation in the ethyl ether was found to be most responsible for interference, resulting in high blank values for color. Normally a reagent blank substituted for a sample in the extraction procedure should have a transmission value of 96-98%. If lower values are obtained, repeated washings of the ether with ferrous sulfate and redistillation are recommended. Initially a great deal of trouble was encountered from substances that were leached from corks used in this test. This difficulty was eliminated by boiling the corks three times until most of the brown pigmentation was removed, drying, and then coating twice with flexible collodion. Some of the collodion at first will be removed during analysis without influencing results, but after one or two runs it will remain fixed. The coated corks used for sealing the extraction flasks may be used repeatedly, if, between analyses, they are rinsed and dried.

None of the trisodium phosphate–sodium sulfate buffer in the extraction flask should be allowed to enter the tyramine receiving flask, since these salts will result in turbidity as a result of the interaction with the Folin-Ciocalteu reagent.

No trouble was experienced with uncoated, clean corks when Millon's reagent was used, and blank values for color normally were negative. Excess turbidity may result, however, when the ether is not fully evaporated at the end and when final solutions are not cooled to room temperature prior to addition of Millon's reagent. For this reason it was found necessary to use the air current from an electric fan to fully remove final traces of ether and to effect cooling.

Temporary emulsions, resulting in a few instances, during extraction may be removed by a gentle shaking of the flask from side to side, by placing the bottom of the flask in hot water for a few minutes, or by centrifuging.

SUMMARY

A rapid quantitative method, specific for free tyrosine in ripened Cheddar cheese, is presented. In this procedure an active bacterial decarboxylase preparation converts free tyrosine to its amine, tyramine, under suitable conditions. A multiple ether extraction at pH 10.5 quantitatively recovers both the initial free tyramine and any resulting from decarboxylation. Then from a second sample of the same cheese not subject to decarboxylation only the initial free tyramine is recovered. Colorimetric analysis of the amine with Millon's reagent, preferably, or Folin-Ciocalteu's reagent follows, and through the use of suitable formulas the free tyrosine of cheese, total tyrosine liberated from protein during ripening, and free tyramine can be calculated.

Recovery experiments, precautions, and an apparatus designed for the rapid and simultaneous extraction of the tyrosine contents of ten samples of cheese as tyramine are described and discussed.

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TYROSINE IN CHEDDAR CHEESE

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In the previous paper, *(11)*, a sensitive method for the quantitative determination of tyrosine in cheese was described. The method is specific for free L-tyrosine, and by the use of appropriate formulas another constant value, total tyrosine liberated from cheese protein, can be obtained.

Dahlberg and Kosikowski *(3)* earlier had reported on the concentration of tyramine found in cheese, and Kosikowski *(4)* had indicated in a general way the relationship between tyrosine and tyramine. Information concerning the amounts of free tyrosine in cheese, its rate of production during ripening, and its relationship to soluble protein is meager.

This study is concerned with the quantitative aspects of free tyrosine in Cheddar cheese and its relationship to some of the other constituents present, as shown by application of the new bacterial enzymatic procedure.

METHODS

Commercial Cheddar cheese. Ten Cheddar cheeses, of the 35-lb. twin type of varying age and history, were obtained from a number of New York state plants. Free tyrosine, total tyrosine liberated from the cheese protein, and free tyramine were determined by the method of Silverman and Kosikowski *(11)*, using the Folin-Ciocalteu reagent. In addition, the percentage of soluble protein was obtained by the procedure used by Dahlberg and Kosikowski *(1)* and phosphatase activity by the Cornell method *(5)*. Quality evaluation of the cheese was conducted by two experienced judges.

Cheese manufactured in the laboratory. Three series of Cheddar cheese totaling ten cheeses in number were made by the authors from raw milk and milk pasteurized at 161° F. for 15.5 seconds. Milk for Series I was obtained from a commercial source in Ithaca, N. Y., and milk for Series II and III was obtained from the Cornell University dairy herd.

The cheeses were manufactured from 300-lb. lots of milk by the procedure of Lochry *et al.* *(7)*. To observe the influence of lactic acid bacteria on the rate of free tyrosine formation, cheeses were made with a 1% inoculum of the conventional lactic acid starter (FL) and also with equal (0.75%) concentrations of this lactic starter and a heat- and salt-tolerant *Streptococcus faecalis* (DK) starter. The enterococci have been recommended previously for Cheddar cheese *(2)*.

After an overnight press in stainless steel square hoops at room temperature the cheeses were cut into 12-oz. blocks, vacuum packed in tin cans, and ripened at 60° F. Within 1 week after manufacture, the cheeses were analyzed by stand-

TABLE 1

An analysis of Cheddar cheese made from raw and pasteurized milk with conventional lactic acid starter (FL) or a combination of FL and DK (S. faecalis) starters and ripened at 60° F.

			Fat	Moisture	Salt	Total protein
Series I			*(%)*	*(%)*	*(%)*	*(%)*
A	Raw	FL	33.25	36.27	1.76	24.26
B	Raw	FL + DK	33.50	36.45	1.72	24.36
C	Past.	FL	33.25	36.70	1.62	23.96
D	Past.	FL + DK	32.75	37.90	1.48	23.76
Series II						
E	Raw	FL	37.25	33.71	1.62	22.92
F	Past.	FL	38.00	33.44	1.66	23.31
G	Past.	FL + DK	36.75	34.62	1.54	23.46
Series III						
H	Raw	FL	34.25	35.88	1.65	24.52
I	Past.	FL	35.00	35.60	1.72	23.51
J	Past.	FL + DK	34.50	36.76	1.51	23.90

ard methods *(12)* for total protein, salt, fat, and moisture. At intervals indicated in Table 1 soluble protein, free tyramine, free tyrosine, and total tyrosine liberated from protein sources were determined on a dry weight basis. The pH of the cheese was measured with a Beckman glass electrode at room temperature.

RESULTS

Observations on the experimental cheese made in this laboratory. Fat, total protein, salt, and moisture (Table 1) were fairly uniform within the same series of cheese but differed between each major series. The pH of the young cheese was generally 5.0 to 5.2.

Rate of free tyrosine production. Free tyrosine in cheese at any given time ranged in concentration during a 176-day ripening period at 60° F. from a low of 52 γ to a high of 2,536 γ per gram of cheese, dry weight (Table 2).

As Cheddar cheese increased in age (Table 2), a steady increase in free tyrosine resulted. The rate of production, however, was not necessarily uniform and there were notable exceptions to this trend with certain cheeses. Such an exception is cheese D, Series I, Table 2, made with a starter consisting of *S. lactis* and *S. faecalis* organisms. Here, within the first 86 days, free tyrosine accumulated at a much slower rate than in the other cheese in the same series but nevertheless had attained a level of 464 γ per gram at the end of this period. At 176 days free tyrosine was no longer present, but the free tyramine had increased substantially. It is evident that the conversion rate of tyrosine to tyramine in this cheese had become so rapid as to completely deplete the supply of free tyrosine accumulated earlier and that tyrosine was apparently being decarboxylated to tyramine immediately upon liberation from peptide sources. Raw or pasteurized milk cheese in these three series containing added *S. faecalis* organisms showed significantly lower concentrations of free tyrosine at comparable ripening intervals than did cheese made with FL starter.

TABLE 2

Free tyrosine, free tyramine,[a] and total tyrosine liberated in Cheddar cheese ripened at 60° F. and calculated on a dry weight basis

	Days of ripening														
	4			33			58			86			176		
	T_A	T_O	T_{TO}	T_A	T_O	T_{TO}	T_A	T_O	T_{TO}	T_A	T_O	T_{TO}	T_A	T_O	T_{TO}
	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)	(γ/g)
Series I															
A	0	165	165	270	892	1162	616	915	1531	1001	1215	2216	2680	903	3583
B	83	52	135	526	210	736	813	280	1093	1380	422	1802	3050	150	3200
C	0	146	146	0	823	823	0	1509	1509	0	2062	2062	341	2536	2877
D	32	81	113	320	264	584	460	277	737	974	464	1438	3740	0	3740
Series II	2			31			59			101			162		
E	0	201	201	11	700	711	48	1128	1176	44	1795	1839	163	2224	2387
F	0	185	185	12	523	535	20	852	872	60	1397	1457	0	1908	1908
G	80	61	141	260	295	555	606	373	979	768	627	1395	1328	829	2157
Series III	2			24			56			95			155		
H	0	69	69	0	621	621	33	1326	1359	268	2308	2576	413	1786	2199
I	0	98	98	5	568	573	0	879	879	20	1413	1433	0	1894	1894
J	93	27	120	190	157	347	446	275	721	806	250	1056	1392	427	1819

[a] All tyramine concentrations have been converted to their equivalent weight of tyrosine for comparative purposes. To find actual free tyramine per gram of cheese, dry weight, multiply by 0.76.

T_A—free tyramine content of cheese.
T_O—free tyrosine content of cheese.
T_{TO}—total tyrosine liberated from cheese protein.
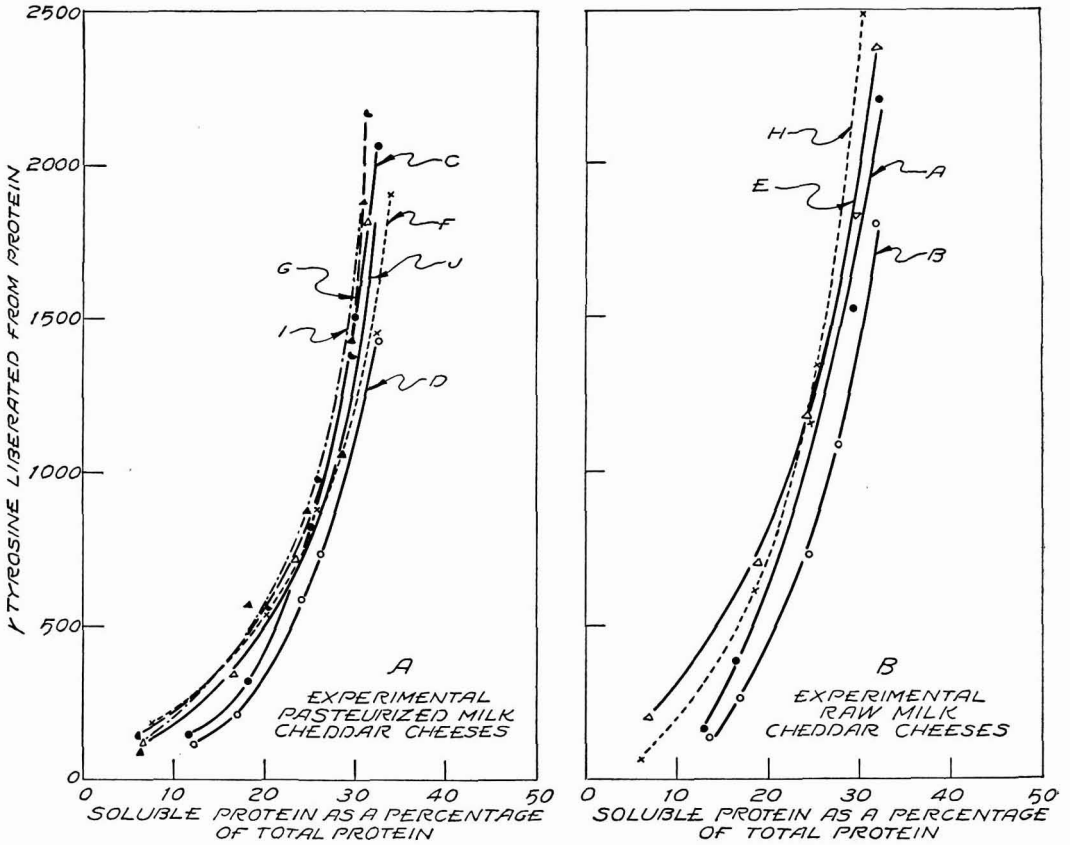
FIG. 1. The relationship between total tyrosine liberated and soluble protein as a percentage of total protein of experimental raw and pasteurized milk Cheddar cheese. Each letter refers to separate cheese listed in Table 1.

Total tyrosine liberated from cheese protein. Compared to free tyrosine values on the same cheese (Table 2), the total tyrosine liberated from cheese protein ranged from a low of 69 γ to a high of 3,583 γ per gram of cheese, dry weight. A steady increase in concentration with ripening was noted for all of these cheeses, and significant differences in the ratios of free tyrosine to free tyramine were evident between certain cheeses. Generally, a greater rate of total tyrosine liberation was observed in raw milk cheese made with regular FL lactic acid starter. After 176 days of ripening at 60° F., as much as 6.0% of the tyrosine of cheese protein was hydrolyzed to its free state, existing either as free tyrosine or, if acted upon by decarboxylase, as free tyramine.

Free tyramine followed the general pattern earlier reported by Dahlberg and Kosikowski *(2, 3)*, increasing rapidly in the cheese containing active decarboxylating enzyme systems and relatively slowly in the cheese in which these systems were suspected as being weak.

Tyrosine versus soluble protein production. Except for the initial state of ripening when most of the action on cheese protein apparently resulted in a predominance of higher peptides, the rate of tyrosine hydrolyzed from peptide bondings exceeded that of the protein solubilized, as shown in Figure 1. A surprisingly close agreement exists among cheeses made from different milk and starters in the nature of the slope presented for each cheese. When the cheese is about 3 months of age the curves incline sharply to the vertical. The slope of the curves for raw milk cheese in the initial stages of ripening was slightly less steep than those for pasteurized milk cheese, but after 3 months at 60° F., near the peak of ripening, differences in the characteristics of the curves were not appreciable. These data suggest that tyrosine is liberated during the latter stages of ripening at the expense of the various peptide sources.

Tyrosine in commercial cheese. In Table 3 it may be observed that the free tyrosine concentration of ten commercial cheeses was of the same order as that found in the experimental cheese, from 348 to 2,630 γ per gram of cheese, dry weight. Cheeses with similar soluble protein concentrations differed in their ratios of tyramine to tyrosine; those having a higher free tyrosine content generally had a lower concentration of free tyramine. The value for total tyrosine liberated from cheese protein ranged from 485 to 2,921 γ per gram of cheese, dry weight. It is interesting to note (Figure 2) that the relationship between total tyrosine liberated and soluble protein among these ten commercial cheeses of widely different histories closely resembled that found with the experimental cheese (Figure 1, A and B).

DISCUSSION

In the early stages of cheese ripening both soluble protein and total tyrosine liberated increased significantly as the experimental cheese aged. However, in a relatively short time the rate of production of soluble protein slowed down considerably, whereas the liberation of total tyrosine maintained its rapid rate, undoubtedly at the expense of the lower peptides. Total tyrosine liberated is apparently a much more sensitive index of protein breakdown of cheese ripening than is soluble protein. Such a sensitive index of cheese ripening should prove to be of value for application to future cheese work.

Some investigators *(8, 9)* have indicated that the free amino acids found in cheese and starter filtrates are present in direct proportion to their concentration in casein. Though conceding that this condition undoubtedly exists for certain of the free amino acids, Kosikowski and Dahlberg *(6)* point out that this does not apply for at least two amino acids, proline and tyrosine. Unless the tyramine content of cheese is first considered, any interpretation of data concerning the liberation of tyrosine from cheese during ripening may be invalid. Essentially, two independent reactions may be occurring simultaneously, proteolysis resulting in the appearance of tyrosine and decarboxylation of this compound to tyramine. The presence and rate of activity, therefore, of decarboxylating enzymes present naturally in the flora of raw milk cheese or added as *S. faecalis* starter influence greatly the free tyrosine content of cheese.

TABLE 3

Analysis of commercial Cheddar cheese made from raw and pasteurized milk

No.	Age	Phosphatase test	Moisture	Soluble protein	Total protein	Tyramine[a]	Free tyrosine	Total tyrosine	Flavor and body Score[c]	Flavor intensity
	(mo.)		(%)	(%)	(%)	(γ/g)	(γ/g)	(γ/g)		
1	15	Past.	35.63	10.54	25.61	291[b]	2630[b]	2921[b]	38.5 sl A	+|-
2	15	Past.	36.28	9.67	24.89	206	2321	2527	38.5 H	+|-
3	13	Raw	37.01	8.31	24.29	1286	476	1762	36.5 UC	++++
4	15	Past.	36.34	9.49	25.39	892	1386	2278	40	++|-
5	16	Past.	33.07	8.02	27.05	0	1912	1912	38.5 Fl	−
6	16	Past.	34.38	8.64	26.29	672	669	1341	37.5 Fr, UC	+++
7	15	Past.	35.10	9.65	25.33	182	1914	2096	39 A	+|-
8	12	Raw	34.84	8.99	25.07	500	1848	2348	37.5 A, B, sl UC	++|-
9	5	Past.	36.25	5.50	25.01	218	428	646	40	++
10	5	Past.	34.87	5.07	24.79	137	348	485	40	+

[a] Expressed as the weight of tyrosine before decarboxylation for comparative purposes.
[b] All values are on a dry weight basis.
[c] A = acid, H = heated, UC = unclean, Fl = flat, Fr = fruity, B = bitter, sl = slightly.
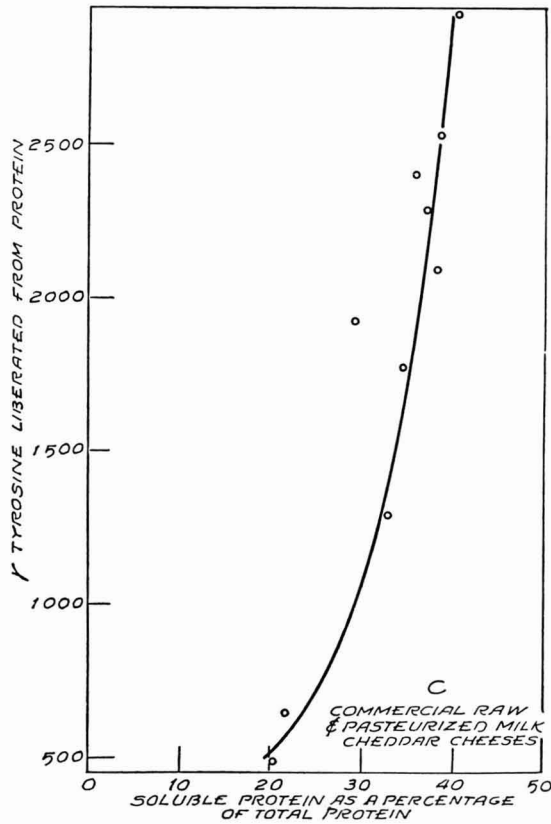
FIG. 2. The relationship between total tyrosine liberated and soluble protein as a percentage of total protein of commercial raw and pasteurized milk Cheddar cheese.

The Folin-Ciocalteu reagent was employed in the present study, as the more specific qualities of the Millon reagent were not fully appreciated until after the experiment was well under way. Actually, for ripened cheese the Millon reagent is preferable, as possible interference with tryptamine or indole and skatole is then ruled out. To evaluate any interference by the last two compounds on the present results, all cheeses were ether-extracted under acid conditions, pH 5.0-5.5, and the resultant color-complex was developed with the Folin-Ciocalteu reagent. No significant color was found in any of these cheese, indicating that indole and skatole were not present. Aromatic amine determination by paper chromatography (10) indicated that tryptamine was not detectable in any of the cheeses with the possible exception of two cheeses showing traces of a compound falling on the tryptamine area on the chromatogram.

Present studies pointing out the intimate relationships existing between free tyrosine, total tyrosine liberated, and free tyramine in ripened cheese perhaps indicate a need for a reevaluation of possible changes which might be occurring among other compounds during all stages of ripening. Critical examination of products of ripening on cheese subjected to highly controlled experimental conditions may show many interesting results, especially if the suspected changes observed by chromatography can be confirmed by sensitive and specific analytical techniques for the compounds in question. The quantitative estimation of the changes taking place in peptides and amino acids during ripening and interpretations concerning their significance to quality and flavor of cheese present a strong challenge to workers in this field.

SUMMARY

A study was made of the quantitative aspects of free tyrosine, the total tyrosine liberated, and the tyramine content of commercial and experimental Cheddar cheese. In well ripened commercial Cheddar cheese amounts of free tyrosine as high as 2,630 γ and as low as 348 γ per gram of cheese, dry weight, were observed, and for experimental cheese similar values were obtained, but with a slightly greater range. The amount of free tyrosine is a variable whose concentration is dependent upon the difference in rates between proteolysis and decarboxylation.

Total tyrosine liberated in Cheddar cheese, a function of the activity of proteolytic enzymes, was not subject to the degree of variability that was observed for free tyrosine. In well ripened commercial Cheddar cheese total tyrosine liberated ranged in value from 1,341 to 2,021 γ per gram of cheese, dry weight. In experimental Cheddar cheese ripened for 176 days at 60° F. values as high as 3,583 γ per gram were obtained. Addition of *Streptococcus faecalis* organisms to commercial lactic acid starter did not materially increase the extent of total tyrosine liberation, but values were greater in experimental raw milk cheese than in their pasteurized milk counterparts.

The total tyrosine liberated in Cheddar cheese during most of the ripening period appears to be the result of hydrolysis at the expense of the lower peptides, appreciable amounts being liberated even after soluble protein had attained its maximum value. Total tyrosine liberated was considered to be a more sensitive criterion of cheese ripening than are soluble protein values.

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PASTEURIZATION EQUIVALENTS OF HIGH-TEMPERATURE SHORT-TIME HEATING WITH ICE CREAM MIX

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Previous studies (7) have indicated that ice cream mix may be pasteurized at temperatures above the boiling point with no deleterious effect on flavor. This is accomplished by heating rapidly to the desired temperature, keeping the holding time to a minimum, and cooling immediately. It is necessary in such a system to know the lowest temperature which, with no intended holding time, will result in bacterial destruction comparable to that obtained by accepted pasteurization methods. The flow diversion valve control may be set at this temperature to comply with minimum pasteurization requirements, even though the product may be processed at much higher temperatures at the option of the operator.

The primary purpose of this study was to determine what processing temperature with no intended holding time is necessary to cause destruction of selected heat-resistant bacteria equivalent to that obtained by laboratory pasteurization at 155° F. for 30 minutes.

EXPERIMENTAL PROCEDURE

Description of Roswell heater. The Roswell heater shown in Figure 1 was used in these studies. It consists of a single heating unit, two cooling units, and two regeneration units. The heating unit consists of a hollow stainless steel tubular element and the chamber housing it. The tubular element, shown par-
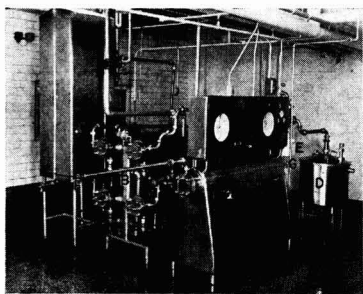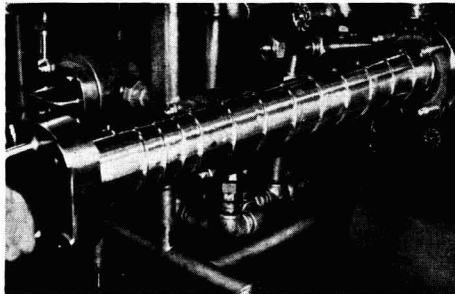
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FIG. 1. Front view of Roswell heater with accessory equipment. A—cabinet cooler; B—regenerator; C—heater; D—surge tank; E—flow diversion value (partially hidden); F—sampling port for raw mix; G—sampling port for heated mix with no intended holding time; H—sampling port for heated mix held for 3 seconds.

FIG. 2. Heating element showing spiraled heating surface partially withdrawn from the heating chamber.

tially withdrawn from the chamber in Figure 2, is 68 in. long and 4 in. in diameter. The element and chamber are both part of the heating system, as high pressure desuperheated steam is present inside the hollow element as well as in the chamber wall. In this manner heat is supplied to the product from both sides. The single-element heater used in this study had a heating capacity of 600,000 to 750,000 B.t.u. per hour.

To prevent "straight through" flow, the element is machined in such a manner that a $\frac{1}{8}$-in. ridge is spiraled around it; this directs the $\frac{1}{8}$-in. film of flowing product in a spiral fashion between the two heating surfaces. The liquid travels a total distance of 22 ft. in passing through the heating unit, which provides 11 sq. ft. of heating surface. The average time required for a particle of liquid to pass through the heating unit was found to be 1.8 seconds when the equipment was operated at a rate of 6,000 lb. per hour. The tolerances incorporated in the design of the heating unit are such that the volume of product making up the film within the heating unit is 1.5 qt.

The cooling and regeneration units of the heater are of the mix-to-water-to-mix type. They consisted of four tubular elements and chambers similar in design to the heating unit. Figure 3 shows the flow of product through the cooling and regeneration system. Although the regenerative unit is a standard part of this heater, in order to simplify the procedure in the experiments reported this feature was not used. Because of the various temperatures used in this study, the flow diversion valve was operated by manual control.

To determine accurately the processing temperatures, a copper-constantan thermocouple lead wire was permanently inserted in the T-joint connected directly to the outlet of the heating unit. The lead was installed in such a manner that the temperature was determined at the center of the flowing stream of mix at a point 3 in. from the heater outlet. A 3-second holding tube was installed for special study.

Preparation of inoculum and bacteriological procedures. Micrococcus sp. MS 102 *(1)* was used as the test organism to determine the equivalence point. The
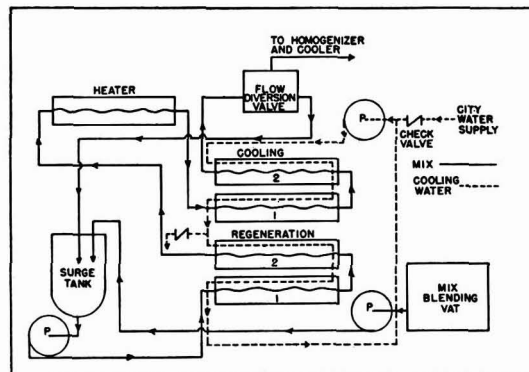
FIG. 3. Flow diagram showing flow of mix from mixing vat to homogenizer and cooler and cooling water from city water supply line to coolers and regenerators.

organism was grown on N-Z-Case[1] agar slants at 32° C. for 24 hours and stored at 40°-45° C. for 48 hours. Large slants of this medium were inoculated from the refrigerated stock culture and incubated at 32° C. for 22-26 hours. The cells were harvested in sterile distilled water and filtered through cotton to remove agar clumps. The filtered suspension was made up to 200 ml. with cold water and mixed in a chilled Waring blendor for 1 minute. To aid dispersion in the mix, sufficient water was added to make 1 liter of inoculum.

Standard bacteriological procedures were used in making plate counts. The initial 1:10 dilution of ice cream mix was prepared by weighing 11 g. of mix into a dilution blank containing 99 ml. of water. All dilutions were plated in duplicate (runs 1-5) or in quintuplicate (runs 6-11) with N-Z-Case medium. Plates were incubated at 32° C. for 72 hours.

Operating procedure during experimental trials. Fourteen hundred pounds of ice cream mix was used in each experimental trial. The mixes contained 12% butterfat and 38-40% total solids. Speck *et al. (4)* have shown that variations in total solids of this magnitude do not exert any significant effect on the inactivation rate of *Micrococcus* sp. MS 102. In preparation, the mix was preheated to 120°-130° F. and 1 l. of inoculum was added. The inoculum was dispersed by means of a funnel pump of the type used to reconstitute dry milk solids. It required approximately 10 minutes of agitation and recirculation to obtain a uniform dispersion of the test organism. An attempt was made in each trial to process mix at temperatures above and below that necessary to give bacterial destruction equivalent to that obtained by laboratory pasteurization at 155° F. for 30 minutes. The mix was processed at the highest temperature first.

Samples for bacteriological plating were taken simultaneously at three different locations with the sampling device described by Kaufmann *et al. (2)*. Sampling ports were located as shown in Figure 1. The raw mix sample was taken at *F;* a sample of the heated mix with no intended hold was obtained at *G*, and mix held for 3 seconds at the processing temperature was sampled at *H*. The duration of the sampling period was standardized to insure uniformity. The raw mix sample was divided into two portions; one was used to determine the initial count of the unheated mix; the other was laboratory pasteurized at 155° F. for 30 minutes. Since homogenization took place after pasteurization (Figure 3), all samples represent unhomogenized mix.

For laboratory pasteurization, 13 ml. of mix was placed in a sterile screw-cap test tube 5 in. long and 0.75 in. in diameter. Care was taken to minimize splashing on the sides of the tube above the liquid level. Mix which contaminated the lip of the tube during the pipetting process was charred by flaming before replacing the cap. To insure reproducibility of the heating and cooling process, ten tubes were always included in the laboratory pasteurization procedure, blanks containing water or mix being used to maintain the quota when the number of test samples was less than ten. One test tube containing mix was used as

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[1] Yeast extract, 1 g.; N-Z-Case, 0.5 g.; glucose, 0.5 g.; K_2HPO_4, 0.4 g.; KH_2PO_4, 0.1 g.; agar, 1.5 g.; water, 100 ml.

a thermometer well. The tubes were placed in a rack to facilitate handling. The tubes were immersed in a 4-in. deep water bath at 130° F. until the mix in the control tube reached this temperature. The rack and tubes were then immersed to the same level in a water bath at 160° F. until the temperature reached 155° F. This required approximately 2 minutes. The tubes were then transferred to a constant temperature water bath adjusted to 155° F. and submerged to the base of the screw-cap for 30 minutes. Rapid cooling was obtained by placing the tubes in an ice water bath. During cooling the tubes were inverted several times.

RESULTS AND DISCUSSION

Results shown in Table 1 indicate that a processing temperature of 187.2° F. with no intended holding time yields bacterial destruction of *Micrococcus* sp. MS 102 equivalent to that obtained by laboratory pasteurization at 155° F. for 30 minutes. A temperature of 181.3° F. with a 3-second holding time gave the same results. The equivalent temperatures were obtained in a similar manner as described by Tobias *et al. (5);* the process is illustrated in Figure 4, which is a plot of log log of the plate count (log of count plotted on semi-log paper) versus temperature at constant time. On this line were located points which corresponded to the log log of the plate count of the laboratory pasteurized samples and corresponding temperatures were read from the graph. Each run was treated separately.
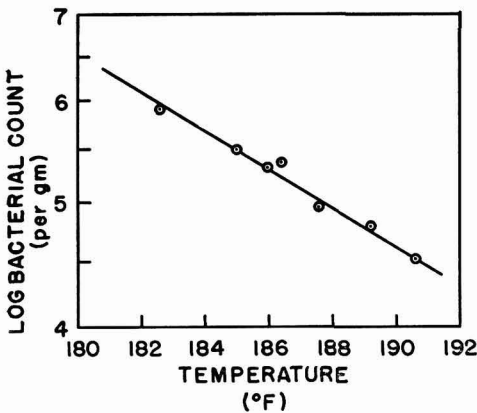
FIG. 4. Log log of bacterial counts versus temperature plot obtained after heating with no intended holding time (data from the last run in Table 1). Equivalent temperatures yielding the same destruction of MS 102 as laboratory pasteurization at 155° F. for 30 minutes were determined from these types of plots.
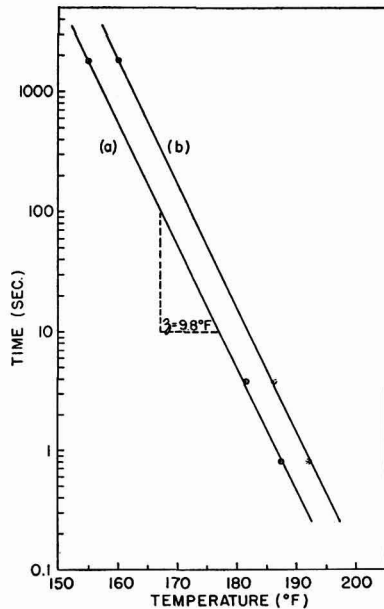
FIG. 5. (Right) Graph showing points of constant population of MS 102 at various time and temperature combinations. Lines pass through 155° F. for 30 minutes [a] and 160° F. for 30 minutes [b].

TABLE 1
Effect of different processing temperatures in the Roswell heater on the destruction of MS 102[a]

Plate count of inoculated mix[b] (per g.) (in thousands)	Processing temperature (° F.)	Plate count of heated mix		Plate count of mix after laboratory pasteurization		Av. process temperature giving destruction of test organism equiv. to lab. pasteurization	
		No hold (per g.)	3-second hold (per g.)	Uninoculated (per g.)	Inoculated[b] (in thousands)	No hold (° F.)	3-second hold (° F.)
820	199.1	60	30	60	53	<198.8	<198.8
	199.5	80	30				
	198.9	50	30				
	198.8	100	80				
2,100	200.6	260	90	1,900	170	<199.6	<199.6
	200.5	100	140				
	199.9	80	130				
	199.6	140	120				
	200.4	130	130				
3,200	194.8	400	120	190	80	188.9	<184.2
	188.7	65,000	300				
	187.0	460,000	130				
	185.3	1,040,000	3,800				
	184.2	1,200,000	11,000				
3,100	190.6	LA[c]	—	600	91	188.5	<185.9
	189.4	45,000	90				
	187.6	165,000	460				
	185.9	670,000	4,700				
2,900	187.6	240,000	LA	260	530	186.1	<181.6
	185.6	1,100,000	5,800				
	184.8	1,300,000	13,200				
	182.0	2,040,000	42,000				
	181.6	LA	131,000				
3,300	189.7	—	—	2,000	92	186.9	180.5
	188.5	27,000	SPR				
	188.1	39,000	SPR				
	186.9	90,000	SPR				
	184.9	450,000	SPR				
	182.9	910,000	28,000				
	180.7	890,000	69,000				

[a] Unit operated at a product flow rate of approximately 6,000 lb. per hour.
[b] Average count of from 4 to 7 replicates.
[c] LA = laboratory accident.

TABLE 1 (Concluded)

Effect of different processing temperatures in the Roswell heater on the destruction of MS 102[a]

Plate count of inoculated mix [b] (per g.) (in thousands)	Processing temperature (°F.)	Plate count of heated mix — No hold (per g.)	Plate count of heated mix — 3-second hold (per g.)	Plate count of mix after laboratory pasteurization — Uninoculated (per g.)	Plate count of mix after laboratory pasteurization — Inoculated [b] (per g.) (in thousands)	Av. process temperature giving destruction of test organism equiv. to lab. pasteurization — No hold (°F.)	Av. process temperature giving destruction of test organism equiv. to lab. pasteurization — 3-second hold (°F.)
1,600	192.0	4,000	—	SPR[d]	54	187.6	181.7
	189.4	27,000	—				
	187.4	134,000	510				
	186.4	95,000	4,000				
	185.0	129,000	15,000				
	184.1	440,000	14,000				
	180.9	640,000	79,000				
1,200	189.0	19,000	1,300		76	186.9	181.1
	188.0	44,000	—				
	185.8	170,000	2,400				
	184.6	300,000	6,000				
	183.8	380,000	14,000				
	181.0	730,000	94,000				
1,700	190.8	11,000	600	3,400	270	185.8	181.1
	190.0	40,000	380				
	186.6	220,000	1,200				
	185.4	340,000	3,200				
	183.7	440,000	19,000				
	182.4	470,000	100,000				
1,800	190.8	18,000	—	660	160	187.2	181.9
	188.4	90,000	800				
	187.4	110,000	1,200				
	185.8	360,000	2,300				
	185.1	780,000	9,200				
	183.8	1,100,000	32,000				
	182.5	1,500,000	14,000				
1,400	190.6	32,000	—	8,700	110	187.2	181.3
	189.2	61,000	6,000				
	187.6	92,000	1,300				
	186.4	250,000	22,000				
	186.0	210,000	11,000				
	185.0	320,000	17,000				
	182.6	800,000	57,000				
				Av.[e]		187.2	181.3

[d] SPR = spreader colonies making count impossible.

[e] Exclusive of runs 1 and 2 with no hold and runs 1 through 5 with 3-second hold,

From these considerations it is evident that 155° F. for 30 minutes, 181.3° F. for 3 seconds, and 187.2° F. with no intended holding time are points of constant bacterial population with respect to the test organism. Therefore, a plot of temperature versus log time should yield a straight line whose slope is the z value of the organism. However, before a plot of these three points could be made, it was necessary to determine the effective holding time when processing was performed with no intended hold. This effective holding time includes the combined lethal effects of heat-up time to processing temperature (0.5 second), discharge time from the heater (0.2 second), sample collection time, and cooling time (0.1 second), all expressed in terms of exposure to the processing temperature. It is obvious that the effective hold can never be zero, and the shorter it is the more difficult it becomes to ascertain its exact extent. In these studies it was estimated that the total effective hold was approximately 0.8 second (0.5 + 0.2 + 0.1). The plot in Figure 5 was made with this value as the time of exposure. The z value so obtained was found to be 9.8° F. Barber (1) reported a z value of 9.7° F. for the same organism. Since the effective holding time does not lend itself to direct measurement, it was thought desirable to calculate the z value, assuming an error of ±50% in the estimated hold. Under these conditions the z values obtained are 8.9° and 10.2° F., as compared with 9.8° F.

Some municipal pasteurization standards require heating to 160° F. for 30 minutes to pasteurize ice cream mix. The equivalent temperatures for this more rigorous standard also may be determined from the data collected in this study, as illustrated in Figure 4. A line passing through 160° F. for 30 minutes was drawn parallel to the one passing through 155° F. for 30 minutes. Theoretically, this line shows all time and temperature relationships giving the same destruction of *Micrococcus* sp. MS 102 as 160° F. for 30 minutes. At the 0.8-second and 3.8-second intercept the corresponding equivalent temperatures obtained from the plot are 192.7° and 186° F., respectively. An examination of the experimental data in Table 1 indicates an equivalent destruction at a temperature of approximately 194° F. at 0.8-second and 187° F. at 3.8-second hold; these values are in reasonably good agreement with those obtained from the plot. The reason an indirect approach was used to determine the temperature equivalents to 160° F. for 30 minutes is that the latter process allows less than 0.1% of the test organisms to survive. The ice cream mix used in these studies was not sterile prior to inoculation with the test organisms and generally contained some spore-formers, which, in low dilutions (1 to 10), exhibited spreader tendencies and inhibitory effects on *Micrococcus* sp. MS 102. It was difficult, therefore, to detect a few test organisms in the presence of an equal or a larger number of these spore-formers.

In order to determine the safety margin afforded by the equivalent temperatures found in this study, the lethality in terms of *Mycobacterium tuberculosis* was determined as proposed by Tobias *et al.* (5). The results are shown in Table 2. It can be seen that the HTST standard of 175° F. for 25 seconds has a lower lethality in terms of *Myc. tuberculosis* than the long-hold standard but still furnishes a good margin of safety. Theoretically, a value of one or over indicates that the heat treatment is of sufficient magnitude to destroy *Myc. tuberculosis*;

TABLE 2

Lethality of various time and temperature combinations in terms of Myc. tuberculosis *(1, 3)*

Temp.	Time	Lethality	Temp.	Time	Lethality
(° F.)	*(sec.)*		*(° F.)*	*(sec.)*	
155.0	1800	57.6	160.0	1800	144.0
175.0[a]	25	32.5	177.5[c]	30	60.0
187.2[a]	0.8	8.6	194.0[d]	0.8	32.8
181.3[a]	3.8	12.2	187.0[d]	3.8	39.9
194.0[b]	0.8	32.8	198.0[e]	0.8	69.0
186.0[b]	3.8	37.2	190.0[e]	3.8	72.2

[a] Time and temperature combinations giving destruction of MS 102 equivalent to that obtained by laboratory pasteurization at 155° F. for 30 minutes, taken from Table 1.

[b] Proposed time and temperature combinations.

[c] Time and temperature combination recommended *(6, 8)* as giving destruction of MS 102 equivalent to that obtained by laboratory pasteurization at 160° F. for 30 minutes.

[d] Time and temperature combinations calculated to give destruction of MS 102 equivalent to that obtained by pasteurization at 160° F. for 30 minutes.

[e] Proposed time and temperature combinations.

the margin of safety is given by the difference between the actual lethality value and one. The equivalent temperatures obtained from consideration of the data in Table 1 have lower lethalities than 175° F. for 25 seconds. The reason is that *Myc. tuberculosis* has a z value of 12.6° F. as compared with 9.8° F. for *Micrococcus* sp. MS 102. Thus, at some temperature higher than any considered in this study, the plots of log time versus temperature at constant population intersect. In order to maintain the same lethality as afforded by the HTST standard, the following time and temperature combinations should be used: 194° F. with an effective hold of 0.8 second or 186° F. with a 3-second hold or a total effective hold of 3.8 seconds. Similarly, where the 160° F. for 30 minutes pasteurization standard is enforced, 198° F. or 190° F. with an effective hold of 0.8 second or 3.8 seconds, respectively, should prove satisfactory.

The use of test organisms in evaluating the adequacy of any pasteurization process utilizing extremely high temperatures was found to be subject to a number of limitations. High temperatures exerted a deleterious effect on the growth of the surviving organisms, and it was necessary to increase the incubation period from 2 to 3 days in order to obtain maximum counts.

Another problem associated with HTST pasteurization arises from a consideration of the z value of *Micrococcus* sp. MS 102. This value is smaller than that of some pathogenic bacteria with the result that equivalence points at high temperatures do not afford the same margin of safety. It would be unwise to propose a pasteurization temperature with a smaller margin of safety than is provided by the accepted standards until positive direct evidence is presented to indicate that the standards are too rigorous. A new test organism which may overcome some of these limitations is now under study in this laboratory.

An evaluation of the processing procedure by means of lethality in terms of *Myc. tuberculosis* has resulted in a more rigorous requirement than that determined from the equivalent destruction of *Micrococcus* sp. MS 102. This technique is applicable to any pasteurization process and furnishes a numerical index of the margin of safety. Thus, it is possible to select the desired safety factor and

to calculate the temperature required to obtain it. Although this technique presents an excellent means of checking the adequacy of any heating process, it does not eliminate the need for experimentally evaluating various types of heaters, preferably with test organisms, to ascertain whether or not the heating characteristics are in accord with those theoretically postulated.

SUMMARY AND CONCLUSIONS

1. Equipment for heating and pasteurizing with no intended holding time possesses some effective holding time which is derived from the lethal effect of the heat-up time, discharge time, and cooling time. It was estimated that the Roswell heater used in these studies had an effective hold of 0.8 second at the processing temperature.

2. The destruction of *Micrococcus* sp. MS 102 equivalent to that resulting from laboratory pasteurization at 155° F. for 30 minutes was obtained at 187.2° F. or 181.3° F. with effective holding times of 0.8 second or 3.8 seconds, respectively.

3. The destruction of *Micrococcus* sp. MS 102 equivalent to that resulting from pasteurization at 160° F. for 30 minutes would be obtained at 194° F. or 187° F. with effective holding times of 0.8 second or 3.8 seconds, respectively, as shown by calculation.

4. Plotting log time of 1,800 seconds versus temperature at 155° F., 3.8 seconds at 181.3° F., and 0.8 second at 187.2° F. (points at which the population of *Micrococcus* sp. MS 102 is constant) yields a straight line with a z value of 9.8° F.

5. Because of the smaller z value of *Micrococcus* sp. MS 102 compared to that of *Myc. tuberculosis (1, 3)*, the margin of safety progressively decreases as higher temperature equivalents are obtained. A numerical index of the safety margin may be obtained from a calculation of the lethality in terms of *Myc. tuberculosis* by means of which time and temperature combinations may be postulated with any desired margin of safety.

6. A temperature of 194.0° F. for 0.8 second or 186.0° F. for 3.8 seconds yields the same lethality in terms of *Myc. tuberculosis* as 175.0° F. for 25 seconds. A temperature of 198.0° F. for 0.8 second or 190.0° F. for 3.8 seconds gives a lethality equivalent to that obtained at 177.5° F. for 30 seconds, conditions which have been found to give results comparable to pasteurization at 160° F. for 30 minutes *(6, 8)*.

ACKNOWLEDGMENTS

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SOME FACTORS AFFECTING THE QUANTITY OF WATER-INSOLUBLE FATTY ACIDS IN CREAM[1]

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Hillig *(3)* in 1947 published a method ''for determination of all water-insoluble fatty acids in butter or cream whether present as such or as alkaline salts and whether in the fat or the water phases of butter.'' The results obtained with butter were subsequently used as evidence of deterioration of cream prior to its use for the manufacture of butter. This situation created a need for information which would indicate some of the factors that might affect the results obtained by the use of this method as a measure of cream degradation. It was recognized that the results of a study of this nature would probably have more significance in areas where cream for churning is often produced in small quantities, marketed infrequently, shipped long distances, or collected at country buying stations. Since such cream is kept at widely varying temperatures and for varied periods of time, an important phase of the study was to determine some of the effects of time and temperature upon the water-insoluble fatty acid (WIA) content of cream. Hillig and Ahlman *(4)* have shown that the WIA in the cream, for the most part, are retained in the butter on churning. Butter from decomposed cream usually contained more WIA than butter from cream classified as satisfactory for butter making.

METHODS

Cream used in this study was obtained by the centrifugal separation of Grade A milk from the University dairy herd unless otherwise designated. The milk was warmed to 95°-100° F. prior to separation, and the cream was cooled in ice water with slow agitation to the temperature at which it was to be stored or laboratory determinations made.

The WIA values were determined by the method of Hillig *(3)*. Acidities were determined by titrating 17.5 ml. of milk or 9 g. of cream with 0.1 N NaOH to the phenolphthalein end point.

Butter was churned by agitation with a laboratory stirrer inserted in a quart Mason jar, approximately half filled with cream. The temperature was adjusted so that churning required 10-20 minutes. Churning was stopped when the granules were approximately the size of wheat kernels, the buttermilk was drained, and the butter granules were washed in two portions of ice water. The granules were gathered together with a spatula, and any excess water was drained. The Babcock method was used for determining butterfat in milk and cream, and a modified procedure with a 6-g. 90% bottle was used for butter.

In the determination of lipase activity, a homogenized 18% fat cream-sucrose substrate containing 42.5% sucrose was used. Twenty grams of the product was added to 200 g. of the substrate and held at 60° F. The increase in titratable acidity was used as a measure of the lipolytic activity. The substrate plus 20 ml. of distilled H_2O was used as a control. Parmelee and Babel (13) noted that saturation of cream of 35% fat with sucrose restricted bacterial growth but did not appear to reduce lipase activity.

Fat acidities were determined on 15.5 g. of the butter oil at 90°-95° F. from melted centrifuged butter samples with $0.1 N$ alcoholic KOH, a micro burette, and neutral phenolphthalein as the indicator. The end point was taken as a distinct red. The results were expressed as milliliters $0.1 N$ alcoholic KOH per 100 g. of fat.

<div align="center">RESULTS</div>

Effect of temperature. Fresh centrifugally separated cream was held at 40°-45° F., 55°-60° F., and 70°-72° F. for 2, 4, and 6 days. The titratable acidity of the cream and the quantity of WIA were determined in the fresh cream and after holding. The data are shown in Table 1.

<div align="center">TABLE 1

Titratable acidities and water-insoluble fatty acids in six lots of cream held for varying times at different temperatures</div>

Description of samples	Titratable acidity		WIA per 100 g. fat	
	Range	Average[a]	Range	Average[a]
	(%)	*(%)*	*(mg.)*	*(mg.)*
Fresh	0.10-0.12	0.11	19-161	102
2 days 40°-45° F.	0.11-0.17	0.15	72-617	277
2 days 55°-60° F.	0.12-0.57	0.27	29-304	164
2 days 70°-72° F.	0.35-0.66	0.56	49-477	148
4 days 40°-45° F.	0.12-0.32	0.19	74-589	283
4 days 55°-60° F.	0.17-0.61	0.38	36-256	124
4 days 70°-72° F.	0.55-0.75	0.66	75-503	298
6 days 40°-45° F.	0.12-0.38	0.22	25-739	308
6 days 55°-60° F.	0.40-0.61	0.53	28-273	143
6 days 70°-72° F.	0.58-0.75	0.66	229-1042	598

[a] Average results for the six lots of cream.

The increase in WIA was more rapid in the cream held at 40°-45° F. than in that held at 55°-60° F. at the end of 2, 4, and 6 days. The increase was greater at 70°-72° F. after 4 and 6 days than at 55°-60° F. Similar results were obtained with 10 lots of cream held 2, 4, and 6 days at 40°-45° F., 55°-60° F., and/or 70°-72° F. Since some of these lots were held only at two of the three temperatures, only the results with the samples held at all three temperatures were included in Table 1.

Decreases in WIA values were noted in some instances. In the 40°-45° F. group the values for two samples were lower at 6 days than at 4 days. In the 55°-60° F. group one sample was lower at 2 days than the fresh sample, two samples were lower after 4 days, and one sample after 6 days.

Effect of acidification of cream. In seeking an explanation for the differences observed at the three temperatures, the acidity of cream was increased by the addition of lactic acid, and WIA were determined in the acidified and unacidified samples. The results are summarized in Table 2.

TABLE 2

The effect of added lactic acid upon the yield of water-insoluble
fatty acids from 27 samples of cream

Description of samples	Fat range	Titratable range	Acidity average[a]	WIA per 100 g. fat	
				Range	Average[a]
	(%)	*(%)*	*(%)*	*(mg.)*	*(mg.)*
Control	19.5-62.0	0.10-0.38	0.13	46-342	159
Acidified	19.5-62.0	0.26-0.60	0.50	5-304	87

[a] Average results for the 27 lots of cream.

In 22 of the 27 samples to which lactic acid had been added, lower WIA values were obtained after acidification, and in two samples no change occurred; nevertheless, the mean difference was 6.2 times its standard deviation. The addition of lactic acid interfered in the removal of fatty acids by the Hillig procedure. The observed effects resulting from the addition of lactic acid suggested determining the effect of time and temperature on the WIA content of acidified cream (Table 3).

TABLE 3

The effect of age upon the yield of water-insoluble fatty acids from
unacidified cream and the same cream after acidification

Description of sample	No. of samples	Temperature held	Average titratable acidity				Average WIA per 100 g. fat			
			Fresh	2 days	4 days	6 days	Fresh	2 days	4 days	6 days
		(° F.)	*(%)*	*(%)*	*(%)*	*(%)*	*(mg.)*	*(mg.)*	*(mg.)*	*(mg.)*
Control	2	40-45	0.12	0.15	0.15	0.16	155	396	366	418
Acidified	2	40-45	0.53	0.48	0.49	0.46	27	18	24	17
Control	2	55-60	0.12	0.16	0.32	0.56	155	240	227	260
Acidified	2	55-60	0.53	0.48	0.48	0.49	27	20	21	108
Control	2	70-72	0.12	0.57	0.72	0.74	155	292	439	709
Acidified	2	70-72	0.53	0.48	0.52	0.72	27	25	309	375
Control	4	40-45	0.12	0.14	0.16	0.17	194	264	361	350
Acidified	4	40-45	0.51	0.49	0.49	0.48	54	28	27	18
Control	4	70-72	0.12	0.60	0.70	0.70	194	401	528	743
Acidified	4	70-72	0.51	0.50	0.52	0.69	54	97	298	2766

In comparing the results for the two samples containing 29.5% butterfat and held at all three temperatures for 2, 4, and 6 days there was no increase in WIA in the acidified cream held at 40°-45° F. or at 55°-60° F. except for one sample, which increased after 6 days at the latter temperature. At 70°-72° F. an increase was observed after 4 days, and a material increase after 6 days. Two additional samples containing 32.5% butterfat were held at 40°-45° F. and 70°-72° F. No increase in WIA occurred in the four acidified samples held at 40°-45° F. for 6 days. At 70°-72° F. two showed an increase after 2 days, all showed increases

after 4 days, and at 6 days all the values greatly exceeded those for the unacidi-
fied cream. Mold growth was observed on the surface of two of these samples on
the 4th day, and this was assumed to account for the high values since mold
growth had been previously found to be associated with high yields of WIA.

In order to further observe the effects of acidification, portions of acidified and
unacidified cream were churned and WIA determinations were made on the
cream and the butter. These results are shown in Table 4. The results indicated

TABLE 4

*The water-insoluble fatty acid content of 10 lots of acidified and
unacidified cream and the butter churned from the cream*

Description of sample	Titratable acidity		WIA per 100 g. fat	
	Range	Average[a]	Range	Average[a]
	(%)	*(%)*	*(mg.)*	*(mg.)*
Control cream	0.10-0.20	0.12	87-187	147
Butter			108-162	133
Acidified cream	0.40-0.57	0.52	9-175	81
Butter			67-165	127

[a] Average results for the 10 lots of cream.

that the inhibiting effects of lactic acid on the separation of WIA from cream
were not equally effective when determining the WIA in butter churned from
that cream.

Effect of ripening cream with starter. The effect of the addition of lactic acid
and aging at 55°-60° F. upon the WIA content of cream prompted the deter-
mination of the effect of increased acidity resulting from the addition of starter.
Inoculation with starter was at the rate of 0.1% and 0.3%, after which the
samples including an uninoculated control were held 2, 4, and 6 days at 40°-45°,
55°-60°, and 70°-72° F.

Titratable acidities were higher and the WIA values were lower for the sam-
ples to which starter had been added after 2 and 4 days at each temperature.
The differences in WIA were not great in most instances and were not of the
magnitude previously observed for acidified and unacidified cream. After 6 days
an equal number of the samples containing starter were higher and lower in WIA
values when compared with the controls.

Effect of agitation of milk. During the progress of these experiments it was
noted that the quantity of WIA in the fresh cream separated from milk produced
by the University Dairy herd was materially higher than they previously had
been. Cream from other sources did not show this change. The method of han-
dling the University milk had been changed from tank cooling in 10-gal. cans to
the use of a pipe line milker and the cooling of the milk in a 500-gal. cold wall vat.
The daily production of the herd at this time was 1,500 to 1,800 lb. Tables 5 and
6 show the effect of agitation of milk upon the quantity of WIA in fresh cream
separated from the milk.

The agitation of milk in the process of cooling in a cold wall tank, or with a
laboratory stirrer, increased the WIA content of the freshly separated cream.

TABLE 5

The effect of the method of cooling milk upon the water-insoluble fatty acid content of fresh cream separated from that milk

Milk source	No. of trials	WIA per 100 g. fat		Method of cooling milk
		Range	Average	
		(mg.)	*(mg.)*	
U. of N. dairy herd	11	371-960	703	Cold wall tank Agitator 50 r.p.m.[a]
	9	165-529	300	Agitator 50 r.p.m.[b]
	5	171-271	247	Agitator 27 r.p.m.[a]
	9	19-295	133	In cans
U. of N. MSH herd	12	39-152	103	In cans
Farm producers	24	10-214	129	In cans

[a] Agitator thermostatically controlled.
[b] Agitator manually controlled to cool milk to 35° F.

TABLE 6

The immediate effect of agitation of milk with a propeller type laboratory stirrer upon the water-insoluble fatty acid content of fresh separated cream

No. of trials	Quantity of milk	Time agitated	Temperature agitated	Average WIA per 100 g. fat	
				Before agitation	After agitation
	(qt.)	*(min.)*	*(° F.)*	*(mg.)*	*(mg.)*
6	3	30	52	224	606
3	3	30	52	215	502
3	3	30	100	215	614

Increasing the temperature at which agitation took place in the laboratory, or increasing the speed, or time, of agitation in the cold wall tank resulted in material increases in the WIA content of the cream.

Differences in the WIA content of the fresh cream separated from the three sources of milk cooled in cans were well within the range of differences observed for cream separated at different times from the same source. The cream from the milk which had been agitated with a laboratory stirrer showed increased lipolytic activity when measured in terms of increase in titratable acidity of a pasteurized-homogenized cream-sucrose substrate to which the cream had been added and held at 60° F. The results are shown in Table 7.

The lipolytic activity of the freshly separated cream from the milk from the three sources, University Dairy herd (DH), University Milking Shorthorn herd (AH), and non-Grade A (C) producers, was not increased to the same extent by agitation of the milk. The cream from the University Dairy herd milk showed the greatest effect from agitation.

This effect of agitation on lipolytic activity was found to be a possible source of error in WIA determinations when comparisons were made between unpasteurized cream and laboratory churned butter, depending upon the type and extent of agitation used in churning the cream. In three trials 50 g. of cream were weighed in duplicate into centrifuge bottles in preparation for WIA determinations. One pair of each duplicate was churned in the centrifuge bottle with

TABLE 7

Lipolytic activity of cream from agitated and unagitated milk, as measured in terms of increase in titratable acidity of a homogenized cream-sucrose substrate to which the cream had been added

	Titratable acidity				
	Fresh	46 hours	113 hours	162 hours	234 hours
	(%)	*(%)*	*(%)*	*(%)*	*(%)*
Substrate + H₂O	0.067	0.070	0.070	0.072	0.081
Substrate + unag. AH		0.090	0.120	0.138	0.155
Substrate + ag. AH		0.105	0.134	0.157	0.180
Substrate + unag. C		0.100	0.130	0.151	0.170
Substrate + ag. C		0.113	0.153	0.171	0.198
Substrate + unag. DH		0.100	0.123	0.142	0.160
Substrate + ag. DH		0.130	0.170	0.198	0.225
Substrate + 15 mg. steapsin		0.225	0.430	0.485	0.560

a laboratory stirrer prior to the determination. The results of the churned samples were 55 mg. (271 to 326), 44 mg. (361 to 405), and 21 mg. (324 to 345) higher, respectively, than the results for the unchurned samples.

Effect of the method of separation and fat content of the cream. After observing these effects of agitation it was thought that centrifugal separation of cream might have a similar effect. Four lots of milk were divided into two portions each, one of which was hand skimmed after 4 hours and the other was centrifugally separated. In each case the WIA content of the centrifugally separated cream was higher than that of the hand skimmed cream. It was found, however, that the higher WIA values for the centrifugally separated cream were apparently related to the per cent of fat in the cream rather than to the effect of centrifugal force. This conclusion was based upon the results of which those given in Table 8 are illustrative.

Effect of individual cows. The milk from two groups of cows, a pasture-fed group and a dry-fed group, was available. The dry-fed group was on nonsucculent feed continuously and the other group was on pasture during the pasture season and received liberal amounts of silage during the nonpasture season. Four

TABLE 8

Effect of method of separation and the fat content of cream upon the yield of water-insoluble fatty acids

Description of sample	Fat	WIA per 100 g. fat
	(%)	*(mg.)*
Hand skimmed	22.0	168
Centrifugally separated		
40 r.p.m. (normal speed 65 r.p.m.)	16.0	181
66-70 r.p.m.	52.0	224
40 r.p.m.	26.5	192
70 r.p.m.	57.5	229
70 r.p.m. + skim	18.0	169
40 r.p.m.	14.0	143
40 r.p.m. concentrated[a]	31.0	175
70 r.p.m.	61.5	258
70 r.p.m. + skim	31.0	180
70 r.p.m. + skim	15.0	107

[a] By centrifuging and removing a portion of the skimmilk layer.

cows of approximately the same age and stage of lactation were selected from these groups. The milk from each cow was centrifugally separated without cooling. Water-insoluble fatty acids were determined in the fresh cream and after aging.

Analysis of variance between the groups showed no significant relationships between groups or between dates. Calculation of correlation coefficients showed no significant correlation between milk fat percentage and WIA or between milk acidity and WIA in either the fresh or aged samples.

The fresh milk from six cows with one or more quarters containing mastitis infection was separated and WIA were determined in the fresh cream and after 4 days at 55°-60° F. The results were comparable to those obtained for fresh cream from the milk of cows with normal udders in the feeding experiment.

Comparison of WIA in cream and butter. In the progress of these studies comparisons were made between the WIA content of cream and the butter churned from it. The results of these determinations have been grouped in Table 9 according to the type of cream.

In each trial the WIA content of the butter from the raw starter-soured cream and the pasteurized sweet cream was less than the WIA content of the cream. In the raw sweet cream, pasteurized-neutralized cream, and the sour acidified cream trials half of the butter WIA values were greater than the cream WIA values, and half were less by approximately the same amount. The WIA values in the butter in the raw sour cream and the acidified pasteurized-neutralized cream trials are sufficiently larger than those of the cream to indicate a trend. A less pronounced but similar trend may be noted for the pasteurized-starter–soured and sweet acidified cream trials.

Relation of fat acidities to WIA values. Fat acidities in terms of milliliters of 0.1 N alcoholic KOH per 100 g. fat were determined and correlated with the WIA values of the cream by the Hillig procedure. The regression line for these data is shown in Figure 1. For the entire data the coefficient r = 0.9012; for the data where the WIA values are 200 or over, r = 0.9002; and for WIA values less than 200, r = 0.3938.

DISCUSSION

The use of low temperatures for holding cream did not assure that WIA would remain at low levels. Peters *et al. (15)* had also found marked increases in WIA in cream held up to 10 days at 38° F. Weckel *(18)* stated that lipase may be quite active in milk held at refrigerator temperatures. Hillig and North *(6)* found no pronounced increase in WIA in cream held at 4° C. until organoleptic classification would condemn its use for butter manufacture; however, rapid deterioration took place when the cream was moved from 4° C. to 25° C. Babel *(1)* reported no significant difference in the WIA content of butter made from portions of the same cream held 10 days at 55° to 75° F. The report of Parmelee and Babel *(13)* that creameries having difficulties with high WIA tended to have higher values in the late winter months implies an effect of low temperatures. Later *(14)* they reported that cream held at 50° F. may contain more WIA than

TABLE 9

Comparative yield of water-insoluble fatty acids from cream and from laboratory churned butter from the same cream

Description of cream	No. of samples	Titratable acidity		WIA per 100 g. fat				Average increase or decrease in butter			
		Initial	After neut. or acid.	Cream		Butter		Increase		Decrease	
				Range	Av.	Av.		No. of samples	WIA	No. of samples	WIA
		(%)	*(%)*	*(mg.)*	*(mg.)*	*(mg.)*			*(mg.)*		*(mg.)*
Raw sweet	18	0.19-0.14		87-632	193	195		9	30	9	25
Raw sour	19	0.38-0.73		52-1048	304	357		9	158	10	32
Raw, starter soured	5	0.41-0.57		161-270	208	187		0		5	21
Past. sweet	15	0.10-0.20		120-342	209	142		0		15	25
Past. neutralized	8	0.43-0.50	0.08-0.27	42-345	188	191		4	33	4	28
Past. starter soured	5	0.43-0.61		45-288	165	174		2	72	3	34
Sweet, acidified	22	0.10-0.20	0.26-0.57	9-680	164	178		10	73	12	35
Sour, acidified	6	0.33-0.57	0.37-0.59	57-694	311	318		3	64	3	51
Acidified, past., neut.	2	0.44-0.55	0.12-0.17	46-47	46.5	214		2	168	0	
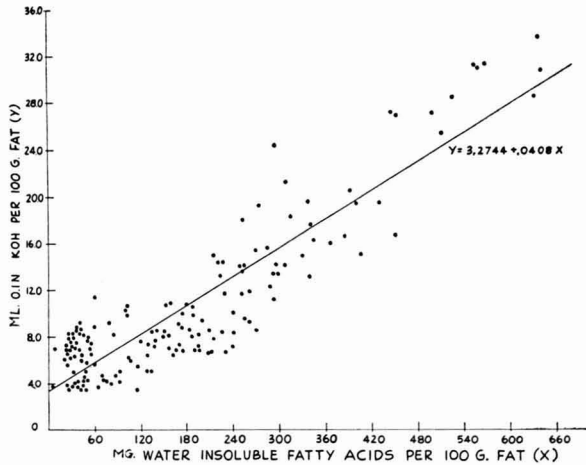
FIG. 1. Distribution and regression of water-insoluble fatty acids and titratable acidity of fat.

cream held at 70° F. The lipolytic effect at low temperatures may result from the favorable conditions for lipolytic bacteria and the slower rate of acid development, which favors the action of natural lipases and those of microbial origin. If the natural lipase activity of the milk is high, a rapid increase in WIA would be expected. This natural lipase action would be supplemented by that produced by microorganisms at the low temperatures (11, 12).

The instances in which decreases in WIA values occurred suggested possible errors in the method of analysis, utilization of free fatty acids by microorganisms, or an effect of developed acidity. The possibility of error in analysis was ruled out after numerous redeterminations to verify these differences, which had been observed in many instances not included with these data. The work of Jezeski et al. (9) supports the suggestion of utilization of fatty acids by microorganisms. Lubert et al. (10) found no microorganisms that produced a bacterial lipase having an optimum activity on the acid side of neutrality. Hood (8) stated that at higher temperatures the activity of bacteria either checks the lipase or uses the fatty acids released. The inhibitory effect of developed acidity on the action of the natural lipases or upon the extraction procedure was considered as a possible factor affecting the results, although the titratable acidities were only 0.13% and 0.32% in the two samples with lower WIA values in the 40°-45° F. group. Peters et al. (16) also reported declines in WIA during the holding of cream and suggested the definite possibility of the utilization of a portion of the acids by microorganisms.

The yield of WIA was reduced in many instances immediately after the acidification of the cream with lactic acid, but this reduction was not evidenced to the same extent in the laboratory-churned butter from the cream. The most satisfactory explanation for the reduction is that the fatty acids were not completely extracted under these conditions.

The low rate or absence of WIA production in acidified cream held at 40°-45° or 55°-60° F. may have resulted from the unfavorable pH for lipolysis or from the addition of lactic acid, which interfered with the extraction of the WIA. The optimum pH range for milk lipase has been reported as 8 to 9 (2, 8, 17), and it is also noted that low pH adversely and permanently affects lipase activity (2). At 70°-72° F. mold growth was observed in some samples on the surface of the acidified cream after 4 days. In all these cases the WIA was high. Hillig and Ahlman (4) found no significant change in WIA in storage butter except where visible mold appeared.

The addition of starter to cream resulted in a decreased rate of WIA production, but not to the extent which resulted from the addition of lactic acid. This supports the conclusion that the effect of added lactic acid was more than a lowering of the pH.

The observance of increased lipolytic activity resulting from agitation is not new (8), but data are presented which show the magnitude of the effect. The production of high WIA values as found here in sweet cream from high quality milk under bulk cooling conditions has significance since high WIA is usually associated with deterioration resulting from microbial growth. The possibility of error in WIA determinations as a result of agitation in laboratory churning of raw cream has been shown. The increase in the acidity of a cream-sucrose substrate to which cream from the agitated milk was added gave an indication of the accelerated lipolytic activity resulting from agitation.

No differences in WIA in fresh or aged cream were found that could be attributed to the effect of pasture or dry feeding of cows or to mastitis infection. This observation of the effect of pasture agrees with Hillig and Palmer (7) but not with Parmelee and Babel (14). There were no apparent differences in the initial WIA of sweet cream from three sources of Grade A milk and one source of non-Grade A milk.

The results of the direct titration of the centrifuged oiled-off fat from churned cream, with 0.1 N alcoholic KOH and phenolphthalein as the indicator, were compared with actual WIA values. The advantages of this procedure are its simplicity and rapidity. The statistical analysis of the results indicated the extent of its usefulness in elimination of high WIA samples.

Based upon the values obtained, samples of butterfat containing 400 mg. WIA per 100 g. would have an average titration value of 20.38 ml., 1% of the titrations of these individual samples could be expected to be 12.6 ml. or lower, and 1% to be 28.1 ml. or higher. The standard error of the titration for butterfat with a WIA value of 400 was 3.25 ml. Therefore, if a titration of 12.6 ml. is used to reject samples because of a presumed high WIA, then more than 99% of butter samples with an actual WIA of 400 or higher would be rejected. On the other hand, some butter samples with less than 400 WIA would also be rejected. In the particular group of samples for which data are presented, 28 of the 159 samples would have been rejected on the basis of titration above 12.6 ml. even though the WIA was actually less than 400 mg. This frequency of rejection of samples with less than 400 WIA, and the necessity for verification of the results

by a more exact procedure, would depend upon the proportion of these samples near the 400 mg. level.

The 28 samples rejected on the basis of the titration but which should not have been rejected on the basis of their actual WIA content contained 214 to 394 mg. WIA, whereas none of those under 214 mg. were rejected by the titration.

The use of the titration procedure to determine WIA resulted in the rejection of a relatively high proportion of butter samples in the WIA range of 214-400 mg. Therefore, the usefulness of this method would depend upon the proportion of butter moving through commercial channels that is within the 214-400 mg. range. To give an actual illustration of this the correlation analysis reported above was applied to the WIA determinations on 400 authentic samples of butter as reported by Hillig *et al. (5)* and by Hillig and Ahlman *(4)*. There were 208 samples in the WIA range of 150 to 400. Of this number 124 would have been rejected on the basis of 12.6 ml. as the maximum permissible. Of the remaining samples 81 were under 150 WIA and 111 over 400 WIA. More than 99% of these would have been accurately accepted or rejected on the basis of the titration.

SUMMARY

Holding cream at low temperatures did not assure a low WIA content.

Reduction in WIA levels during the holding of cream at 40°-45° F. and 55°-60° F. were obtained.

Extraction of fatty acids by the Hillig procedure from cream acidified with lactic acid was incomplete.

High WIA values in cream resulted from excessive agitation of milk during cooling.

The accuracy of identification of cream with a high WIA content by direct titration of the fat has been shown.

ACKNOWLEDGMENT

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MICROBIOLOGY OF THE SURFACE RIPENING OF BRICK CHEESE[1,2]

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The flavor of mild, cured brick cheese is one of its most important characteristics, yet comparatively little is known concerning the causes of the desired flavor or methods of producing it. The characteristic brick flavor has been attributed chiefly to the action of the microbial flora that grows as a "smear" on the surface of the cheese during the early part of ripening. Langhus *et al.* *(12)* reported growth of yeasts, micrococci, and a rod-shaped bacterium in that order, but did not identify them. Iya and Frazier *(9)* identified the film yeasts isolated from brick cheese smears at different factories as species of *Mycoderma*.

The surface flora of other varieties of surface ripened cheeses has been studied, but few of the microorganisms concerned have been identified. Usually, yeasts, micrococci, and bacilli were reported. Grimmer and Aronson *(7)* found the yellow-pigmented *Micrococcus flavus*, *M. citreus*, *M. sulfureus*, and *M. aurantiacus* on the surface of Tilsit cheese. Janiak *(10)* found the yeasts on Tilsit and Limburger cheese to be species of *Torula* and *Mycoderma* and one of the rods to be *Bacterium linens*. *B. linens* has been found in cheese smears by numerous other workers. Recently Hartley and Jezeski *(8)*, working with the slime on curing blue cheese, found yeasts and mold, micrococci, and rods appearing in that order. On cheese ripened at 46-49° F. the rod was *B. erythrogenes*, and on cheese cured at 55-58° F. it was *B. linens*.

The color of the smear on cheese surfaces may vary from yellow through orange to an almost reddish tinge. The yellow color has been attributed, in part at least, to pigmented micrococci, but the orange to reddish brown shades have been attributed to *B. linens* by a long series of workers, including Weigmann *(15)*, Grimmer and Aronson *(7)*, and Albert *et al.* *(1)*.

A number of workers have concluded that the enzymes of the surface flora of cheese were not significant in the splitting of protein within cheese of the following varieties: brick *(12)*, Gruyère and Tilsit *(11)*, Wilster Marschkäse *(2)*, Belpaese *(16)*, and Trappist *(6)*. Most workers agree, however, that the surface flora contributes to the flavor of the cheese, although the chemical compounds responsible for the flavor have not been identified.

METHODS

Cheeses from which isolations of pure cultures were made came from three factories in Wisconsin, from the Department of Dairy and Food Industries of

this University, and from the curing rooms of the Kraft Foods Company of Wisconsin at Beaver Dam. The cheeses were 7 days old when received from outside and were cured to completion in curing rooms here.

Manufacture of cheese. Cheeses to be inoculated on the surface with selected isolated cultures were made in the University Dairy by means of the ''sweet curd'' method recommended by Buyens (4) from milk pasteurized at 162° F. for 16 seconds. Before use, all equipment that would contact the milk or cheese was treated for at least 5 minutes with a hypochlorite solution containing 200 p.p.m. of available chlorine. To increase the number of cheeses that could be cured at once, normal-sized cheeses were halved by means of a sterilized metal follower covered with sterilized Viscon paper, while the cheese was in the hoop and after it had been turned the second time. Because of the increased ratio of surface area to volume, the time of salting was reduced from 24 to 20 hours.

The cheeses rested on sterilized aluminum trays in the curing chamber, which was a low temperature incubator made from a refrigerator. The humidity was kept high by means of sheets of cheesecloth draped along the sides and across the bottom of the refrigerator, with the ends of the sheets resting in water-filled trays. A thin coating of mineral oil prevented an accumulation of ice on the refrigerating unit. The curing temperature was 60° F.

Inoculation of cheese. The inoculation of film yeasts onto the surface of the cheeses was accomplished by the use of pasteurized 22% brine, inoculated with a suspension of the selected yeast and used in lieu of the usual brine or salt bath. When each cheese was removed from the brine it received an additional inoculum of film yeast suspension, of micrococci, of *B. linens*, or of combinations of these organisms. This inoculum was rubbed over the surface of the cheese by an operator wearing sterilized rubber gloves. The cultures for the preparation of inocula were grown originally in broth on a rotary shaker; and the cells were collected from 100 ml. of culture by centrifugation, washed twice with physiological salt solution, and resuspended in 10 ml. of the saline solution.

The yeasts were grown in a broth, hereafter called Iya broth:

KH_2PO_4	0.5 g.	Bacto-Tryptone	5 g.
$NaNH_4HPO_4 \cdot 4H_2O$	1.0 g.	Liver extract*	100 ml.
Cerelose	50.0 g.	Carrot extract*	100 ml.
NaCl	3.0 g.	Tap water	800 ml.

* One pound of ground, fresh beef liver is steamed with 1 l. of distilled water for 30 min. After filtration through several layers of cheesecloth, the extract is bottled and sterilized for 20 min. at 121° C. The carrot extract is prepared in a similar manner.

The micrococci and *B. linens* were grown in a broth of the following formula: 5 g. glucose, 10 g. Bacto-Tryptone, 5 g. Bacto-Yeast Extract, 2 g. K_2HPO_4, 1,000 ml. distilled water; pH 6.9-7.1.

Handling during ripening. Every other day during the 2-week ripening period the cheeses were rubbed and turned, the operator wearing sterilized rubber gloves previously moistened with sterile water.

Sampling during ripening. Samples were taken just before the cheese was handled so that 48 hours of undisturbed growth had been permitted. A thin piece of rind was cut from the upper surface of the cheese with a sterilized scalpel, deposited in a sterilized petri dish, trimmed to an area of 1 sq. cm., and then shaken in 10 ml. of 0.5% sodium citrate solution. After dispersion of the rind further dilutions were made as desired.

To prepare contact slides for the microscopic examination of the surface flora, glass slides were pressed against the surface of the cheese and the adhering material was spread uniformly over the slide with the aid of a loopful of water. The slides were dried, defatted with xylene, fixed with alcohol, and stained by the Hucker modification of the Gram method.

Grading of ripened cheese. After the cheese had been paraffined it was cured at about 40° F. for 6 weeks and then graded by three judges, who evaluated the flavor and total grade of the cheese numerically as follows:

1. Excellent
2. Desirable
3. Satisfactory
4. Objectionable
5. Very objectionable
6. Not salable as original cheese

Grading was within half points, and results were expressed as averages of the points allotted by the respective judges. Special attention was paid to the evaluation of flavor. The authors joined the three judges in this evaluation.

Isolation and identification of cultures. The micrococci and yeasts were isolated by plating in appropriate media and were purified by streaking procedures. No attempt was made to identify the yeasts as to genus. Purified cultures of micrococci were studied by means of methods described in the *Manual of Methods for the Pure Culture Study of Bacteria (14)* and identified as to species on the basis of descriptions in *Bergey's Manual of Determinative Bacteriology (3)*. Organisms were tested for their proteolytic ability on milk agar, which was flooded with 1% HCl after incubation. Tests for lipolysis were made on agar containing tributyrin (Bacto-Tryptone, 10 g.; Bacto-Yeast Extract, 3g.; K_2HPO_4, 5 g.; tributyrin emulsion, 50 g.; agar, 15 g.; distilled water, 1,000 ml., pH 5.5) and on the same basal medium with tributyrin replaced by Nile blue sulfate-saturated butterfat.

Effect of yeasts on micrococci. Two selected yeast cultures (B6 and B7) were grown with *M. caseolyticus* culture (Mc32) in milk at pH 4.9 and 6.4 to show the influence of the yeasts on the growth of the micrococci, especially under acid conditions. Also, autolysates of washed yeast cells were tested for their effect on the growth of three species of *Micrococcus*.

Tests on odor production. Seven kinds of culture media were inoculated with pure cultures of yeasts and micrococci from brick cheese and observed for the sweaty odor characteristic of the brick cheese smear. These media included: skimmilk, whole milk, Bacto-Casitone (0.5%) broth plus Bacto-Yeast Extract (0.01%), Bacto-nutrient broth with and without 3% melted and filtered butterfat, and a broth containing 0.5% Bacto-Tryptone and 0.3% beef extract, with

and without 3% melted and filtered butterfat. Also, whole milk was inoculated with a mixture of two yeasts and a micrococcus. The lower volatile saturated fatty acids produced by some of the cultures were estimated by partition chromotography by the methods of Ramsey and Patterson *(13)* and Claborn and Patterson *(5)*.

<center>RESULTS</center>

The 136 cultures of film yeasts isolated from cheese smears and brines fall into four groups on the basis of the shape of the cells and the type of film formed on liquid media. No attempt was made to identify the yeasts, but a representative (B7) of the most commonly occurring group and a yeast that was both proteolytic and lipolytic (B6) were selected for further study. The majority of the film yeasts, as represented by B7, were neither proteolytic nor lipolytic. Iya and Frazier *(9)* had identified the predominant film yeasts of smears as species of *Mycoderma*.

The 329 cultures of micrococci isolated from cheese smears were classified into several groups, representatives of which were studied in detail to identify them as to species. The species found most often was a colorless variant of *M. varians* (Mv), which made up 80% of all cultures isolated and was found in all cheese smears tested. Found less frequently were *M. caseolyticus* (Mc) and *M. freudenreichii* (Mf).

Associative growth of film yeasts and micrococci. Since the film yeasts of cheese smears have been found by Iya and Frazier *(9)* to have a stimulating effect on *B. linens*, it was suspected that the micrococci of the smear might be stimulated in a similar manner. All of the micrococci isolated from the smears were found able to grow at pH levels of 4.8 to 5.5 in broth, a pH range similar to that on the surface of freshly made brick cheese, but developed at an increased rate as the pH approached neutrality. Therefore, the reduction of the acidity of the cheese at the surface, accomplished by film yeasts according to Iya and Frazier, might be expected to favor the growth of the micrococci. This was demonstrated by growing a culture of *M. caseolyticus* (Mc32) in skimmilk at 30° C. with and without the film yeasts B6 and B7. The milk was at pH 6.4 or was adjusted to pH 4.9 to simulate the pH of the surface of a freshly dipped cheese. Counts of both micrococci and film yeasts were made with tryptone (0.5%), glucose (0.1%), beef extract (0.3%) agar and of film yeasts alone with agar made by adding 1.5% agar to the special Iya broth described in *Methods*.

The results in Table 1 show that the micrococcus by itself or in combination with film yeasts grew much better in skimmilk at pH 6.4 than at pH 4.9. The micrococcus alone made only a 1.7 fold increase in 56 hours in milk at pH 4.9, but a 6.9 fold increase in the presence of film yeasts. That the stimulating effect of the yeasts was not entirely due to destruction of lactic acid was indicated by the better growth of the micrococcus in milk at pH 6.4 in the presence of film yeasts than in their absence. The film yeasts seemed to obtain no benefit from growing with the micrococcus.

TABLE 1

Growth of micrococcus Mc 32 and film yeasts B6 and B7 separately and together

| | | | Number of organisms per ml. ($\times 1,000$) after incubation at 30° C. for | | | | | | | |
| | | | 0 hours | | 33 hours | | 56 hours | | | |
Flask[b]	Plating medium[a]	Initial pH of milk	(a) Micrococci	(b) Yeasts	(c) Micrococci	(d) Yeasts	(e) Micrococci	(f) Yeasts	Ratio e:a	Ratio f:b
A	TGE	6.4	5,400		27,000		87,000		16.1	
B	TGE	4.9	3,200		3,200		5,400		1.7	
C	Iya	6.4		110		500		1,700		15.5
D	Iya	4.9		120		340		930		7.8
E	TGE	6.4	1,850	50	18,770	230	70,560	440	38.1	8.8
E	Iya	6.4		50		230		440		8.8
F	TGE	4.9	1,500	100	3,330	170	10,400	600	6.9	6.0
F	Iya	4.9		100		170		600		6.0
G	TGE	6.4	0	0	0	0	0	0	–	–

[a] TGE = tryptone glucose beef-extract, which grows both micrococci and film yeast.
Iya = Iya's agar acidified to pH 4.1 with lactic acid, which grows only film yeasts.
[b] A and B inoculated with micrococcus Mc 32.
C and D inoculated with film yeasts, both B6 and B7.
E and F inoculated with micrococcus Mc 32 and film yeasts B6 and B7.
G uninoculated.

To demonstrate that the stimulating effect of the film yeasts on the growth of the micrococci was due to more than the reduction of the acidity by the film, four cultures of micrococci, representing the three species of *Micrococcus* found on cheese smears, were grown in skimmilk at room temperature with and without added autolysate of each of two film yeasts. The results in Table 2 indicate that the autolysates of both film yeasts were stimulatory to the growth of all four cultures of micrococci tested. The stimulation is attributed to available nitrogen compounds and accessory food substances in the autolysates of the yeasts.

Odors of cultures of smear organisms. Brick cheese that has developed a smear normally has a characteristic odor that has been described as sweaty, a term used also to describe the odor of some of the higher volatile fatty acids, especially isovaleric acid. An effort was made to determine whether the micrococci or film yeasts or combinations of them could be demonstrated to be responsible for the brick cheese odor. Pure cultures of five of the micrococci and the two

TABLE 2

Effect of two film yeast autolysates on the growth of micrococci in skimmilk at room temperature

| | Numbers of micrococci per ml. ($\times 1,000$) | | | | | |
| | Control[a] | | B6 autolysate[b] | | B7 autolysate[c] | |
Micrococcus	0 hr.	48 hr.	0 hr.	48 hr.	0 hr.	48 hr.
Mc 11	280	640,000	190	1,400,000	330	1,300,000
Mc 32	6,900	1,100,000	6,700	2,200,000	9,600	1,700,000
Mf 43	10,000	3,800,000	17,000	7,800,000	14,000	6,400,000
Mv 22	220	490,000	220	1,200,000	190	800,000

[a] Control = 5 ml. sterile water added to 50 ml. skimmilk.
[b] B6 autolysate = 5 ml. B6 autolysate added to 50 ml. skimmilk.
[c] B7 autolysate = 5 ml. B7 autolysate added to 50 ml. skimmilk.

film yeasts were grown for 2 weeks at room temperature in various media to test for the development of the sweaty odor characteristic of the cheese smear. The most commonly found micrococcus of the smear, *M. varians*, did not produce any sign of the odor when grown in skimmilk, whole milk, or Bacto-Casitone plus yeast extract. Cultures of *M. caseolyticus* and *M. freudenreichii* produced a sweaty odor in one or more of these media. The proteolytic and lipolytic film yeast (B6) produced a sweaty odor in skim and whole milk, whereas the nonproteolytic and nonlipolytic film yeast (B7) did not. *M. caseolyticus* did not cause the sweaty odor when grown in peptone or tryptone nutrient broth, with or without added butterfat. A combination of the two film yeasts and *M. caseolyticus* (Mc32) yielded a marked sweaty odor when grown in whole milk, an odor stronger than that produced by the micrococcus or film yeast alone. It was observed that the sweaty odor could be produced in either skimmilk or whole milk but usually more strongly in the latter.

An analysis by partition chromatography for volatile acids produced by the micrococci and yeasts in pure culture in Bacto-Casitone containing 0.01% Bacto-Yeast Extract showed that the yeast and micrococcus cultures differed in their ability to form butyric, propionic, and acetic acids in the medium. The strains of *M. caseolyticus* yielded considerably more butyric acid than the other organisms tested. Levels of acetic and propionic acid were comparatively low except in the instance of the proteolytic and lipolytic film yeast (B6). Higher volatile acids were not present in measurable amounts.

Because cultures of micrococci streaked on milk agar plates often gave off an odor resembling that of brick cheese, an attempt was made to identify some of their odoriferous compounds. About 20 plates of each culture were flooded with 1% hydrochloric acid, and the washings were pooled for extraction with ether and fractionation by chromatography. Three fractions were collected, two of which were identified, one as butyric and the other as acetic acid. The third fraction, which was composed of higher fatty acids than butyric, gave a residue, on evaporation of the solvent, the odor of which resembled that of the streak plate cultures. The odor was pungent and resembled that of butyl alcohol plus the odor of the fatty acids, valeric and caproic. It is evident, then, that the micrococci can produce odors like those of the smear on ripening brick cheese.

Flavor of surface-inoculated cheeses. The real test of the ability of the pure cultures isolated from brick cheese smears to affect the flavor of the cured cheese was a series of experiments in which the surfaces of cheeses were inoculated with one or more cultures by the procedures described under *Methods*. Preliminary experiments had shown that autoclaved cheese vat brine or 22% sodium chloride brine was not as satisfactory as pasteurized 22% brine for salting the cheese. Inoculation of the surface of cheeses with a pure culture of either film yeast (B6 or B7) did not bring the flavor of these cheeses up to that of the control cheeses, even though the yeasts grew well and some micrococci appeared after 7 to 9 days on the cheeses inoculated with film yeasts. The addition of the two film yeasts together seemed to cause some improvement in flavor, which, however, was not as good as that of the control cheeses.

Cheeses inoculated with *B. linens* alone and with *B. linens* and the two film yeasts did not develop the characteristic brick cheese flavor possessed by the control, although the flavor was mild and pleasant. Contrary to expectations, no Limburger flavor developed upon the addition of *B. linens*, but this organism did not grow on the surface according to microscopically obtained evidence.

Cheeses inoculated with each of two strains of *M. caseolyticus* but no film yeasts did not show any typical brick cheese flavor and usually were inferior to the control cheeses in flavor. Microscopic tests on the smear showed that the micrococci decreased in numbers throughout the curing period.

The foregoing experiments had demonstrated that film yeasts should precede the micrococci in order to get appreciable growth and activity of the latter. Therefore, a series of cheeses was inoculated with both film yeasts and with single cultures of representatives of the three species of *Micrococcus* previously identified. The results in Table 3 indicate that a brick cheese flavor was obtained more often with strains of *M. caseolyticus*, but that *M. freudenreichii* and *M. varians* could contribute this flavor.

TABLE 3

Effect upon the flavor of brick cheese of adding various micrococci plus a pair of film yeasts to the surface

Micrococcus added [a]	Flavor grade [b]	Total grade	Brick cheese flavor	Description of flavor
Mc 11	4.0	3.7	−	salty, acid, fermented
Mc 32	4.0	3.7	−	salty, sl. bitter, sl. acid, fermented
Control	4.0	3.7	−	salty, acid, fermented
Mc 11	3.0	3.3	−	salty, bitter, acid, yeasty, pleasant
Control	2.3	2.0	−	yeasty, pleasant
Mc 11	3.8	4.0	+	salty, sl. acid, yeasty, sl. fermented
Mc 32	3.2	3.3	+	salty, sl. acid, fermented, glutamic
Mc 58	3.8	3.8	++	salty, bitter, acid, glutamic
Mf 15	3.2	3.5	−	salty, sl. acid, yeasty, sl. fermented
Mf 43	3.2	3.3	+++	salty, sl. acid, sl. unclean, yeasty
Mv 22	3.8	3.8	+	salty, bitter, sl. acid, yeasty, fermented
Control	3.2	3.5	−	salty, acid, sl. unclean, fermented
Mc 11	2.7	2.8	++	salty, sl. acid, sl. yeasty, pleasant
Mc 32	2.6	2.6	+	salty, sl. bitter, sl. yeasty, acetic
Mc 58	2.8	3.8	−	salty, sl. acid, sl. yeasty, sl. unclean
Mf 15	3.0	3.1	−	salty, sl. acid, yeasty, acetic
Mf 43	2.9	2.9	−	salty, sl. acid, yeasty, sl. unclean, fruity
Mv 22	2.7	2.7	−	salty, sl. acid, yeasty, acetic
Control	3.6	3.7	−	salty, acid, yeasty, Limburger, acetic

[a] Cheese previously inoculated with a saline suspension containing both B6 and B7 film yeasts.
[b] See *Methods* for numerical grading.

The presence or absence of characteristic brick flavor in the cheese is of primary interest in the data shown. Flavor grade, total grade, and descriptions of flavor are included to indicate that other flavors might mask the brick flavor and that flavor and total grade did not necessarily parallel the intensity of the brick flavor. Microscopic examinations of the smears showed that *M. caseolyticus* and *M. freudenreichii* grew well in the smear during ripening but that *M. varians* did not increase in numbers until about the ninth day.

TABLE 4

Effect upon flavor of brick cheese of adding mixture of six micrococci and a pair of film yeasts together to the surface

	Flavor grade[a]	Total grade	Brick cheese flavor	Description of flavor
Test	4.0	4.0	+	salty, acid, fermented, sl. Limburger
Control	3.2	3.5	−	salty, acid, sl. unclean, yeasty
Test	2.8	3.8	+	salty, acid, yeasty, sl. fermented, sl. Cheddar
Control	3.6	3.7	−	salty, acid, yeasty, Limburger, acetic

[a] See *Methods* for numerical grading.

Since strains of all three species of *Micrococcus* could contribute characteristic flavor to brick cheese by growing in the surface smear, a combination of strains should be effective. A combination of six strains, representing the three species of *Micrococcus*, and the two film yeasts was inoculated onto the surfaces of two lots of cheese, which along with controls were cured as usual. The results in Table 4 show that the combinations of cultures produced brick cheese flavor in both test cheeses, whereas the control cheeses of this lot were devoid of the typical flavor.

DISCUSSION

It is evident from the results that micrococci are the chief organisms concerned in the production of the characteristic odor of the smear of brick cheese and of the flavor given the cheese by that smear and that previous growth of film yeast in the smear intensifies the flavor production. However, the most commonly encountered type of film yeast cannot cause the flavor by itself. The lipolytic and proteolytic yeast used in some of the experiments can contribute flavor, but this type of film yeast apparently is so rare in occurrence (found once in 136 isolations) as to be of little significance. The function of the film yeasts is primarily to improve growing conditions for the micrococci which follow.

The most numerous micrococci in smears, *M. varians*, do not seem to be as active in flavor production as the two other species ordinarily found growing along with it, *M. caseolyticus* and *M. freudenreichii*, but a combination of these micrococci is effective.

The flavoring substances formed by the micrococci of the smear have been shown to include butyric and acetic acids and sometimes propionic acid, but it is the higher volatile fatty acids that are responsible for the sweaty odor of the brick cheese smear, as in the surface taint of butter.

Hastening of development of the smear on cheese results when cultures of smear organisms are rubbed onto the ripening cheese. The selection of film yeasts and micrococci known to produce desirable flavors should help guarantee the prompt production of good flavor in brick cheese.

SUMMARY

Pure cultures of film yeasts and of micrococci were isolated from surface smears of brick cheese and from cheese brines. A culture of the most commonly found film yeast and one that was both lipolytic and proteolytic were selected

for experiments on associative growth and inoculation of cheese surfaces. The micrococci were found to be predominantly colorless variants of *M. varians*. Next in order of occurrence were *M. caseolyticus* and *M. freudenreichii*. The associative growth of film yeasts and micrococci was studied and attempts were made to ascertain the source and the cause of the typical odor of a brick cheese smear. Experimental cheeses were inoculated on the surface with film yeasts and micrococci, alone or in combinations, to find their effect on flavor.

Growth of micrococci with film yeasts indicated that the yeasts stimulate the growth of the cocci on the surface of a brick cheese by reducing the acidity of the cheese surface and furnishing accessory food substances to the cocci.

M. caseolyticus and *M. freudenreichii*, but not *M. varians*, were found to be able to produce the characteristic sweaty odor of a brick cheese smear when grown in milk and other media, as could the proteolytic and lipolytic film yeast, but not the film yeast most commonly found in smears.

Butyric and acetic acids were identified as products of the growth of cultures of micrococci on milk agar, but the fraction with an odor characteristic of brick cheese smear was one containing higher fatty acids.

Inoculation of cheese surfaces with single cultures or combinations of them gave the following results: For the most part the addition of the two film yeasts to the surface, alone or combined, or the separate addition of *B. linens* or of single cultures of micrococci did not improve the flavor of the brick cheese. The addition of *B. linens* plus the two film yeasts neither improved the flavor nor produced a Limburger flavor. Brick cheese flavor was detected in seven of 15 cheeses inoculated with separate strains of micrococci combined with the two film yeasts. The typical flavor was found most frequently in cheeses to which *M. caseolyticus* had been added, but one cheese inoculated with *M. freudenreichii* had a very pronounced brick cheese flavor. In both trials in which a combination of six micrococci, which included the three species, and the two film yeasts were added to the cheese, brick cheese flavor was produced.

It is concluded that micrococci, especially combinations of them, in the smear are important in the production of the characteristic brick cheese flavor, and that this flavor is accentuated by the previous growth of film yeasts.

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APPLICATION OF A LATIN SQUARE CHANGE-OVER DESIGN TO DAIRY CATTLE GRAZING EXPERIMENTS

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In feeding experiments with dairy cows it is reasonable to expect the results to vary not only according to the feeds given but also from other causes. One cause, the differences among cows, was recognized early, and various change-over experimental designs were devised to separate the source of variation due to cows from that due to feeds or treatments. Cochran *et al.* (1), in developing a Latin square change-over design for feeding trials with dairy cows, found not only cows but also experimental periods to be significant sources of variability.

It was anticipated that both cows and experimental periods would be sources of variation in the yields from a rotational grazing experiment. Pastures may vary in rates of growth through the grazing season and consequently offer different amounts of forage at successive grazing periods. Therefore, a Latin square change-over design was selected for this experiment.

EXPERIMENTAL PROCEDURE

Three pasture mixtures, alfalfa–orchard grass, Ladino clover–orchard grass, and Ladino clover–Kentucky 31 fescue, were planted in 2-acre lots. The mixtures were randomly assigned as to location within each 6-acre block or replicate. The four blocks or replicates were located adjacent to one another in a rectangular area 24 acres in size.

Trios of cows of similar ages and calving dates were selected, then randomly assigned to one or another of three experimental groups. Such groups constituted the "tester" cows. Various numbers of "spare" cows were added and removed as appeared necessary in attempts to adjust numbers of cows to quantities of forage available.

The grazing plan followed was based on a 3 × 3 Latin square change-over design. This design was essentially the same as that used in feeding experiments with dairy cows and in recent Georgia work with winter pastures (7). One major difference, however, was that each group of cows grazed successively on four replicates of the same pasture mixture before going to the second mixture and similarly grazed on four replicates of the second before going to the third. Another difference was that the grazing season did not end, except in one year, when one or more "squares" of the design were completed. The grazing plan for a single replicate consisted of short periods when all three cow groups were grazing, with somewhat longer periods for recovery while the cow groups grazed

the three other replicates. Grazing periods within a replicate were about 1 week in length, with intervals of about 3 weeks for recovery of pasture lots.

Cows were weighed at the beginning and end of each grazing period. Weight of each milking from each cow was recorded. Monthly butterfat tests were made on 1-day milk samples. Changes in body weights of cows were estimated as average linear changes over a number of weeks, rather than the somewhat erratic apparent changes over single grazing periods.

Data secured were used to estimate yields of FCM *(4)* and TDN *(6)* from the pastures. Fat-corrected milk for tester cows only was considered to be a suitable measure of pasture quality, whereas estimates of total digestible nutrients obtained by all cows (testers and spares) from the various pastures were assumed to measure quantity *(8)*. The use of only tester cows permitted comparisons of pasture quality over all lots and replicates which involved the same cows. The same use would have been desirable for comparisons of TDN, but the yields from some lots during some grazing periods were much greater than could have been utilized by the tester cows alone.

ANALYSIS OF RESULTS

The estimated TDN values from the 1951 grazing season (Table 1) will be used to illustrate the method of analysis. Five grazing periods, i.e., one and two-thirds squares of the grazing plan, were completed by the end of the grazing season. The A_i represents cow groups; the B_j, pasture mixtures; and the C_k, grazing periods.

Since the data were not orthogonal with respect to the A_i and B_j, it was necessary to fit constants and obtain the corresponding deviations *(a_i and b_j)* from the block means in order to calculate unbiased sums of squares for cow groups and pasture mixtures *(2, 5)*. The solution for a_1 in Block 1 is shown below.

The equations for Block 1 can be determined from inspection of Table 1. They are

$$
\begin{aligned}
5M + 5a_1 &+ b_1 + 2b_2 + 2b_3 = 4{,}673 \\
5M + 5a_2 &+ 2b_1 + 2b_2 + b_3 = 4{,}884 \\
5M + 5a_3 &+ 2b_1 + b_2 + 2b_3 = 4{,}469 \\
5M + a_1 + 2a_2 + 2a_3 &+ 5b_1 = 3{,}846 \\
5M + 2a_1 + 2a_2 + a_3 &+ 5b_2 = 5{,}186 \\
5M + 2a_1 + a_2 + 2a_3 &+ 5b_3 = 4{,}994
\end{aligned}
$$

Only the first and fourth equations are needed to solve for b_1, since the coefficients of b_1 can be eliminated from the second and third equations by applying the restriction $\Sigma a_i = \Sigma b_j = 0$ and doing a little algebraic manipulation.

The initial equations for solving for b_1 are

$$
\begin{aligned}
5M + a_1 + 2a_2 + 2a_3 + 5b_1 &= 3{,}846 \\
5M + 5a_1 + b_1 + 2b_2 + 2b_3 &= 4{,}673
\end{aligned}
$$

Applying the restriction and rearranging,

$$
\begin{aligned}
a_2 + a_3 + 5b_1 &= 3{,}846 - 4{,}675 \\
5a_1 + b_2 + b_3 &= 4{,}673 - 4{,}675
\end{aligned}
$$

TABLE 1
Estimated yields of TDN (in lb.) per 2-acre lot

Block	Grazing	B_1[a]		B_2		B_3		Sums
	1st C_1	A_1[b]	349	A_3	666	A_2	1,043	2,058
	2nd C_2	A_2	1,011	A_1	1,822	A_3	1,442	4,275
1	3rd C_3	A_3	834	A_2	1,260	A_1	1,088	3,182
	4th C_4	A_2	962	A_1	830	A_3	837	2,629
	5th C_5	A_3	690	A_2	608	A_1	584	1,882
	Sums		3,846		5,186		4,994	14,026
	Group sums	1—4,673		2—4,884	3—4,469			M = 935
	1st C_1	A_1	784	A_3	1,849	A_2	1,665	4,298
	2nd C_2	A_2	858	A_1	1,076	A_3	1,012	2,946
2	3rd C_3	A_3	950	A_2	1,169	A_1	817	2,936
	4th C_4	A_2	628	A_1	672	A_3	674	1,974
	5th C_5	A_3	276	A_2	254	A_1	244	774
	Sums		3,496		5,020		4,412	12,928
	Group sums	1—3,593		2—4,574	3—4,761			M = 862
	1st C_1	A_1	624	A_3	1,337	A_2	1,726	3,687
	2nd C_2	A_2	1,541	A_1	1,066	A_3	873	3,480
3	3rd C_3	A_3	687	A_2	1,248	A_1	841	2,776
	4th C_4	A_2	874	A_1	743	A_3	791	2,408
	5th C_5	A_3	281	A_2	240	A_1	251	772
	Sums		4,007		4,634		4,482	13,123
	Group sums	1—3,525		2—5,629	3—3,969			M = 875
	1st C_1	A_1	332	A_3	812	A_2	1,030	2,174
	2nd C_2	A_2	1,802	A_1	1,216	A_3	1,234	4,252
4	3rd C_3	A_3	773	A_2	1,012	A_1	1,049	2,834
	4th C_4	A_2	905	A_1	715	A_3	602	2,222
	5th C_5	A_3	481	A_2	415	A_1	430	1,326
	Sums		4,293		4,170		4,345	12,808
	Group sums	1—3,742		2—5,164	3—3,902			M = 854

[a] B_1 was alfalfa–orchard grass, B_2 Ladino clover–orchard grass, B_3 Ladino clover–Kentucky 31 fescue.
[b] A_1, A_2, and A_3 represent cow groups.

Further simplifying, and solving

$$-a_1 + 5b_1 = -829$$
$$5a_1 - b_1 = -2$$

$$24a_1 = -839$$
$$a_1 = -35.0$$

Similar solutions were made for the other constants, and the following values for Block 1 were obtained:

$$a_2 = 56.8$$
$$a_3 = -21.6$$
$$b_1 = -172.8$$
$$b_2 = 97.9$$
$$b_3 = 75.2$$

Inasmuch as grazings were orthogonal with respect to other variables, no fitting of constants for grazings was needed, and the sum of squares was calcu-

lated in the usual manner. The calculation of this and the other sums of squares are shown below for Block 1.

Correction factor: $(14,026)^2/15 = 13,115,245$ $= C_1$
Total: $(349)^2 + \ldots\ldots + (584)^2 - C_1$ $= 1,927,763$
Grazings: $[(2,058)^2 + \ldots + (1,882)^2]/3 - C_1$ $= 1,247,981$
Groups (unadjusted): $[(4,673)^2 + (4,884)^2 + (4,469)^2]/5 - C_1$ $=\quad 17,224$
Groups (adjusted): $-35.0(4,673) + 56.8(4,884) - 21.6(4,469)$ $=\quad 17,326$
Mixtures (adjusted): $-172.8(3,846) + 75.2(5,186) + 97.9(4,994)$ $=\quad 214,311$
Remainder (error): $1,927,763 - 1,247,981 - 17,224 - 214,311$ $=\quad 448,247$

Note that an unadjusted sum of squares for groups was calculated separately, and used in obtaining the error term (5). Data from Blocks 2, 3, and 4 were handled in the same manner as shown above.

For summary purposes and for use in the combined analysis, the block totals for pasture mixtures were adjusted for effects of cow groups (Table 2). For example, the B_1 mixture in Block 1 was grazed only once by Group 1 and two times each by Groups 2 and 3. Therefore, the constant for Group 1 was added algebraically to 3,846, and the adjusted value became 3,811.

The combined analysis for all four blocks or replicates required several additional calculations, including a new correction factor for all blocks, a total sum of squares over all blocks, a sum of squares for blocks, and a block-by-mixture interaction sum of squares. Sums of squares for cow groups and for grazings were obtained by pooling the respective individual block values. The four unadjusted sums of squares for groups that were obtained in the individual block analyses were pooled and used in obtaining the error term for the combined analysis. In calculating the blocks-by-mixtures interaction, use was made of the adjusted values for mixtures (Table 2). The combined analysis for all four blocks is shown in Table 3.

The pooled error mean square (54,854) was used in testing blocks and mixtures. The residual mean square (59,545) was used to test grazings, groups, and the interaction term. The highly significant differences among grazings within blocks agrees rather well with the variation among grazing totals in Table 1.

TABLE 2
Estimated yields of TDN (in lb.) adjusted for group effect

Block	Pasture mixture			Sums
	B_1	B_2	B_3	
1	3,811	5,164	5,051	14,026
2	3,313	5,144	4,472	12,929
3	3,815	4,560	4,748	13,123
4	4,184	4,089	4,535	12,808
Sums	15,123	18,957	18,806	52,886
Means per acre	1,890	2,370	2,351	2,204
Ranking of means	2 [a]	1 [b]	1 [b]	

[a] Significantly less than B_2 or B_3 ($P < 0.01$).
[b] B_2 and B_3 not significantly different.

TABLE 3
Combined analysis of variance for all four blocks

Source	D.F.	Sum of squares	Mean square	F-ratio
Total	59	9,979,483		
Blocks	3	60,937	20,312	0.37
Grazing within blocks	16	6,890,389	430,649	7.23 [b]
Groups within blocks	8	1,006,924	125,866	2.11
Mixtures	2	473,211	236,606	4.31 [a]
Mixtures × blocks	6 ⎱30	216,540 ⎱1,645,626	36,090 ⎱54,854	1.64
Error term	24 ⎰	1,429,086 ⎰	59,545 ⎰	

[a] Significantly different at 5% probability level.
[b] Significantly different at 1% probability level.

The nonsignificance of differences among cow groups was not anticipated, since cow differences often are significant in barn feeding trials. The significance of differences among pasture mixtures needed qualification. Upon examining the means according to a test proposed by Duncan (3), it was found that B_2 and B_3 did not differ significantly from each other whereas B_1 was lower (Table 2).

Estimated yields per acre of TDN for 1950 were 1,964 lb. for alfalfa–orchard grass, 2,185 lb. for Ladino clover–orchard grass and 2,289 lb. for Ladino clover–Kentucky 31 fescue. These yields, in contrast with the 1951 values, were not significantly different.

DISCUSSION

A major advantage of the Latin square change-over design for grazing is that it can lead to more efficient evaluation of treatment differences than would be possible with certain other designs. Relative efficiency of a design may be calculated as the inverse of relative size of error mean square in the analysis of variance. The error mean square in Table 3 would have been increased from 54,854 to 72,069 with the cow group classification omitted, to 201,506 with grazings omitted and to 189,156 with both left out of the design. Relative efficiencies would have been 100%, 78%, 27%, and 30%, respectively. The practical implication of such omissions is that treatment effects would have been evaluated improperly, since known sources of variability (cow groups and grazing periods) would have been included in the error term.

Disadvantages include (a) inability to end the grazing season when exactly one or more "squares" of the design have been completed and (b) tendencies toward loss of orthogonality and confounding. The former increases work in analysis of the data; the latter may reduce true efficiency of the experiment by partially confounding pasture mixture effects with those due to cow groups, grazing periods, and replicates. Confounding may result from (a) adding or removing spare cows to or from groups and (b) changes in quality and quantity of forage available from month to month.

The irregular addition and removal of spare cows may give biased comparisons of pasture mixtures, since apparent differences among the mixtures may be due not only to their yielding abilities but also in part to the cows which graze them. The changes in quality and quantity of forage with time may affect estimates of replicate yields, since the replicates are grazed successively (rather than

simultaneously) and may be in different stages of growth at each grazing. For instance, a few spare cows in early stages of lactation might be used on Mixtures B_1 and B_2 but not on B_3. The resulting yields of FCM from B_1 and B_2 might then be greater than from B_3, not only because of differences in nutritive values of the pastures but also partly because of the cows which grazed them. Similarly, if Block 2 is grazed one week later than Block 1, the yield from Block 2 will differ somewhat from that of Block 1 because of its later stage of growth.

There is need for additional work on designs for use in dairy grazing experiments. Such designs should (a) be flexible enough to utilize all forage available at all times, (b) minimize the confounding of variables with one another and permit them to be estimated most efficiently, and (c) permit highly efficient testing of significance among treatment differences.

SUMMARY

A Latin square change-over design was applied to dairy cattle grazing experiments. Experimental procedure is given. Analysis of the data is presented, including the fitting of constants for nonorthogonal variables. The design gave substantial gains in efficiency over single grouping or complete randomization but required more work to analyze the data and may have led to some confounding of effects of pasture mixtures with effects of other variables.

ACKNOWLEDGMENTS

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APPLICATION OF AN INFLATABLE URETHRAL CATHETER FOR URINE COLLECTION FROM COWS[1]

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Many techniques have been employed in an attempt to obtain urine quantitatively from cows. Manual collection *(4)* has largely been outmoded by various separating devices. These may consist of an apron to which is attached a short tube that is inserted into the vagina *(2)* or an apron with a wire frame which can be bent to fit the shape of the pin bones *(1)*. Branding cement has also been employed to hold separating equipment in place *(5)*. Urine may be drawn off through a large tube and the feces collected in a bag or metal box. These types of apparatus require frequent adjustment and do not avoid contamination of urine with mucus or other secretions of the vaginal tract. Bassett *(3)* has reported the use of a Jaques rubber urethral catheter for urine collection from ewes on pasture. The catheter is strapped into place with harness and is connected to a plastic container fitted snugly under the belly of the ewe. A larger diameter catheter is used when the urethral orifice of the ewe becomes enlarged through catheterization or pregnancy, but even with this precaution Bassett states that some ewes will strain and cause urine to escape around the periphery of the catheter.

A similar catheter tested in this laboratory was found to be unsuitable for quantitative urine collection from cows because it could be readily expelled and urine was lost around the outside of the tube. The inflatable urethral catheter described in the present report eliminates loss of urine, and no harness is required to keep the apparatus in place.

METHODS AND MATERIALS

The initial experiments were conducted with inflatable catheters constructed in the laboratory from rubber tubing, balloons, and rubber cement. These were used for several months but were discarded in favor of similar catheters (Figure 1) of more durable material and smoother design which are used in the practice of medicine. These can be purchased from medical supply houses.[2]

A size 24, Bardex hemostatic catheter with a 75-ml. balloon (Bard catalogue No. 113) was found to be satisfactory for mature cows, and a smaller catheter (size 20, No. 123A) was more easily inserted through the urethral orifice of young dairy heifers.

The urethral orifice of the cow lies on the floor of the vagina about 3 to 4 in. from the entrance to the vulva. It should not be confused with a small blind sac, the suburethral diverticulum, which is located immediately posterior to it. A clean, sterile catheter is lubricated with a mild disinfectant jelly and introduced through the urethral orifice until the balloon lies just inside the neck of the
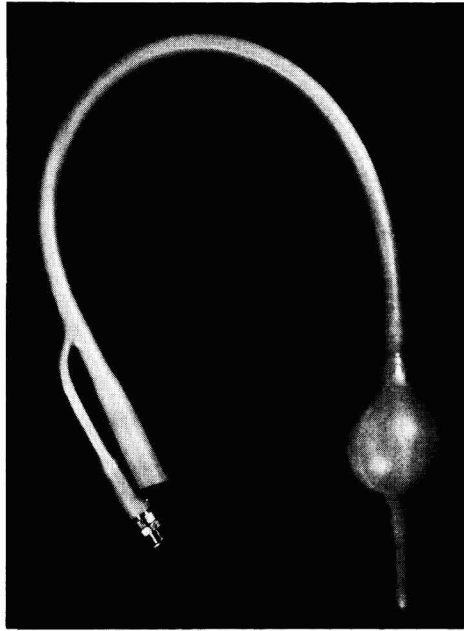
FIG. 1. A size 24, 75-ml. rubber inflatable urethral catheter. Note the valve in the inflation tube.

bladder. The balloon is inflated with 50 to 75 ml. of warm water by means of a hypodermic syringe and the inflation tube is sealed with a clamp or with a special valve[3] which is made to fit the nozzle of the syringe. The catheter is then pulled gently so that the bulb fits snugly into the neck of the bladder and prevents any loss of urine around the tube. With a little practice an operator can insert and inflate the catheter within 1 or 2 minutes.

A 3-in. length of $\frac{1}{4}$-in. diameter stainless steel tubing is used as a connection between the catheter and a 3/16-in. bore rubber tube, which drains the urine into a carboy (Figure 2). The tube is tied to the carboy with sufficient slack to allow for the movements of the cow. Occasionally a cow switches its tail excessively against the collection tube. The resultant jerks on the catheter can be prevented by tying the tube to a 4-in. length of 2-in. canvas strap attached to the cow with branding cement a few inches below and to one side of the vulva.

RESULTS AND DISCUSSION

The Foley catheter was employed with 14 cows and heifers during the past 2 years for the collection of urine in digestibility and hormone studies. It was inserted for periods up to 3 weeks without any apparent adverse effects. Young heifers and some cows may show slight discomfort when first catheterized but they soon become accustomed to the apparatus and seldom show any further annoyance. Four cows producing 10 to 30 lb. of milk per day showed no drop in production when catheterized for periods of 1 to 3 weeks.

[3] Bard-apter valve, catalogue No. 480, C. R. Bard, Inc., Summit, N. J.
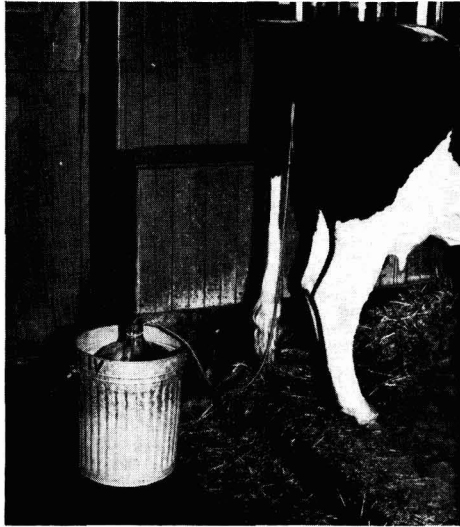
FIG. 2. Collecting urine for hormone work from a cow in a stanchion. A sufficient length of tubing is allowed to enable the cow to move from side to side and to lie down.

The equipment was also tested under loose-housing conditions in preparation for digestibility studies on pasture. The feces of two 7-month-old heifers were collected in canvas bags, and the urine was drawn off with inflatable catheters. The catheters were left in place for 3 weeks without producing any undue strain upon the animals. At no time were any clinical signs of infection observed in fresh samples of urine from animals that had been catheterized for periods of 1 week or more.

SUMMARY

An inflatable rubber urethral catheter has been employed for the continuous collection of urine from dairy cows and heifers. Urine has been collected by this method for periods of up to 3 weeks without causing infection or imposing any undue strain upon the animal.

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THE EFFECT OF FREQUENCY OF EJACULATION ON THE SEMEN PRODUCTION, SEMINAL CHARACTERISTICS, AND LIBIDO OF BULLS DURING THE FIRST POST-PUBERAL YEAR[1]

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With the rapid expansion of artificial insemination to the point where thousands of cows are bred to a single sire in a year, it becomes imperative to prove and select sires as early as possible in order to maximize the use of those proven desirable. The usual recommendations are that bulls should not be placed in service at less than 1 year of age and that they be used for not over 25 services during the first year of service. Following these practices, it is possible to prove a bull by the time he is 5 years old, though seldom do sufficient progeny complete lactations to make the proof reliable before the bull is 6 years old. Sperm have been found in the testes of the bull as early as the 224th day *(13)*. This finding suggests the possibility of placing bulls in service earlier than the usual recommendation.

The limits of usage consistent with satisfactory fertility in bulls have been studied through surveys of breeding results of bulls being used at various intervals, and by planned experiments. In studies of the first type, Dawson *(4)* found fertility to be related to the number of services in the preceding month, and Lasley and Bogart *(9)* found positive highly significant correlations of fertility with the interval from the previous service. In contrast to these reports, Ellenberger and Lohmann *(5)* found neither the interval from the last ejaculation nor the number of ejaculations in the previous 30 or 60 days to be significantly correlated with fertility of bulls ejaculated at various intervals. In planned experiments, Kirillov *(6)* and Kirillov and Morozov *(8)* found that bulls mated on alternate days produced more semen and more sperm per ejaculation, but less for the total period than those on two or four matings per day. No significant differences in the percentage of ejaculates considered suitable for use in artificial insemination or the percentage of cows not returning to service within 30-60 days were found by Patrick *et al. (12)* when bulls were ejaculated once every 4th day, twice every 8th day, or three times every 12th day. Mercier *et al.* *(11)* found a higher percentage of ejaculates good enough for use in artificial insemination when bulls were ejaculated once every 6th day as compared with twice every 12th day or three times every 18th day. Most of the work on frequency of ejaculation in the bull has been done with mature bulls. Roberts *(14)* studied the problem with bulls 12-15 months old, obtaining the highest numbers of sperm per ejaculation on two ejaculations per week. However, Roberts' experiments were of short duration.

Except for preliminary reports of these investigations *(1, 2)* there appears to be no record of studies on the effect of frequency of ejaculation on the sperm production, seminal characteristics, and libido of bulls ejaculated at regular intervals from puberty. It is the purpose of this paper to present the results of experiments designed to determine the age at which bulls become sexually functional and the effect of frequency of ejaculation on semen production, seminal characteristics, and libido of early post-puberal bulls and the effects on later semen production of the same bulls.

EXPERIMENTAL PROCEDURE

Nine Holstein bull calves were divided at random into three groups of three each to be assigned to collection frequencies of one, two, and three times per week. The bulls were tested at weekly intervals, each bull being allowed 10 minutes to express sexual interest by mounting a heifer. As soon as a bull would mount and ejaculate, he was placed upon the preassigned collection schedule and kept on that schedule for 1 year. One false mount was allowed before each semen collection. If a bull had not mounted within 10 minutes after approaching within 5 ft. of the teaser, or had not ejaculated within 10 minutes after first mounting, a failure was recorded for that collection period. One teaser, a freemartin heifer, was used throughout most of the experiment.

All semen samples were examined for volume, per cent motile sperm, sperm concentration, and pH. Counts of morphologically atypical sperm were made on approximately 50 samples from each collection frequency group.

In the analyses of variance, the frequency mean squares were tested against the bulls within frequency mean squares. The period mean squares were tested against the period times frequency interaction mean squares. The bulls within frequency mean squares and the period times frequency interaction mean squares were tested against the period times bulls within frequency interaction mean squares.

RESULTS

The bulls showed overt expression of sexual interest at a mean age of 29.4 $\pm$ 2.14 weeks. They first ejaculated at a mean age of 38.9 $\pm$ 1.30 weeks. The mean values of the seminal characteristics are summarized by quarter-year periods in Table 1. This table shows that semen quality was lowest during the first period and improved in most respects during the first three-quarters of the year. The differences shown between quarters during the year were highly significant for all semen characteristics measured except pH and total sperm per ejaculate. The differences in pH were significant.

In Table 2 the seminal characteristics are tabulated according to frequency of ejaculation. It will be noted that there was little difference in semen volume at the various frequencies of collection. The largest ejaculates were produced by bulls collected once per week but the differences were not significant. Although highly significant differences in per cent motile sperm, sperm concentration, total sperm per ejaculate, and total motile sperm per ejaculate were found between bulls within frequencies, the group differences were not significant for these

F. N. BAKER ET AL

TABLE 1

Summary of seminal characteristics by quarter-year periods

Characteristic units	Quarter-year period			
	1	2	3	4
	Mean S. E.	Mean S. E.	Mean S. E.	Mean S. E.
Semen volume/ejaculate *(ml.)*	1.84 ± .075	2.57 ± .083	3.12 ± .092	3.09 ± .074
Motile sperm *(%)*	34 ± 1.5	52 ± 1.0	52 ± 0.6	52 ± 1.0
Semen pH	6.87 ± .023	6.73 ± .026	6.71 ± .069	6.81 ± .022
Sperm concentration *(10⁶/ml)*	303 ± 22	668 ± 36	936 ± 39	868 ± 38
Total sperm/ejaculation[a] *($\times 10^6$)*	708 ± 71	1836 ± 128	3191 ± 184	3028 ± 174
Total motile sperm/ejaculation *($\times 10^6$)*	338 ± 39	1035 ± 78	1723 ± 119	1639 ± 121
Atypical sperm *(%)*	12.3 ± 1.09	9.5 ± .81	7.4 ± .80	5.8 ± .59

[a] The mean total sperm per ejaculate cannot be calculated from the mean volume and mean sperm concentration since the latter two are arithmetical means and the former is a weighted mean.

characteristics. Highly significant interactions between period and frequency were found for sperm concentrations and total sperm per ejaculate. The two-time group produced the lowest average semen volumes and sperm numbers. As a result of the random assignment of bulls to collection frequency groups, three bulls with a lower than average semen producing ability were assigned to this group. The variation in semen volume and sperm numbers is thought to be due to this, rather than to collection frequency.

Figure 1 shows the total sperm produced during the 1-year experimental period. The one, two, and three times per week collection groups produced a total of 400, 418, and 935 billion sperm, respectively. All bulls produced in successive quarter-year periods 150, 361, 603, and 639 billion sperm.

Table 3 gives the number of trials and the number of failures to mount and failures to ejaculate after mounting. Total refusals for the one- and two-time groups were two and one out of 156 and 312 trials, respectively, whereas the three-time group refused 83 times in 468 trials. The proportions of failures to mount and of failures to ejaculate of the three-time group were highly significantly different from those of the one- and two-time groups.

DISCUSSION

The puberty data suggest a transition period of approximately 10 weeks from the infantile to the sexually functional bull. They further indicate that puberty

TABLE 2

Summary of seminal characteristics by frequencies of ejaculation

Characteristic units	Frequency of ejaculation per week		
	1×	2×	3×
	Mean S. E.	Mean S. E.	Mean S. E.
Semen volume/ejaculate *(ml.)*	3.12 ± .105	2.50 ± .065	2.57 ± .069
Motile sperm *(%)*	49 ± 1.3	44 ± 1.1	49 ± 0.9
Semen pH	6.77 ± .024	6.89 ± .017	6.69 ± .041
Sperm concentration *(10⁶/ml)*	763 ± 47	462 ± 23	845 ± 30
Total sperm/ejaculation *($\times 10^6$)*	2705 ± 214	1412 ± 100	2561 ± 125
Total motile sperm/ejaculation *($\times 10^6$)*	1488 ± 97	736 ± 56	1394 ± 86
Atypical sperm *(%)*	6.7 ± .61	12.9 ± .87	6.1 ± .53
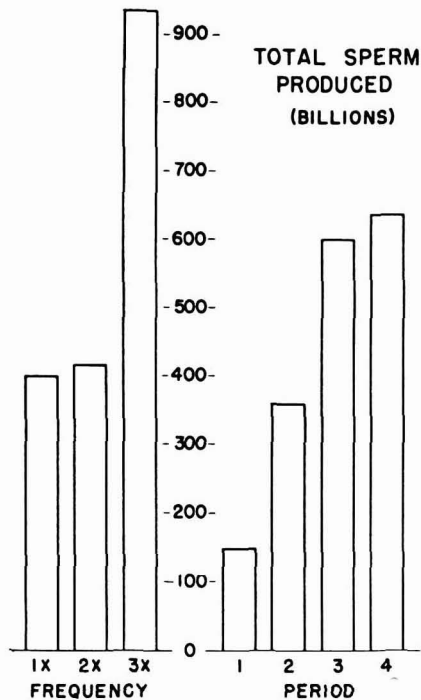
FIG. 1. Total sperm produced in 1 year by bulls on collection frequencies of one, two, and three times per week and total sperm produced by all bulls by quarter-year periods following puberty (3 bulls in each frequency group, 9 bulls in each quarter-year total).

is reached earlier than the commonly accepted serviceable age (over 1 year). The data show progressive increases in semen volume, sperm concentration, total sperm per ejaculation, and total motile sperm per ejaculation through at least three-quarters of a year after puberty. The apparent leveling off during the fourth quarter-year may have been due to seasonal effects, since the bulls showed further increases in these characteristics in a subsequent experiment. These increases are attributed to the growth of germinal epithelium and accessory gland secretory tissue associated with increasing age. As has been noted above, semen volume, sperm concentration, and sperm motility were lowest during the first

TABLE 3
Summary of failures to mount and failures to ejaculate

Weekly collection frequency	Total trials (3 bulls/group)	Refusals		
		To mount	To ejaculate	Total refusals
1×	156	2	0	2
2×	312	1	0	1
3×	468	52	31	83

quarter-year. Experiments are needed to test the fertility of the early post-puberal bull.

Semen volume per ejaculation was highest for the bulls ejaculating least frequently (once per week) and lowest for those ejaculating most frequently (three times per week). These findings are consistent with the reports of Kirillov (6), Kirillov and Morozov (7), and Roberts (14). The failure to find significant differences in sperm motility asociated with different frequencies of ejaculation is in agreement with Patrick et al. (12). The finding of the highest sperm concentration in the group collected most frequently (three times per week) is in contrast with the reports of Kirillov, Kirillov and Morozov, and Roberts. No trends relative to frequency of ejaculation were apparent in total sperm per ejaculation or total motile sperm per ejaculation. The finding that the most frequently collected group (three times per week) produced the most sperm during the experimental period is in agreement with the data of Chang (3) for rams and Roberts (14) for bulls. This finding may be indicative of a failure in the one- and two-time groups to ejaculate a high proportion of the sperm actually formed. However, it is not to be inferred that three times per week is an optimal frequency of ejaculation for early post-puberal bulls or that the sperm obtained from three ejaculations per week is an accurate measure of the rate of spermatogenesis.

Libido, as measured by the proportions of failures to mount and failures to ejaculate, was the only characteristic studied in which clear-cut differences were noted between frequencies. The bulls scheduled to ejaculate three times per week actually ejaculated only two and one-half times, on the average, whereas the other bulls failed less than 1% of the time. The reduction in libido with frequent ejaculation is in agreement with the findings of Rodolfo (15) and McKenzie et al. (10) in swine and Kirillov (6) in bulls. However, it should be noted that one bull in the three-time group was responsible for only three of 83 failures to ejaculate. This bull has ejaculated three times per week (11 failures to ejaculate excepted) for 2½ years after puberty. During this time there has been a gradual increase in the amount and quality of semen, which would be expected with growth. Recently, this bull was proven fertile. In contrast, another bull from the three-time group was maintained on the same schedule for over 2 years, failing frequently (one run of failures extending to 15 consecutive collection periods). Thus, it is seen that some bulls are better able to withstand the effects of frequent ejaculation upon libido than others.

The results of these investigations suggest that bulls might be placed in service younger and used more frequently than heretofore recommended. Service up to three times per week is apparently not harmful to sperm production except as the latter may be affected by reduced libido. However, the fertility of early post-puberal bulls should be tested before they are put to extensive use.

SUMMARY

In a study using nine Holstein bulls, sexual interest was first expressed at an average age of 29.4 weeks. Ability to ejaculate was first demonstrated at an

average of 38.9 weeks. Approximately 10 weeks was required for the transition from the infantile to the sexually functional bull. Highly significant differences were found in semen volume, per cent motile sperm, sperm concentration, total sperm per ejaculation and total motile sperm per ejaculation as the bulls aged during a 1-year period after puberty.

No significant differences in semen volume, sperm concentration, per cent motile sperm, semen pH, total sperm per ejaculation, total motile sperm per ejaculation, or per cent atypical sperm were found attributable to frequency of ejaculation when frequencies of once, twice, and three times per week were compared. A highly significant reduction of libido, as measured by the proportion of failures to mount and ejaculate, was found in bulls scheduled to ejaculate three times per week. Ejaculation frequencies up to three times per week were not harmful to the seminal characteristics nor to spermatozoan production of young bulls except as they adversely affected libido.

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THE EFFECT OF VARIOUS RESTRICTED DIETS ON THE GROWTH AND ON CERTAIN BLOOD COMPONENTS OF YOUNG DAIRY CALVES[1,2]

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The recent trend toward increased popularity of whole milk replacements for young calves has focused attention on the need for more information on the interrelationships among various dietary components and the resulting physiological responses. Therefore, further work on the more fundamental aspects of the nutrition of the young calf seemed warranted. The objective of the present work was to determine the effects of restricted diets containing various types and amounts of fats and other constituents upon certain physiological responses of the young dairy calf. The criteria employed were body weights; incidence of diarrhea; vitamin A, carotenoids, fat, calcium, and inorganic phosphate levels of blood plasma; and hemoglobin and reducing sugar levels of the blood.

EXPERIMENTAL PROCEDURE

A preliminary 8-week feeding trial with 35 calves was used to determine ration formulas for subsequent use. This work resulted in the selection of six diets containing various combinations of nonfat dry milk solids, dried whey product, hydrogenated soybean oil, butter oil and water (Table 1). Whole milk containing approximately 3% fat was employed as a control diet. All reconstituted diets were homogenized at approximately 3,000 lb. pressure. The butter oil was decanted from melted butter, and the hydrogenated soybean oil likewise was melted prior to incorporation in the rations. The water used was warmed sufficiently so that the final product was near body temperature when fed. A

TABLE 1
Composition of diets

		Components				
Diet	Water	Nonfat dry milk solids	Dried whey product	Hydrogenated soybean oil	Butter oil	Whole milk (3% fat)
		(lb. per 100 lb. body wt. daily)				
1	8.7	1.0		0.3		
2	8.7	0.5	0.5	0.3		
3	8.2	1.8				
4	8.2	1.3	0.5			
5	8.7	1.0			0.3	
6	8.7	0.5	0.5		0.3	
7						10.0

total solids level of 18% was selected for the low-fat rations so that all diets would be approximately comparable in energy content.

Calves from the Iowa State College dairy herd were allotted to seven groups, each of which contained one Ayrshire, one Brown Swiss, and five Holsteins. For the first 3 days after birth, all calves remained with their respective dams. On the afternoon of the third day they were removed to individual pens and the following morning were fed whole milk containing 3% fat. On the afternoon of the fourth day, samples of venous blood were obtained and body weights were recorded prior to the first feeding of the respective experimental diets. Subsequently, blood samples were obtained and weights were recorded at weekly intervals. Calves were fed twice daily, by nipple pail, at the rate of 10 lb. daily per 100 lb. body weight. This amount was reduced only when part was refused by the calves or in the event of severe diarrhea. Vitamin A, 12,000 I.U. per 100 lb. body weight, was administered once daily by gelatin capsule. No additional supplements were offered. Feed consumption, appearance of calves, and incidence of diarrhea were recorded twice daily.

The butter was obtained in lots of about 20 lb. each, and the vitamin A and carotenoid contents of the butter oil were determined for the first and last portions of each supply used. The procedure involved saponification, subsequent extraction (5) of carotenoids and vitamin A, and colorimetric estimation with activated glycerol dichlorohydrin (2). At frequent intervals fresh samples of the whole milk were assayed for vitamin A and carotenoids by a modification of the method of Sobel and Rosenberg (24). Composite samples of milk were preserved with mercuric chloride, and the fat percentage was determined by the Babcock test.

At weekly intervals whole blood (sodium citrate anticoagulant) was analyzed for reducing sugars by the Somogyi method (25) and for hemoglobin by the acid hematin method (10). Plasma from the citrated blood was analyzed for vitamin A and carotenoids with activated glycerol dichlorohydrin (3), for inorganic phosphate by the Fiske and SubbaRow method (12), for fat by the method of Allen (1), and for calcium by the method of Clark and Collip (9) by using Wang's wash solution (27).

<center>RESULTS AND DISCUSSION</center>

Plasma fat. The group mean plasma fat values of calves fed the reconstituted milks exhibited marked initial decreases as contrasted to an increase in the group fed whole milk (Figure 1). The blood plasma fat of calves which were fed reconstituted rations containing added fat increased during the second week of the experimental period, and in the butter oil groups this trend continued throughout the experiment. The increase did not continue so long in the hydrogenated soybean oil groups, and the levels were markedly lower in these calves than in subjects which were fed milk fat. The low-fat rations resulted in an initial diminution in blood plasma fat concentration. This decrease was less marked after the second week, however, and the values were essentially constant subsequent to the fifth week.
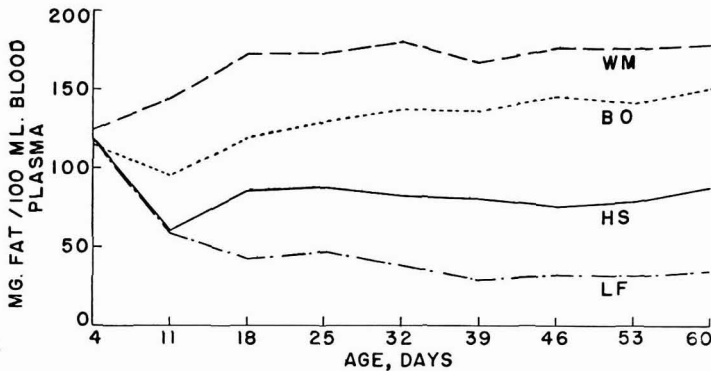
FIG. 1. Effect of type of dietary fat on blood plasma fat levels of dairy calves (WM = whole milk, BO = butter oil, HS = hydrogenated soybean oil, LF = low fat).

Analyses of variance (23) of the data for the Holstein calves indicated that the blood plasma fat levels in the animals fed whole milk were significantly higher than those for calves on the other rations. Moreover, significant differences also were observed among the groups not fed whole milk. Substitution of dried whey product for a portion of the nonfat dry milk solids in the reconstituted milks produced no significant effect upon blood plasma levels of fat. Therefore, data for diets similar in fat content were combined (Figure 1).

The present data offer no explanation for the initial decreases in plasma fat of calves fed reconstituted milks. This observation, however, is in agreement with the results of previous investigations (4, 22). Since blood plasma fat decreased initially in calves fed diet 5 as contrasted to an increase in the plasma fat levels of calves that received whole milk, and since the major components of the diets were similar, the difference may be due to dissimilarities in the physical nature of the milks. All diets except whole milk (No. 7) were reconstituted and homogenized. Moreover, since the phospholipid content of butter oil is much lower than that of whole milk fat, variation in this component might contribute to the observed phenomenon (16).

Higher concentrations of fat in blood plasma of calves which were fed diets containing butter oil than in those fed similar diets containing hydrogenated soybean oil may be associated with absorption and/or metabolism. The simplest explanation appears to be that the difference may be due to less efficient absorption of hydrogenated soybean oil. Limited studies with four calves indicated apparent digestibilities of hydrogenated soybean oil ranging from 83 to 99% and averaging 92.5%. Since hydrogenated soybean oil thus apparently is highly digestible, the low blood fat levels resulting from its dietary use may indicate a more rapid utilization or storage than occurs with butter oil. The oils differ considerably in fatty acid components and in phospholipid content as well as in physical nature. Any or all of these properties may be implicated.

Although it recently has been demonstrated that dietary lipids are required by the young calf (16), the amount and type of lipid required are not entirely

clear. That very small amounts are satisfactory for a limited time is indicated by performance of subjects on diets 3 and 4. Moreover, the data demonstrate that the blood plasma fat levels of the calf may vary over a wide range, depending on the composition of the diet.

Plasma carotenoids. Group mean blood plasma carotenoid concentrations for calves which were fed milk fat and those of calves which received low carotenoid diets are shown in Figure 2. Although diets 1, 2, 3, and 4 did not promote quite as good growth and general appearance as did diets 5, 6, and 7, there apparently were no deleterious effects attributable to the low concentration of carotenoids in the blood plasma of calves receiving low-fat or hydrogenated soybean oil diets. Since in the present investigation, however, low plasma carotenoid values were associated with low plasma vitamin A values (because of the nature of the dietaries), further study of the physiological significance of carotenoids seems desirable. Plasma carotenoid values were not influenced by the presence of whey product in the ration.

Plasma vitamin A. Since there were no major differences due to whey product feeding, the mean blood plasma vitamin A levels of calves fed low fat diets (3 and 4), hydrogenated oil diets (1 and 2), and milk fat diets (5, 6, and 7), respectively, were compared (Figure 2). All calves were fed vitamin A by cap-
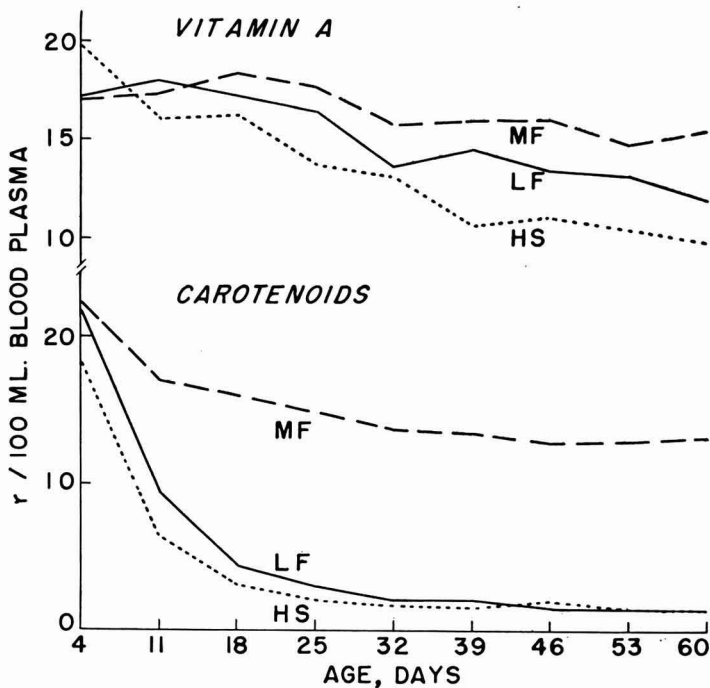
FIG. 2. Blood plasma vitamin A and carotenoid levels (MF = milk fat, from whole milk and butter oil; HS = hydrogenated soybean oil; LF = low fat). Dietary vitamin A equivalents per 100 lb. body weight daily were 18,000 I.U. for MF and 12,000 I.U. for HS and LF.

sule at the rate of 12,000 I.U. per 100 lb. body weight daily, an amount which is in excess of recommended allowances (17). Nevertheless, the blood plasma vitamin A values for calves not fed milk fat decreased steadily during the course of the experiment, suggesting that the supply of vitamin A was inadequate. Since the supplemental vitamin A was the natural ester form in oil and was fed by capsule, which results in a slow rate of absorption (15), the relatively inefficient utilization of this nutrient may have been due in part to the form and method of administration.

The dietary intakes of vitamin A in the low-fat and the hydrogenated soybean oil groups were similar and the calves in the latter groups had slightly lower plasma vitamin A levels, indicating that this lipid had no major effect on vitamin A absorption in the dairy calf. Other data indicate that fat is not essential for vitamin A absorption in human subjects (28) and in rats (11). The higher blood plasma vitamin A levels exhibited by calves which were fed diets containing milk fat were to be expected since the vitamin A activity of the dietary fat increased the total intake to approximately 18,000 I.U. per 100 lb. body weight daily. Even this high level was not sufficient to maintain the plasma vitamin A at initial levels. The apparent inefficient utilization of vitamin A by calves in all groups suggests that some characteristic of all diets in this regime may have interfered with normal vitamin A absorption and/or metabolism.

Hemoglobin. Hemoglobin levels in the blood of calves on all dietary regimes indicated variable but definite downward trends. Since no significant among-group differences were apparent, the data were combined (Figure 3). Wise and associates (29) observed similar levels in calves fed normal diets, except that a slight upward trend was apparent after the fourth week of life. Thomas *et al.* (26) observed somewhat similar changes except that the upward trend occurred later.

Wise *et al.* suggested the need for determining whether erythropoiesis in 4-week-old calves is stimulated by the stage of growth or by dietary changes. The present results indicate that there is no particular stimulation of erythropoietic ability with advancing age in young calves, since there was no marked change from the downward trend in blood hemoglobin concentration. It appears more likely that the calf is born with a supply of iron and copper sufficient for hemoglobin formation during the first several weeks of life. The slightly different results obtained by Wise *et al.* may be due to the fact that these workers provided hay and grain concentrates for all calves after the second week, whereas calves of the present investigation subsisted entirely on the respective "milks" supplemented with vitamin A. The relatively slow rate of growth observed in the present study also could have contributed to the resistance to development of anemia. High blood hemoglobin levels usually were associated with severe diarrhea, and in most instances sharp declines occurred upon recovery, thus indicating that the calf is subject to rapid onset of, and quick recovery from, dehydration.

Blood reducing sugars. Levels of reducing sugars in the blood were subject to irregular fluctuation within groups, and differences between group means were
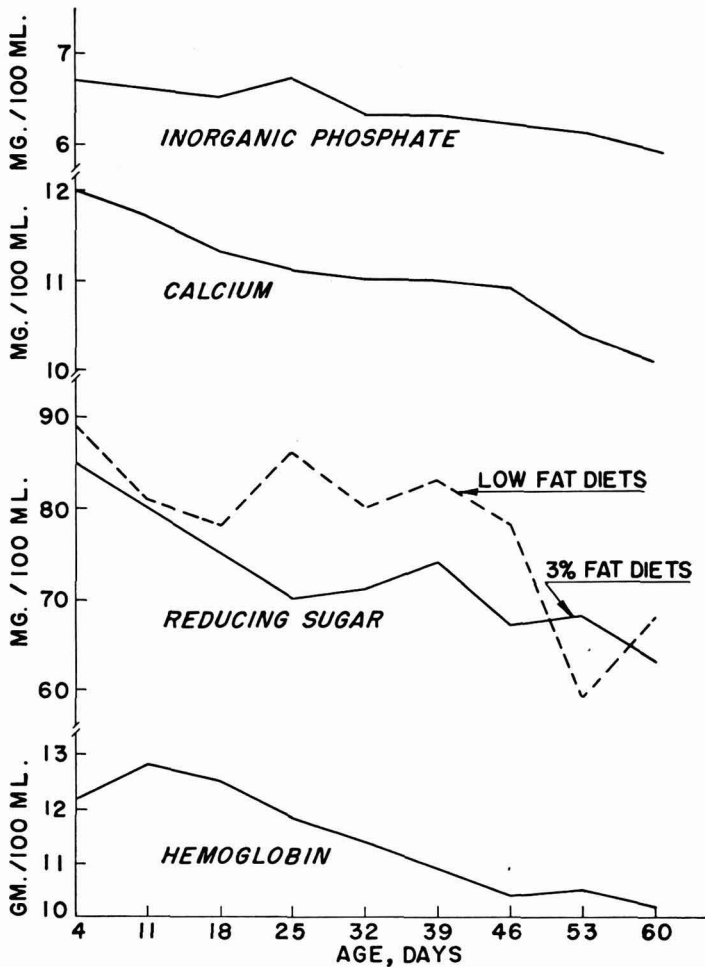
FIG. 3. Mean plasma inorganic phosphate, plasma calcium, blood reducing sugar and hemoglobin values.

small except that calves fed low-fat diets (which were higher in milk solids other than fat) usually exhibited higher blood reducing sugar levels than those fed the other diets. The mean values for calves in the low-fat groups (No. 3 and 4) and for those in the other groups are shown in Figure 3. In both instances the general trend over the 8-week period was downward.

Since varying degrees of excitement, which influence blood reducing sugar levels, accompany most blood collections, the fluctuations observed are not surprising. The significance of the increases which occurred in the mean reducing sugar values at 39 days of age is not clear. Possibly this can be attributed to experimental error, although it seems unlikely since the change was so marked and the number of calves reacting similarly was so large.

Downward trends, similar to those of the present study, have been observed by McCandless and Dye *(19)* in calves fed normal diets. These workers found the glycemic level in calves to vary inversely with the development of the rumen and concluded that rumen fermentation of sugars accounted for the progressive decrease in blood sugar concentration as age increased. This fails to explain the results of the present investigation in which, because of the nature of the dietary, rumen function hardly could have developed in a normal fashion. The calves were fed by nipple pail throughout the experiment, thus insuring passage of most of the material fed directly to the omaso-abomasal area. It must be recognized, however, that complete inactivity of the rumen is difficult to achieve, since a small amount of liquid may be regurgitated to the ruminoreticular area through the reticulo-omasal orifice and since hair and bedding (wood shavings in this instance) sometimes may be ingested.

Plasma calcium. Since no significant group mean differences in plasma calcium levels were observed, all data were composited to establish normal trends for calves fed under these dietary conditions (Figure 3). These values were not markedly different from serum calcium concentrations reported by Wise *et al.* *(29)* for calves receiving normal rations. A gradual decrease in serum calcium was noted following the colostral period. These authors found that at 3 days of age the average for 11 calves was 11.9 mg. per 100 ml. serum. This decreased to 10.7 mg. per 100 ml. when the calves were 60 days of age. The slightly faster rate of decline observed in the current study probably is not significant.

An adverse effect upon calcium metabolism in rats has been reported when high concentrations of lactose were fed; however, blood levels were not altered *(21)*. The relatively high lactose rations included in the present study did not result in blood plasma calcium levels significantly different from those of calves fed other diets. It should be noted, however, that diets 3 and 4 were considerably higher in calcium, as well as in lactose, than the other diets.

The effect of fat upon calcium utilization in laboratory animals has been a controversial issue. Reports of both inhibition *(8)* and promotion *(6)* of calcium utilization by inclusion of dietary fats have been published. The present data do not indicate any influence upon calcium in the blood plasma under the conditions observed. However, the blood plasma calcium level is not considered a positive criterion of the adequacy of calcium nutrition except in cases of extreme deficiency or deranged metabolism *(18)*.

Plasma inorganic phosphate. Mean concentrations of blood plasma inorganic phosphate were similar in all dietary groups. Averages for all calves were plotted as an indication of normal trends under conditions of the experiment (Figure 3). A rise in plasma inorganic phosphate levels was observed during the third week of the present investigation. Flipse *et al.* *(13, 14)* also observed a slight rise in the plasma phosphate levels at about 3 weeks for calves fed restricted diets containing lactose. However, the increases in both instances probably were too small to be significant.

The mean levels of blood plasma inorganic phosphate tended to decrease with age, whereas serum inorganic phosphate observed by Wise *et al.* *(29)* increased.

This difference may be due to diet, since the animals in the latter experiment consumed grain and hay whereas the subjects in the present investigation and those used by Flipse *et al. (13, 14)* were restricted to milk diets. It should be noted, however, that feeding diets high in milk solids other than fat, and therefore higher in phosphorus (diets 3 and 4), did not result in levels of plasma inorganic phosphate significantly different from those in calves fed whole milk or reconstituted milks similar in composition to whole milk.

Phosphate metabolism in the rat appears to be influenced by dietary fat *(7)* and lactose *(20)*. Whether these dietary substances also influenced blood plasma inorganic phosphate levels was not indicated, but such a relationship apparently is not manifested in the dairy calf, since blood plasma levels of inorganic phosphate were comparable for calves fed milks that varied in concentrations of calcium, lactose, and fat and in type of fat.

Body weight changes. Mean weight gains of calves in the various dietary groups are shown in Table 2. Feeding low-fat diets resulted in mean gains in body weight similar to those of calves receiving reconstituted diets containing

TABLE 2

Effect of type of milk on group mean body weight changes [a]

Diet	Age, days		
	4	32	60
		(lb.)	
1	96	107	119
2	91	97	108
3	94	109	123
4	96	109	126
5	86	100	118
6	85	94	110
7	84	98	114

[a] Five calves on diet 4, seven on each of the others.

butter oil. However, two calves fed the low-fat diet containing dried whey product (No. 4) died during the experiment. The average weight gain of calves receiving hydrogenated soybean oil was inferior to that of calves on the other rations. Analysis of variance of the body weight gains indicated that none of these differences were statistically significant.

Since all rations were approximately equivalent with respect to energy content, differences in caloric intake may be disregarded in evaluating the data. The protein content, however, was not constant among rations. The National Research Council *(17)* recommends for young calves a protein intake similar to that normally consumed when whole milk is fed. Diets 2 and 6, which seemed to promote somewhat less growth than diets 1 and 5, contained the same amount of solids-not-fat as whole milk, but half was in the form of dried whey product. Since dried whey product is relatively low in protein (approximately 17%), calves which were fed diets 2 and 6 received less than the recommended allowance of protein. This was not true with diet 4 because of its high content (13%) of nonfat dry milk solids (protein content approximately 37%). Thus, reduced

protein intake may have tended to depress growth rate of calves fed diets containing both fat and whey product.

Incidence of diarrhea. Scouring was most frequent in the low-fat groups, indicating that diets high in concentrations of milk solids-not-fat and/or low in fat are likely to cause diarrhea. This probably was due at least in part to the higher lactose content of the low-fat diets. Moreover, diarrhea was more frequent in calves fed hydrogenated soybean oil than in calves fed butter oil.

Scouring was increased in all instances by the inclusion of dried whey product in the reconstituted diets. This cannot be attributed to the lactose content, since the amounts of this constituent in nonfat dry milk solids and in dry whey product are similar. It seems unlikely that diarrhea could result from differences in protein content of rations containing dried whey product. Therefore, the increase in diarrhea probably resulted at least in part from the higher mineral content of the whey product and/or differences in type of curd formed after ingestion.

SUMMARY

Seven groups of calves were fed, respectively (from 4 to 60 days of age), whole milk and various reconstituted diets which contained different types and amounts of fats and milk solids other than fat. Vitamin A, 12,000 I.U. per 100 lb. body weight daily, was the only supplement. This level of vitamin A supplementation was not adequate to prevent a decrease in plasma vitamin A levels of calves fed low-fat and hydrogenated soybean oil diets. Diets containing milk fat (approximately 18,000 I.U. total vitamin A equivalent per 100 lb. body weight daily) maintained blood plasma vitamin A at relatively constant levels.

Some variations among groups in weight gains were observed but the differences were not significant statistically. Mean blood plasma fat concentrations were highest for calves in the whole milk group, followed in decreasing order by those in groups fed butter oil, hydrogenated soybean oil, and low-fat diets. Feeding of reconstituted diets resulted in decreased blood fat levels during the first week in all instances, in contrast to a rise in blood fat levels in calves which were fed whole milk. Mean blood reducing sugar levels decreased for all groups during the 8-week experimental period. In most instances the values for calves on low-fat diets (high in lactose) were higher than for calves in the other groups. Since there were no apparent differences among the various dietary groups in the other blood constituents investigated (plasma calcium, plasma inorganic phosphate, and hemoglobin), the group mean values were combined to establish average trends.

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YIELDS OF HOLOCELLULOSE PREPARED FROM RUMINANT FECES BY ACID CHLORITE TREATMENT

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A previous report has shown that satisfactory recoveries of the cellulose and hemicellulose fractions of forage are retained in holocellulose prepared by acid chlorite treatment (7). There have been no reports of holocellulose preparations from the feces of ruminant animals. The holocellulose fraction comprises approximately 40 to 60% of the dry matter of common forages (6). In order to study the digestibility of holocellulose and its fractions in various forages it is necessary to have a procedure for the preparation of holocellulose from feces that will give satisfactory yields.

The object of this investigation was to ascertain the yields of holocellulose obtained from ruminant feces when using an acid chlorite treatment that was satisfactory for the preparation of holocellulose from various forages (7).

EXPERIMENTAL PROCEDURE

Wheat straw; alfalfa and corn silage; and timothy, clover, soybean, lespedeza, alfalfa, and orchard grass (young and mature) hays were fed to 10 cows as their entire ration. After a 15-day preliminary feeding period fecal samples were collected four times daily for three consecutive days. Each collection was dried in porcelain trays on a circulating air sample dryer operating at 60 to 70° C. The dried samples for the three consecutive days were compounded and ground through a medium screen Wiley mill and stored for analyses after equilibrating with atmospheric moisture.

Analyses of the fecal samples for protein, ether extract, crude fiber, nitrogen-free extract, and ash contents were made according to procedures described by the Association of Official Agricultural Chemists (1). Lignin was determined by the method of Ellis *et al.* (5). Total carbohydrates were calculated by using the following formula:

$$\text{Total carbohydrates} = (\text{crude fiber} + \text{N.F.E.}) - \text{lignin}$$

Extractive-free feces samples were prepared by extracting with alcohol-benzene followed by hot water extraction according to the procedure described for forage material (7). Analyses of the extractive-free fecal samples for protein, ash, and lignin were made by the procedures used for the original feces. The theoretical holocellulose contents of the various feces were calculated as the lignin and protein-free organic matter of the extractive-free feces dry matter:

$$\text{Theoretical holocellulose} = 100 - (\text{ash} + \text{protein} + \text{lignin})$$
(Ash, protein, and lignin as per cent of extractive-free feces dry matter)

RESULTS AND DISCUSSION

The proximate composition and the lignin and calculated total carbohydrate contents of the 10 fecal samples are presented in Table 1. The feces dry matter had protein contents ranging from 6.0 to 20.0%, crude fiber from 21.4 to 41.4%, lignin from 16.5 to 30.9%, and total carbohydrates from 40.4 to 58.5%. The protein, ash, and lignin contents of the dry matter of the extractive-free fecal samples are given in Table 2. These analyses were used for the calculation of the theoretical holocellulose contents of the feces which are also given in Table 2.

TABLE 1

Per cent composition of dry matter of feces

Animal	Forage fed	Protein	Ether extract	Crude fiber	N.F.E.	Ash	Lignin	Total carbohy-drates
x65	Wheat straw	6.0	1.5	29.8	39.9	22.8	16.5	53.2
N666	Timothy hay	7.3	2.5	33.5	44.6	12.1	24.0	54.1
N645	Corn silage	12.0	1.7	27.3	48.4	10.6	22.6	53.1
N291	Clover hay	12.1	1.9	40.3	37.9	7.8	26.7	51.5
N833	Mature orchard grass	9.5	3.9	33.4	46.8	6.4	21.7	58.5
N295	Soybean hay	10.3	2.4	37.8	31.3	18.2	24.3	44.8
N335	Lespedeza hay	14.4	4.5	31.0	40.3	9.8	30.9	40.4
N689	Alfalfa hay	11.1	3.8	40.5	31.3	13.3	23.4	48.4
N685	Alfalfa silage	15.3	2.7	41.4	31.1	9.5	30.1	42.4
N622	Young orchard grass	20.0	8.1	21.4	41.4	9.1	19.5	43.3

TABLE 2

Per cent composition of dry matter of extractive-free feces

Animal	Forage fed	Protein	Ash	Lignin	Theoretical holocellulose
x65	Wheat straw	5.2	17.0	23.5	54.3
N666	Timothy hay	6.2	11.6	23.4	58.8
N645	Corn silage	11.0	8.2	25.2	55.6
N291	Clover hay	10.9	6.5	29.9	52.7
N833	Mature orchard grass	7.0	4.3	23.7	64.8
N295	Soybean hay	8.9	16.7	26.4	48.0
N335	Lespedeza hay	13.9	8.8	35.8	41.5
N689	Alfalfa hay	8.9	10.5	27.0	53.6
N685	Alfalfa silage	14.4	7.2	32.9	45.5
N622	Young orchard grass	17.1	7.5	27.8	47.6

TABLE 3

Yields of holocellulose from the feces of cows fed ten forages

Animal	Forage fed	No. samples	Average holocellulose yield[a]	Recovery[b]
x65	Wheat straw	8	45.6	98.7
N666	Timothy hay	8	48.5	102.5
N645	Corn silage	8	43.7	99.7
N291	Clover hay	9	50.6	111.7
N833	Mature orchard grass	9	51.7	102.4
N295	Soybean hay	8	42.0	103.4
N335	Lespedeza hay	9	41.2	121.3
N689	Alfalfa hay	9	45.5	105.7
N685	Alfalfa silage	8	45.5	117.9
N622	Young orchard grass	9	31.5	109.7

[a] Ash, protein, and lignin-free holocellulose as per cent of original feces dry matter.
[b] Per cent of calculated theoretical holocellulose.

The average yields of holocellulose (ash, protein, and lignin-free) from each of the feces samples are given in Table 3. The yields are given as the per cent of holocellulose in the original feces dry matter and as the per cent of the theoretical holocellulose recovered. Recoveries of 97 to 103% of the theoretical holocellulose are considered satisfactory. The data in Table 3 show that the recovery of theoretical yields of holocellulose was satisfactory with five of the feces and that the yields were much higher from the remaining feces. The individual preparations from each of these samples were uniformly above the theoretical holocellulose yields. The reducing sugar contents of the hydrolyzed holocellulose samples, with higher than the theoretical yields, were no greater than from the hydrolyzed holocellulose samples with satisfactory yields (6). This indicates that a noncarbohydrate or nonreducing carbohydrate contaminant is present in the holocellulose samples that gave higher than expected yields.

Holocellulose yields from wood that are higher than the theoretical yields have been reported by Campbell and McDonald (3, 4), Herbst (8), Jayme and Finck (9), Muller (10, 11), and Suzuki (12). Total acid hydrolysis of the holocellulose prepared by Muller frequently gave more than the calculated values for reducing substances (10). Suzuki found that lignin partially lost its condensation ability when oxidized by sodium chlorite treatment and dissolved in the acids during the analytical procedure for lignin. The determined lignin contents were lower than the theoretical. Browning and Bublitz (2) reported that an acid-soluble lignin fraction was present in holocellulose products obtained by acid chlorite treatment. Campbell and McDonald found that wood residues (crude holocellulose) after acid chlorite treatment contained a modified lignin that was not determined by the 72% sulfuric acid method. Muller (10, 11) found that when the holocellulose prepared from beechwood shavings by sodium chlorite and chlorine treatment was extracted with potassium carbonate, approximately theoretical yields of holocellulose were obtained. Potassium carbonate, however, removed substantial amounts of pentosans from the holocellulose. Wise et al. (13) pointed out that the accuracy that had been claimed for summative analysis of wood involving holocellulose determinations was fortuitous and many times arose through compensating errors. We have previously discussed this in relation to its use with forage materials (7). Greater errors may be involved in its use with fecal samples.

Additional studies were made to ascertain the effect of repeated acid chlorite treatments on the yield and composition of holocellulose from feces when using two, three, and four treatments. These results are presented in Table 4. We have arbitrarily selected from 97 to 103% of the theoretical holocellulose as a satisfactory yield. When two acid chlorite treatments were used, the yields of holocellulose were unsatisfactory with five of the ten feces samples studied. When three or four acid chlorite treatments were used, the recoveries of holocellulose were unsatisfactorily high in four and three of the samples, respectively. The recoveries were unsatisfactorily low with one and four of the samples studied when three and four acid chlorite treatments, respectively, were used. Satisfactory yields of holocellulose were obtained from the feces samples studied wtih

TABLE 4

Yields and composition of holocellulose from ten feces samples when using 2, 3, and 4 acid chlorite treatments

Animal	Forage fed	Theoretical holocellulose[a]	Holocellulose yield[b] Treatments			Recovery (%) Treatments			Lignin (%)[c] Treatments			Protein (%)[c] Treatments		
			2	3	4	2	3	4	2	3	4	2	3	4
x65	Wheat straw	46.2	45.7	44.2	44.4	98.9	95.7	96.1	4.5	4.0	3.8	2.0	1.7	1.5
N666	Timothy hay	47.3	47.4	47.4	46.5	100.2	100.2	98.3	5.9	5.1	5.1	3.2	4.0	2.0
N645	Corn silage	43.8	45.1	42.5	40.4	103.0	97.0	92.2	5.2	4.5	4.0	5.6	4.0	4.2
N291	Clover hay	45.3	49.6	49.5	49.0	109.5	109.3	108.2	7.1	5.5	4.3	4.7	3.2	3.1
N833	Mature orchard grass	50.5	52.8	51.3	47.2	104.6	101.6	93.5	6.2	6.5	6.8	3.4	2.4	2.7
N295	Soybean hay	40.6	42.1	41.8	38.2	103.7	103.0	94.1	9.7	7.4	7.7	3.8	3.1	3.4
N335	Lespedeza hay	34.0	41.4	40.7	39.8	121.8	119.7	117.0	11.0	8.9	10.4	6.9	4.7	4.5
N689	Alfalfa hay	43.1	45.3	44.4	43.6	105.1	103.0	101.2	12.8	8.9	7.6	4.6	3.9	3.3
N685	Alfalfa silage	38.5	46.5	46.2	42.5	120.8	120.0	110.4	10.9	9.5	8.2	6.4	5.0	5.0
N622	Young orchard grass	28.7	31.5	31.2	29.7	109.8	108.7	103.5	16.3	14.4	12.0	10.4	7.5	6.3

[a] Theoretical holocellulose as per cent of original feces D.M.
[b] Ash, protein, and lignin-free holocellulose yields.
[c] Composition of crude holocellulose.

five, five, and three of the ten samples when using two, three, and four acid chlorite treatments, respectively. These data indicate that a uniform delignification treatment can not be used for the preparation of crude holocellulose samples by gravimetric procedures from a variety of feces. This does not limit the usefulness of the holocellulose procedure with feces, however, since carbohydrate determinations of the crude holocellulose preparations may obviate the necessity of protein and lignin determinations. Inaccuracies of the lignin determinations may seriously limit the validity of the calculated theoretical holocellulose values. Crude holocellulose preparations from feces may also provide satisfactory material for the determination of the alpha cellulose and hemicellulose contents. Wise *et al.* (13) have pointed out that holocellulose preparations from wood are of more value for the determination of the alpha cellulose and hemicellulose contents than for holocellulose as an entity.

The data in Table 4 indicate that the removal of protein and lignin from feces occurs at a slower rate during the fourth acid chlorite treatment than with the second and third treatments. These data also indicate that there is some loss of carbohydrate material with four of the feces samples when given four acid chlorite treatments.

Further studies are necessary to ascertain the nature of the material accounting for higher than theoretical yields of holocellulose from certain feces.

SUMMARY

Holocellulose was prepared from the feces of cows fed ten different forages as their entire ration. A procedure was used that was satisfactory for the preparation of holocellulose from the ten forages fed. Five of the feces samples gave satisfactory recoveries of the calculated theoretical holocellulose contents and five gave higher than the calculated content. Repeated acid chlorite treatments caused a loss of carbohydrate from the holocellulose fraction with certain samples. Reducing properties of the holocellulose hydrolyzates indicate that a noncarbohydrate and nonreducing carbohydrate constituent is present in the holocellulose preparations which gave high yields. A uniform number of acid chlorite treatments with the conditions studied will not prepare holocellulose in theoretical yields with all samples from a variety of feces.

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THE FEEDING VALUE OF EXCELLENT FORAGE
FOR MILK PRODUCTION[1]

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In an earlier publication, McCullough *et al.* *(1)* reported high rates of gain by growing dairy heifers grazing annually seeded winter forages. Although dairy farmers in the South have used this type of forage for several years, few data have been reported relative to its effect on milk production. The experiments described in this paper were initiated to study the use of winter forage in the dairy ration.

EXPERIMENTAL PROCEDURE

The pasture mixture used throughout the experiments was a combination of oats, rye grass, and crimson clover. The experimental animals were Jersey and Guernsey cows from the Station herd which were between 40 and 180 days in lactation at the beginning of the experiments. Within each experiment the animals were grouped according to milk production and stage of lactation. Hay was group fed in racks, grain was fed individually at milking time, and silage was group fed in outside troughs. All feeds were weighed and weighbacks were recorded. Milk weights were recorded daily and 2-day samples for butterfat were taken at weekly intervals.

With the exception of Experiments 2 and 3, forage intake and digestibility measurements were made as described in an earlier publication *(2)*. In Experiments 2 and 3, chromic oxide was administered by capsule (7 g.) twice daily at the morning and evening milkings. Fecal samples were obtained twice daily at each milking (6 A.M. and 4 P.M.) and compounded for each animal for 4 days. The methods of analysis were the same as those referred to above. Chemical compositions of the feeds used are shown in Table 1.

RESULTS AND DISCUSSION

Experiment 1. The original intent in the experiment was to determine the level of milk production which could be maintained with forage alone. Two high and two medium production cows were assigned to a 2.5-acre field and given only minerals and salt in addition to the forage. The data by periods are shown in Table 2. The two high producing cows were about 6 weeks in lactation and the two low producers were about 12 weeks. This may explain the greater response of the two high producers in Period 2, but of greater interest is the persistency of all four cows throughout the experiment. The dry matter intake (estimated with Cr_2O_3) averaged 33 lb. per cow per day for all four cows. The estimate was

TABLE 1

The chemical composition of the feeds and forages fed[a] (dry matter basis)

Feed	Experi-ment	Crude protein	Crude fiber	Ether extract	NFE	Ash
		(%)	*(%)*	*(%)*	*(%)*	*(%)*
Pasture forage (3/5)	1	24.39	17.89	4.02	44.67	9.03
Pasture forage (3/24)	1	21.02	23.02	3.50	42.81	9.65
Pasture forage (4/6)	1	17.44	23.43	3.51	46.33	9.29
Pasture forage	3	28.54	14.21	7.10	43.82	7.10
Hay[b]	3	11.22	29.09	3.87	51.80	3.99
Corn silage	3	6.29	29.71	4.11	56.48	3.39
Grain rations	2 and 3	15.11	9.17	5.26	66.38	4.06
Grain ration	4	26.34	6.88	5.23	55.86	5.69
Hay[c]	4	5.35	33.12	1.51	55.26	4.75

[a] Chemical analysis of hay used in Experiment 2 was not available. The alfalfa was U.S. No. 2 and the lespedeza U.S. No. 2.

[b] Lespedeza hay—U.S. No. 1.

[c] Common Bermuda—sample grade.

an average of four measurements taken at 2-week intervals. At the end of 8 weeks the animals had dropped only 8% in milk production. In the first month the animals gained 8% in milk production, and at the end of 6 weeks milk production was 3% greater than at the beginning. The changes in body weight were erratic but were of interest if only to indicate that even the high-producing cows did not lose body weight.

So long as the forage dry matter remained above 70% digestible, the forage stimulated milk production at a high level, but at 66% digestibility production fell rapidly. A point of practical significance in pasture studies was the apparent low utilization of the forage by the two low producing cows. If it can be assumed that four cows with the same production level would have reacted the same as the two high producers, the pasture would have been credited with 3,983 lb. milk per acre. Four low producers averaging the same as the two cows used would have credited the pasture with 2,066 lb. milk per acre. The gain in body weight with the two groups would have been 92 lb. and 146 lb., respectively. To obtain a proper evaluation of the milk producing capacity of this type of forage apparently requires the use of cows producing above 40 lb. of FCM per day.

Experiments 2 and 3. The object of these two experiments was to determine if limited quantities of this excellent forage could be used to stimulate the milk production of cows being fed good barn rations. In Experiment 2, 12 cows were divided into two groups of six cows. Group 1 was fed alfalfa hay free choice and Group 2 was fed kobe lespedeza hay free choice. All cows were permitted to graze for 2 hours after the morning milking. In Experiment 3, two groups of four cows each were fed 50 lb. of corn silage and 10 lb. of lespedeza hay per cow per day. Group 1 received no grazing and a low protein grain ration at the rate of 1 lb. of grain to 4 lb. of milk, and Group 2 received 2 hours of grazing and no grain. The data for the 30-day period are shown in Table 3. The cows being fed good legume hay and a small amount of grain maintained milk production at a higher than expected rate with the 2 hours of grazing. Apparently this type of ration would be suitable for cows producing about 20 lb. of milk per day. The

TABLE 2

Milk production, weight changes, and dry matter digestibility of cows receiving forage alone
(Experiment 1)

Cow	March 1-14			March 15-28			March 29-April 12			April 13-27		
	Av. FCM	Weight change	D.M. dig.	Av. FCM	Weight change	D.M. dig.	Av. FCM	Weight change	D.M. dig.	Av. FCM	Weight change	D.M. dig.
	(lb.)	(lb.)	(%)	(lb.)	(lb.)	(%)	(lb.)	(lb.)	(%)	(lb.)	(lb.)	(%)
Julia	40.3	+77	78	47.1	−40	78	46.1	+2	74	40.5	−12	67
Lassie	42.2	+19	75	49.1	+49	76	49.3	+4	75	40.4	+16	64
Jan	27.7	+47	80	27.6	+38	79	21.4	−10	73	22.2	+32	69
M. June	23.7	−34	76	20.9	+78	81	20.6	+32	74	20.4	0	66
Period av.	33.4	+27	77	36.1	+31	78	34.4	+7	74	30.8	+15	66

TABLE 3

The feed consumption, milk production, and weight changes of the cows in Experiments 2 and 3

| Group | Feed consumed (D.M. basis) | | | | Beginning milk prod. (FCM) | 30-day change | |
	Forage	Silage	Hay	Grain		FCM	Body wt.
	(lb.)	*(lb.)*	*(lb.)*	*(lb.)*	*(lb.)*	*(%)*	*(lb.)*
Experiment 2							
1 (Alfalfa)	4.9	0.0	22.0	3.3	21.7	+1	+14
2 (Lespedeza)	8.9	0.0	18.0	2.5	20.5	+6	−21
Experiment 3							
1	0.0	15.0	9.4	7.4	28.0	−3	−15
2	2.4	14.0	9.4	0.0	27.0	+13	+3

cows in Experiment 3 increased in milk production, although the small amount of forage was replacing 7 lb. of grain. In both experiments the forage dry matter digestibility was about 75%. It should also be noted that the hay and silage used in both experiments was apparently high quality roughage.

The forage dry matter intake was of interest in the two experiments, since it showed an apparent effect of feeding on forage intake. When the cows were consuming 18 lb. of lespedeza hay they consumed an estimated 8.9 lb. of forage dry matter, but with a ration of 50 lb. of silage and 10 lb. of hay the forage intake was 2.4 lb. Since the supply of this high quality forage is often a limiting factor in its use, the feeding of a full feed of good roughage may be a practical way of extending its use.

Experiment 4. The results of Experiment 4 are presented here only to show that small amounts of this high quality forage did not greatly improve the ration when poor hay was fed. The hay used was a stemmy, bleached, common Bermuda hay that would have graded sample. The results are shown in Table 4. The results of this trial indicate that the stimulating qualities of excellent forage are dependent upon an adequate quantity of nutrients in the ration and should not be depended upon to maintain milk production by increasing the utilization of the feed to overcome the effects of poor feeding.

SUMMARY

The results of four experiments involving the use of excellent winter pasture are shown. The forage maintained milk production at a level in excess of 40 lb. per day when fed as the sole ration so long as the dry matter digestibility re-

TABLE 4

The changes in milk production and body weight of the cows in Experiment 4

| Ration | Beginning milk prod. | 21-day change | |
		FCM	Body wt.
	(lb.)	*(%)*	*(lb.)*
Hay 20 lb. + 2 hr. grazing	28	−31	−5.6
Hay 20 lb. + 7 lb. grain + 2 hr. grazing	28	−22	−1.0
Hay 20 lb. + 9 lb. grain	26	−31	−3.0

mained above 70%. Two hours of grazing per day on similar quality forage maintained a 20 lb. level of production when the cows were also fed U. S. No. 2 hay free choice. The same amount of grazing maintained a 28-lb. level of production when the cows also received good corn silage and U. S. No. 1 lespedeza hay. The limited forage was not adequate to maintain production when fed with poor hay, apparently because of lack of ration nutrients. The free choice feeding of good roughage may also reduce the amount of pasture consumed and thus extend the number of cows which can be grazed per acre.

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METABOLISM OF BULL SEMEN. I. INORGANIC AND
TOTAL PHOSPHORUS RELATIONSHIPS[1]

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Excellent reviews *(17, 19, 22)* on semen metabolism have indicated the important role of phosphorus-containing enzymes in sperm glycolysis. Furthermore, glycolysis is an important source of sperm energy and appears to be the preferential source when ample quantities of suitable substrates are available *(17, 18, 20)*.

Many enzymes have been identified in the semen of several species with indication of considerable interspecies variation *(17, 22)*. MacLeod *(18, 19)* and Lundquist *(16, 17)* have observed enzymes in human semen capable of dephosphorylating phosphate esters.

Although semen of the bull is not particularly high in alkaline phosphatase and even lower in acid phosphatase *(22, 24)* as compared with the other species, it is a particularly rich source of 5-nucleotidase *(21)*. Washed sperm suspensions utilize glycolyzable sugars in preference to phospholipids *(14)*. Phospholipids provide a source of energy in sugar-free suspensions under aerobic conditions *(12, 13)*. Phosphate is required for maintenance of glycolysis and motility *(15)*, though it was recently suggested that phosphate-containing diluents are associated with motility inhibition even when peroxide accumulation is prevented with catalase *(2)*. Concentration of some, but not all, inorganic constituents in semen has been reported *(1, 5, 6, 9, 10, 11, 16, 21, 23, 27)*.

Inorganic phosphorus and total phosphorus were selected for more intensive study to determine: *(a)* levels of inorganic and organic phosphorus in semen of highly fertile bulls, *(b)* how these levels relate to other physical and metabolic measures of semen quality, and *(c)* how these compare with nonreturn rates.

PROCEDURE

Semen samples collected between June, 1952, and January, 1953, from 36 bulls (four breeds) in routine artificial service at Northwest Coop Breeders, Burlington, Wash., were used during this study. Bulls were fed good quality alfalfa hay, silage, and concentrates. Semen was collected daily excepting Sundays and diluted with antibiotic-treated heated homogenized milk at a rate giving in excess of 10^7 sperm per milliliter of diluted semen. Sixty- to 90-day nonreturns were determined from first and second services, with 70% of the services occurring the day after collection. Initial motility ratings were made as previously

described (2). Concentration of sperm was determined with a calibrated Leitz. Photelometer.

Immediately after collection of semen, 0.5 ml. was added to 2.0 ml. of 2.9% Na citrate buffer at 37° C. One ml. of this mixture was pipetted directly into 19 ml. of 10% trichloracetic acid. Determinations of fructose and inorganic phosphorus were made on the filtrate obtained by centrifugation. This process was repeated after incubation for 1 hour. Fructose was determined by the method of Roe (25) modified by taking 2 ml. of the trichloracetic acid filtrate and adding 6 ml. of a solution which is 10 N HCl (80% by volume) and 0.034 g. of resorcinol per 100 ml. Colorimetric determinations were made with an Evelyn Colorimeter.

The method of Fiske and SubbaRow (4) was used for determination of phosphorus. Total phosphorus was determined by the same method after wet ashing 0.02 ml. of the Na citrate-semen mixture with perchloric acid. Statistical procedures were as outlined by Snedecor (26).

RESULTS AND DISCUSSION

Determinations were made on 506 semen samples (36 bulls) of which 400 (35 bulls) were used for 20 or more first and second services (3). As shown in Table 1, total phosphorus increased almost in direct proportion with increase in sperm concentration. Increase in inorganic phosphorus was not as pronounced, though there was a positive relationship which amounted to approximately a 30% increase through the concentration range shown. The ratio of organic to inorganic phosphorus shows nearly a threefold increase through the concentration range under study.

As shown in Table 2, inorganic phosphorus increased 1.4 mg. % or 8.2% during incubation (P < 0.01). Of these 400 samples, 281 increased and 119 decreased. This small increase was not unexpected since acid and alkaline phosphatase has previously been found low (22, 24) for bull semen. This is in contrast with human semen, since Lundquist (16, 17) and MacLeod (18, 19) have demonstrated its ability to dephosphorylate phosphate esters. Lundquist observed an increase in inorganic phosphate from 21.6 to 64.2 mg. % during 20 minutes incubation.

Phosphorus turnover is cyclic and can transfer from one organic molecule to another without passing through the inorganic stage or be exchanged freely between the two phases (8). This may account for the failure of inorganic phosphorus to show greater accumulation under the conditions of this study. Washed sperm suspensions allowed to metabolize in the absence of fructose should furnish inorganic phosphorus, since Lardy and Phillips (12, 13, 14) have shown that endogenous metabolism is supported by phospholipid utilization.

However, 75 semen samples in this study with less than 0.75 mg. % of fructose per milliliter of semen after incubation showed only 0.7 mg. % increase in inorganic phosphorus level, compared with 1.8 mg. % for 74 samples containing more

TABLE 1

Relation of sperm concentration and phosphorus levels in semen

Concentration	Samples	Conc.	Total P	Inorganic P			Organic P[a]		Organic:inorganic	
				Initial	1 hour[b]	Change	Initial	1 hour[b]	Initial	1 hour
(10⁶/ml)	(No.)	(10⁶/ml)	(mg.%)	(mg.%)	(mg.%)	(mg.%)	(mg.%)	(mg.%)	(Ratio)	(Ratio)
400-499	6	445	64.3	16.5	15.7	-0.8	47.8	48.6	2.90	3.10
500-599	3	560	62.3	14.3	15.3	-1.0	48.0	47.0	3.36	3.07
600-699	9	642	71.7	13.0	12.4	-0.6	58.7	59.3	4.52	4.78
700-799	17	754	85.8	15.0	15.6	0.6	70.8	70.2	4.72	3.03
800-899	18	840	92.4	13.5	17.2	3.7	78.9	75.2	5.84	4.37
900-999	16	931	100.6	15.6	15.9	0.3	84.4	84.7	5.41	5.33
1,000-1,099	16	1,055	96.1	15.0	20.0	5.0	81.1	76.1	5.40	3.81
1,100-1,199	25	1,134	116.9	16.9	18.0	1.1	100.0	98.9	5.92	5.49
1,200-1,299	32	1,246	116.6	15.2	17.3	2.1	101.4	99.3	6.67	5.74
1,300-1,399	23	1,348	122.0	14.4	15.3	0.9	107.6	106.7	7.47	6.97
1,400-1,499	23	1,435	118.2	14.4	17.5	3.1	103.8	100.7	7.21	5.75
1,500-1,599	43	1,538	132.4	16.8	17.1	0.3	115.6	115.3	6.88	6.74
1,600-1,699	30	1,651	145.6	17.8	18.6	0.8	127.8	127.0	7.18	6.83
1,700-1,799	31	1,760	148.5	16.6	17.8	1.2	131.9	130.7	7.95	7.34
1,800-1,899	27	1,861	156.8	16.9	17.7	0.8	139.9	139.1	8.28	7.86
1,900-1,999	24	1,940	156.9	18.0	17.5	-0.5	138.9	139.4	7.72	7.97
2,000-2,099	39	2,038	169.3	17.1	18.2	1.1	152.2	151.1	8.90	8.30
2,100-2,199	22	2,152	170.3	15.9	17.4	1.5	154.4	152.9	9.71	8.79
2,200-2,299	15	2,234	181.7	18.3	18.5	0.2	163.4	163.2	8.93	8.82
2,300-2,399	33	2,331	193.5	20.8	23.1	2.3	172.7	170.4	8.30	7.38
2,400-2,499	13	2,426	207.5	21.7	21.6	-0.1	185.8	185.9	8.56	8.61
2,500-2,599	16	2,537	219.0	20.3	22.3	2.0	198.7	196.7	9.79	8.82
2,600-2,699	9	2,649	220.2	19.9	25.6	5.7	200.3	194.6	10.07	7.60
2,700-2,799	8	2,728	216.5	26.1	25.5	-0.6	190.4	191.0	7.30	7.49
2,800-2,899	8	2,921	254.9	17.5	20.9	3.4	237.4	234.0	13.57	11.20
Total	506	1,672	145.9	17.0	18.4	1.4	128.9	127.5	7.58	6.93

[a] Total phosphorus minus inorganic phosphorus.
[b] After incubation for one hour at 37° C.

TABLE 2
Average of semen quality measurements

Measurement	Sample	Inorganic P			Total P	Total P adj. to zero sperm concentration [c]	Sperm conc.	Initial motility	Initial fructose	Fructose Utiliza. [b]	60 to 90 day non-return
		Initial	1 hour [a]	Change							
	(No.)	(mg. %)	(mg. %)	(mg. %)	(mg. %)	(mg. %)	(10^6/ml)	(Rating)	(mg/ml)	(mg/ml)	(%)
Guernsey	143	16.5	17.1	0.6	126	31.5	1,400	7.3	7.33	2.95	70.6
Holstein	144	16.4	19.0	2.5	152	29.9	1,720	7.8	6.08	3.34	72.6
Jersey	78	16.4	17.8	1.4	163	34.9	1,960	8.4	4.93	3.54	73.9
Hereford	35	18.5	18.8	0.3	127	21.1	1,550	7.1	5.17	2.26	70.2
Total	400	16.6	18.0	1.4	143	31.4	1,636	7.7	6.22	3.15	71.9
Standard deviation	400	7.6	7.4	6.5	43	4.4	551	1.3	1.86	.93	8.9
Standard error of mean	400	0.4	0.4	0.3	2	0.2	28	0.1	.09	.05	0.4

[a] Incubation at 37° C.
[b] Utilized during incubation for one hour at 37° C.
[c] Linear regression coefficient = 0.068 mg. % increase in total phosphorus as sperm concentration increased 10^6.

than 5.00 mg. % of fructose after incubation. Although these samples were incubated under aerobic conditions (12, 13), they still contained fructose (17, 18, 20) and other constituents of seminal plasma (17, 19, 22).

The average for inorganic phosphorus of 16.6 before and 18.0 mg. % after incubation (Table 1) is slightly higher than the average of 13.7 mg. % reported by Bernstein (1), when the latter's data are expressed as phosphorus instead of P_2O_5. The average for total phosphorus was 143 mg. % for this study compared with 55 mg. % from the study of Bernstein. This rather wide discrepancy may be due almost entirely to differences in sperm concentration (Table 1). Estimating total phosphorus by using the linear regression coefficient for total phosphorus and concentration yields an average estimated value for seminal plasma of 31 ± 1.0 mg. % (Table 2). Breeds and bulls within breed were significantly different (P < 0.01), but the variance component was only 4.6 and 1.2%, respectively, for breeds and bulls as compared with 21.4 and 34.9% for total phosphorus (Table 3). Hence, there may not be any real discrepancy between these two studies.

Differences associated with bulls and months are shown in Table 3. Of 36 bulls in the study, 16 were used regularly during all months. Analysis of variance was made by using the bull × months means. Considerable monthly variation was observed for inorganic phosphorus content (P < 0.01) before and after incubation, with no significant changes during incubation. Bulls did not vary significantly. Total phosphorus was relatively constant for months but was significantly different for bulls (P < 0.01). This can be accounted for by the wide variation in

TABLE 3

Variations in inorganic and total phosphorus in semen for months, bulls, and breeds

Source of variation		Inorganic P			Total P	Total P adj. to zero sperm concentration
		Initial	1 hour [a]	Change		
(months)		(mg. %)	(mg. %)	(mg. %)	(mg. %)	
July		16.7	17.5	0.8	149	
August		13.3	14.1	0.8	145	
September		18.8	18.7	−0.1	158	
October		22.1	24.4	2.3	149	
November		19.2	21.7	2.5	166	
December		15.1	17.2	2.1	145	
January		13.8	16.1	2.3	138	
Average		17.0	18.5	1.5	150	
Component of variance						
	D.F.	(%)	(%)	(%)	(%)	(%)
Bulls	15	2.5	0.0	2.5	45.6 [b]	
Months	6	39.3 [b]	41.6 [b]	0.6	3.6	
Bulls × months	90	58.2	58.4	96.9	50.8	
Breeds	3	0.0	0.7	1.7 [c]	21.4 [b]	4.6 [c]
Bulls within breed	31	1.3	0.0	0.9	34.9 [b]	1.2 [c]
Sample within bull	365	98.7	99.3	97.4	43.8	94.2

[a] Incubation at 37° C.

[b] Significant at 1% level of probability.

[c] Significant at 5% level of probability.

average concentration of sperm for the 16 bulls included in this study (range 1,101 to 2,272 10^6 sperm per milliliter).

When all bulls and 400 semen samples were used (Table 2), no significant differences were observed between breeds or bulls for levels of inorganic phosphorus with highly significant differences between breeds and bulls within breed for total phosphorus.

Comparison of inorganic and total phosphorus with other semen measures is shown in Table 4. As would be expected from results shown in Table 1, total phosphorus was highly correlated with concentration of sperm ($r = 0.88$). This accounts for 77% of between-sample variation. Total phosphorus was also significantly correlated with initial motility rating, initial fructose, and fructose utilization. These measures were also significantly correlated with concentration and in essentially the same manner.

Inorganic phosphorus before and after samples were incubated showed small but significant correlation with sperm concentration, total phosphorus, and initial fructose. Change in inorganic phosphorus during incubation was not correlated with any measures shown in Table 4 except levels of inorganic phosphorus before and after incubation.

Neither inorganic nor total phosphorus showed any relation to nonreturn rates. Under the conditions of this study, the inorganic phosphorus content of semen shows little variability between bulls other than small differences associated with varying concentration. Neither is it related to initial motility or nonreturn, though Lardy and Phillips (15) observed phosphate necessary for maintenance of motility, and Bishop and Salisbury (2) found excess phosphate inhibitory. These findings are not necessarily discrepant since experimental objectives were markedly different.

The relationship with fructose and fructose utilization (a measure of glycolytic ability) exists but is not significant under the conditions of this study. Different experimental methods may later show different results, since it is felt that the Seliwanoff reaction as used measured only fructose per se and not the intermediate esters of fructolysis, such as 1,6-diphosphofructose. On the other hand, in analysis of total phosphorus the latter are certainly included. Furthermore, study of low rather than high fertility bulls may reveal imbalances not present in semen of high fertility.

Total phosphorus content of semen shows great variability between bulls and samples within bulls, which is directly associated with differences in concentration. This source of variation should not be overlooked when comparing results of different experiments.

SUMMARY

Inorganic and total phosphorus determinations on 506 semen samples collected over a 7-month period from 36 bulls of high fertility showed a marked relationship between sperm concentration and total phosphorus ($r = 0.88$). Inorganic phosphorus content of semen before and after incubation for 1 hour at

TABLE 4

Analyses of covariance

Linear correlation

Initial inorganic phosphorus

Measurement	Total	Breeds	Bulls	Residual
Degrees of freedom	399	3	31	365
Inorganic P initial				
Inorganic P 1 hour	0.62ᵃ	0.24	0.61ᵃ	0.63ᵃ
Inorganic P change	-0.46ᵃ	-0.42	-0.57ᵃ	-0.45ᵃ
Sperm concentration	0.23ᵃ	-0.18	0.52ᵃ	0.21ᵃ
Total P	0.29ᵃ	-0.38	0.48ᵃ	0.31ᵃ
Initial motility	0.07	-0.50	0.20	0.07
Initial fructose	-0.15ᵃ	-0.30	-0.29	-0.17ᵃ
Fructose utilization	0.12ᵇ	-0.81	0.39ᵇ	0.12ᵇ
Per cent nonreturn	-0.03	-0.48	-0.09	-0.02

Inorganic phosphorus after incubation

Measurement	Total	Breeds	Bulls	Residual
Degrees of freedom	399	3	31	365
Inorganic P initial	0.62ᵃ	0.24	0.61ᵃ	0.63ᵃ
Inorganic P 1 hour				
Inorganic P change	0.41ᵃ	0.78	0.30	0.41ᵃ
Sperm concentration	0.21ᵃ	0.54	0.47ᵃ	0.17ᵃ
Total P	0.29ᵃ	0.55	0.50ᵃ	0.26ᵃ
Initial motility	0.04	0.22	0.02	0.04
Initial fructose	-0.15ᵃ	-0.58	-0.20	-0.16ᵃ
Fructose utilization	0.09	0.19	0.44ᵇ	0.04
Per cent nonreturn	0.03	0.44	0.08	0.02

Change in inorganic phosphorus

Measurement	Total	Breeds	Bulls	Residual
Degrees of freedom	399	3	31	365
Inorganic P initial	-0.46ᵃ	-0.42	-0.57ᵃ	-0.45ᵃ
Inorganic P 1 hour	0.41ᵃ	0.78	0.30	0.41ᵃ
Inorganic P change				
Sperm concentration	-0.03	0.62	-0.13	-0.05
Total P	0.00	0.08	-0.01	-0.01
Initial motility	-0.04	0.54	-0.21	-0.03
Initial fructose	0.01	-0.35	0.14	0.02
Fructose utilization	-0.04	0.70	-0.01	-0.10
Per cent nonreturn	0.07	0.50	0.18	0.04

Total phosphorus

Measurement	Total	Breeds	Bulls	Residual
Degrees of freedom	399	3	31	365
Inorganic P initial	0.29ᵃ	-0.38	0.48ᵃ	0.31ᵃ
Inorganic P 1 hour	0.29ᵃ	0.55	0.50ᵃ	0.26ᵃ
Inorganic P change	-0.01	0.08	-0.01	-0.01
Sperm concentration	0.88ᵃ	0.97ᵇ	0.94ᵃ	0.84ᵃ
Total P				
Initial motility	0.56ᵃ	0.95ᵇ	0.61ᵃ	0.46ᵃ
Initial fructose	-0.45ᵃ	-0.75	-0.35	-0.48ᵃ
Fructose utilization	0.55ᵃ	0.85	0.65ᵃ	0.46ᵃ
Per cent nonreturn	0.08	0.99ᵃ	0.16	-0.02

ᵃ Significant at 1% level of probability.
ᵇ Significant at 5% level of probability.

37° C. was most closely correlated with sperm concentration (r = 0.23) and showed unimportant relationships with initial motility, fructose, and fructose utilization.

Inorganic phosphorus increased 1.4 mg. % or 8.2% (P < 0.01) during incubation for 1 hour at 37° C. This change was not related to bulls, breeds, months, or any of the semen quality measures. There was no relationship between inorganic and total phosphorus and nonreturn rate.

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RECTAL TEMPERATURE AND RESPIRATORY RESPONSES OF JERSEY AND SINDHI-JERSEY (F₁) CROSSBRED FEMALES TO A STANDARD HOT ATMOSPHERE

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In the course of systematic studies of crossbred cattle resulting from an experimental breeding program made with Jerseys and Red Sindhi as the parent stocks, a number of F₁ Sindhi-Jersey females have been subjected to a standard hot atmosphere to determine their responses to hot atmospheric conditions. The effect of such exposures on rectal temperature and respiratory rate is reported in this paper.

EXPERIMENTAL CONDITIONS

Each female produced by the breeding program is subjected to the standard hot atmosphere for 6 hours at approximately 2-month intervals from 6 months of age onward except during the first lactation, which is left undisturbed for record purposes, and during the 45 days preceding each expected parturition. The test animals are placed in a climatic chamber on the afternoon preceding the test, but without any heat. Water, grain, and hay are allowed ad libitum during the afternoon. Grain is also offered the following morning, but all feed is withdrawn before the test is started. Water is available to the animal up to the time of starting the test, but no feed or water is offered during the test period. At about seven in the morning, the heaters and humidifiers are switched on, and the air conditions rise fairly rapidly (15 minutes in summer, 20 minutes in winter) to the desired 105° F. and 34 mm. Hg vapor pressure (corresponding to a wet bulb temperature of 92° F. and a relative humidity of 60%). They are maintained at these levels, within 2° F. dry- and wet-bulb temperatures, for the remainder of the 6-hour test period. Air temperatures near the floor are somewhat lower than 105° F. until the end of the first hour.

Rectal temperature is measured by a 5-in. clinical thermometer inserted into the rectum and left in position for 3 minutes. Under hot conditions the chance of error from variations in the position of the thermometer is small. Respiratory rate is measured by counting flank movements over an undisturbed period of 1 minute. Readings are made just before the heat is switched on and at hourly intervals thereafter. Other responses measured during the test, such as water loss and skin wetness, will be reported elsewhere.

RESULTS

To reduce the numerous individual observations to manageable proportions, the mean of the observations during the test has been taken as the representative datum. The use of the mean has the advantage of minimizing the error inherent

in single observations and avoids the uncertainty of interpretation attending such criteria as maximum temperatures. The trapezoidal mean has been used in preference to the straight mean, as more representative of the animal's state while under test, especially when the initial rise of rectal temperature is rapid.

$$t_m = (0.5t_o + t_1 + t_2 + t_3 + t_4 + t_5 + 0.5t_6)/6$$

where t_m is trapezoidal mean temperature; t_1, t_2, etc. are temperatures observed after 1, 2, etc. hours of exposure.

Rectal temperature. The results obtained in these studies are illustrated in Figures 1 and 2, and the analyses of variance are given in Table 1. In these analyses the method recommended by Snedecor *(6)* for dealing with disproportionate subclass numbers was used. Data are given for both the absolute rectal temperature during exposure and the rise of rectal temperature above the initial value. The latter is probably a better indicator of the additional strain developed in the animal by exposure to heat, but the former indicates the total strain experienced by the animal and thus probably comes closer to suggesting the economic consequences of the situation.

Exposure to heat results in a marked rise of rectal temperature in all three categories of animals—heifers, dry cows, and lactating cows. If final, or maximum, rectal temperatures had been reported instead of the exposure means, the rise would have been even more striking, since the rectal temperature tends to go on increasing throughout the exposure, especially in the less tolerant animals.
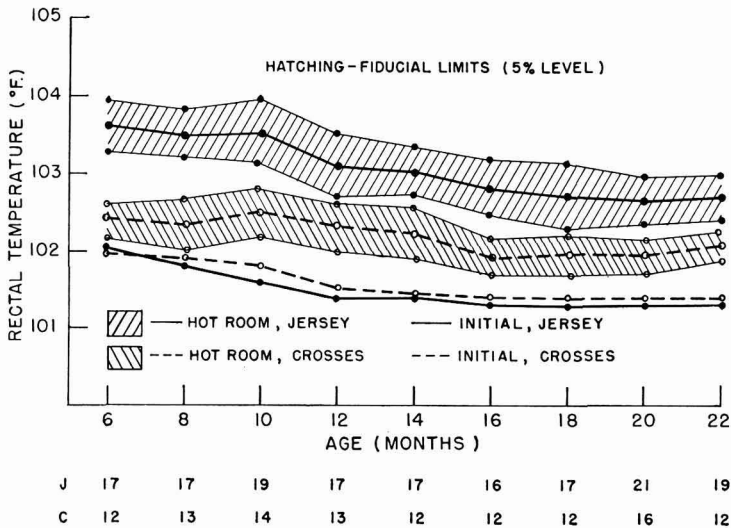

FIG. 1. Initial and mean rectal temperatures of heifers during exposure to a standard hot atmosphere (6 hours at 105° F. with 34 mm. Hg vapor pressure). Numbers at foot indicate the number of animals, Jerseys (J) and crossbreds (C), tested at each age.
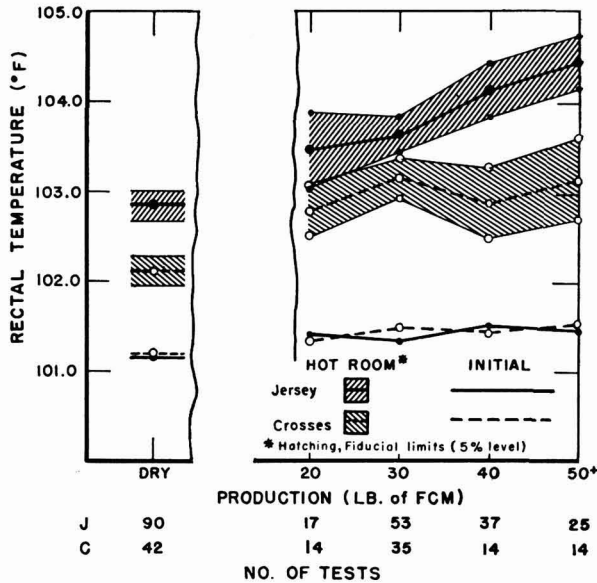
Fɪɢ. 2. Initial and mean rectal temperatures during exposure to a standard hot atmosphere (ʋ hours at 105° F. with 34 mm. Hg vapor pressure) by average level of milk production on the five days preceding the test. Numbers at foot indicate tests performed on Jerseys (J) and crossbreds (C) at various levels of production.

In all three categories both the mean rectal temperature during exposure and the mean rise in rectal temperature above the initial value are greater to a highly significant degree in the Jerseys than in the Sindhi-Jersey crossbred animals.

In heifers, the rectal temperature during exposure is higher in the younger animals, but so is the initial temperature, so that the rise on exposure is substantially the same at all ages (Figure 1 and Table 1). Dry cows show much the same reaction as do the older heifers (Figure 2).

In lactating cows, the stage of lactation has little effect upon the reactions to heat. In Jerseys, the rectal temperature on exposure tends to be higher in the earlier months of lactation, but this is also accompanied by a higher initial temperature, so that the rise is substantially the same at all stages. In Sindhi-Jersey crossbred cows, neither the initial rectal temperature nor the mean obtained during exposure is affected by the stage of lactation.

The level of production, in sharp contrast to the stage of lactation, has a highly significant effect upon both the absolute and the relative rectal temperature during exposure (Figure 2). When dry cows are included, a significant difference is seen between the development of this effect in the two groups. The rectal temperature during exposure tends to rise steadily with the production level in the Jerseys, but in the Sindhi-Jersey crossbred cows it rises with the lower levels of production to a value which is not further affected by further increases in production. This interaction between breed and production level is

TABLE 1

*Analyses of variance of the initial rectal temperature, mean rectal temperature
on exposure, and rise in rectal temperature with exposure*

Source of variation	Degrees of freedom	Mean squares		
		Initial rectal temp.	Mean rectal temp. on exposure	Mean rise in rectal temp. with exposure
(a) Heifers				
Breeds	1	0.325	50.289**	55.472**
Ages	8	1.910**	3.085**	0.656
Interaction	8	0.022	0.369	0.382
Error	258	0.132	0.426	0.396
(b) Lactating cows by months of lactation				
Breeds	1	0.300	16.554**	20.349**
Months	3	0.640*	1.807*	0.648
Interaction	3	0.083	0.654	1.178
Error	130	0.165	0.562	0.527
(c) Dry cows				
Breeds	1	0.016	15.307**	15.311**
Error	130	0.118	0.502	0.503
(d) Cows by levels of production (excluding dry cows)				
Breeds	1	0.215	35.314**	37.634**
Levels	3	0.152	2.398**	1.656*
Interaction	3	0.126	2.239**	1.761*
Error	201	0.185	0.408	0.608

* Significant at the 5% level of probability.
** Significant at the 1% level of probability.

substantiated by a highly significant mean square in Table 1. The initial rectal temperatures of lactating cows are higher than those of dry cows, but there is no progressive rise with the level of production.

Since seasonal variations in the maintenance environment of animals subjected only periodically to testing may influence their reactions to such tests, the responses of animals to the standard hot atmosphere were examined in relation to season. The analysis and results have been discussed at some length in a previous publication (3), and only a summary needs to be given here. It was found that there was little repeatability in the responses of the animals unless they were first sorted according to season. When the responses of heifers and dry cows were plotted by months of the year, an unexpected trend was revealed. The responses showed a maximum in February with a smaller maximum in August, separated by a marked minimum in May-June. The values in November-December were intermediate. This type of fluctuation was easily observed in the Jerseys, which have the greater response, but it was still discernible in the crossbred animals. When the responses for heifers were sorted by season, as suggested by this variation, a fairly high repeatability was found (Table 2). No repeatability was found with lactating cows, however, as was to be expected in view of the multiplicity of factors which affect lactation and its consequences for the animal.

Respiration rate. Respiratory data are given in Figures 3 and 4, and the analyses of variance in Table 3. Exposure to heat results in a very marked rise

TABLE 2

Repeatability [a] *of mean body temperature responses* [b] *of heifers (8-22 months of age)
to a standard test hot atmosphere (6 hours at 105° F. and 34
mm. Hg vapor pressure) when sorted by season*

		Repeatability within seasonal group		
Breed	Item	Apr.-July (low response)	Aug.-Sept. & Feb.-Mar. (high response)	Oct.-Jan. (intermediate response)
Jersey	Repeatability	0.93**	0.72**	0.60**
	No. of animals	23	22	20
	No. of tests	60	53	48
Sindhi-Jersey (F1) crosses	Repeatability	0.54**	0.67**	0.78**
	No. of animals	21	17	18
	No. of tests	48	38	44

** Significant at 0.01 level of probability.
[a] Coefficient of intra-animal correlation *(1, 6)*.
[b] Trapezoidal mean for a period of 6 hours exposure.

of respiration rate in all three categories of animals. Observations made through-
out the period of exposure indicate that the respiration rate, unlike the rectal
temperature, rises rapidly during the first hour and then remains at about the
same level or falls somewhat as exposure proceeds. This is in accordance with
observations previously reported on cows *(5)* and sheep *(4)*. In heifers, both the
mean respiratory rate during exposure and the mean rise in respiratory rate
above the initial value are greater to a highly significant degree in the Jerseys
than in the Sindhi-Jersey crossbred animals. (The lower significance in Table 3

TABLE 3

*Analyses of variance of the initial respiration rate, mean respiration rate on
exposure, and rise in respiration rate with exposure*

		Mean squares		
Source of variation	Degrees of freedom	Initial respiration rate	Mean respiration rate on exposure	Mean rise in respiration rate with exposure
(a) Heifers				
Breeds	1	1,982.829**	16,140.750**	4,766.164**
Ages	8	200.291	299.784	364.463
Interaction	8	246.127	75.969	664.346
Error	258	152.192	822.283	413.380
(b) Lactating cows by months of lactation				
Breeds	1	970.196*	5,802.264**	1,631.130*
Months	3	230.999	390.430	698.430
Interaction	3	93.547	10.584	69.709
Error	130	241.309	199.478	316.618
(c) Dry cows				
Breeds	1	535.759*	1,041.460	363.77
Error	130	119.384	357.52	404.22
(d) Cows by levels of production (excluding dry cows)				
Breeds	1	15.669	3,805.264**	4,776.468**
Levels	3	733.186*	262.580	236.096
Interaction	3	577.953	399.170	48.467
Error	201	231.041	211.85	321.141

* Significant at the 5% level of probability.
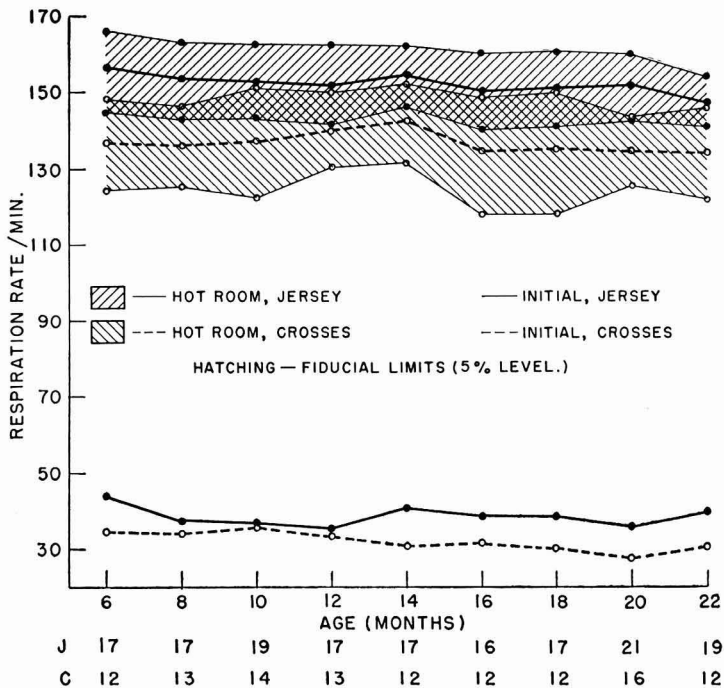** Significant at the 1% level of probability.

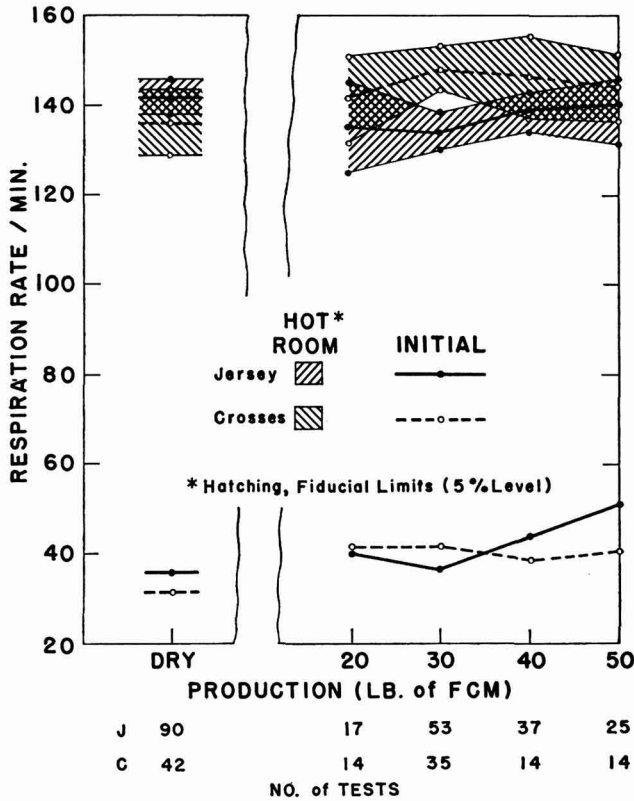FIG. 3. Initial and mean respiratory rates of heifers to a standard hot atmosphere (6 hours at 105° F. with 34 mm. Hg vapor pressure). Numbers at foot indicate the number of animals, Jerseys (J) and crossbreds (C) tested at each age.

for breed differences in cows sorted by stages of lactation is attributed to the smaller number used in this analysis.) In lactating cows, however, the mean respiratory rate during exposure was higher in the crosses.

The initial respiration rate of the Jerseys is the greater to a highly significant degree in heifers and to a significant degree in dry cows, but the difference in lactating cows is obscured by the effects of levels of production. The response to heat is not significantly affected by age in heifers, stage of lactation, or level of production.

<center>DISCUSSION</center>

It is clear that cows of the F_1 cross between Jersey and Red Sindhi show a smaller rise of rectal temperature when exposed to severe heat than do comparable purebred Jerseys. This is true whether the comparison be made as young heifers, older heifers, dry cows, or lactating cows at the same production level or stage of lactation.

It seems reasonable to suppose that all bodily processes, including milk production, will be adversely affected in an animal which shows a marked rise of rectal temperature. But, from a practical point of view, an animal might have such a high productive capacity that it would continue to yield more milk under

Fig. 4. Initial and mean respiratory rates during exposure to a standard hot atmosphere (6 hours at 105° F. with 34 mm. Hg vapor pressure) by average level of milk production on the five days preceding the test. Numbers at foot indicate tests performed on Jerseys (J) and crossbreds (C) at various levels of production.

hot conditions, in spite of the disability, than a more resistant cow with lower productive capacity. The concept of heat tolerance is a complex one, not to be defined in terms of any one response.

The diminution of body temperature response with age is to be expected, since the temperament of the animals becomes more placid and they acquire a certain familiarity with the proceedings. It is possible that the reduction in basal metabolic rate, which commonly occurs during the early life of most animals, plays a part in reducing the response of body temperature to heat, as to other stresses.

The effect of milk production upon the response of body temperature is most interesting. That the response should increase with lactation and that the degree of rise should bear some relation to the level of production are to be expected. The interest lies in the comparative behavior of the two breeds. It is well known that Zebu types have a lower level of production than do the better European

types, and it is sometimes argued that the superior heat tolerance of the Zebu would be nullified if its production were raised to the European level, especially if this increase in production were effected by introduction of production genes from European stock.

It is clear, however, from the curves of Figure 2 that half-breed animals at the same level of production as pure Jerseys still show a greater resistance to heat stress (as judged by rise of body temperature). The greater heat resistance of the Zebu is therefore not entirely due to low production. It will be interesting to see if this superiority is maintained in the F_2 generation and at what combination of European and Zebu "blood" the productive superiority of the one will offset the heat resistance of the other.

The high repeatability of body temperature response to the test conditions, once biassing factors such as season and milk production are excluded, justifies the continuance of present procedures for measuring comparative responses of rectal temperature to heat stress, but caution must be exercised in identifying such responses with the wider concept of "heat tolerance" until more extensive studies have been made.

It is interesting to note that the Sindhi-Jersey crossbred animals not only maintain a lower rectal temperature than the Jerseys but do so with a lower respiration rate. This is in full agreement with the contention argued elsewhere *(2)* that differences in respiratory activity are the result and not the cause of differences in heat tolerance. That age in heifers and level of production in lactating animals should not have any effect on respiration, although they do affect body temperature response, is not surprising when one realizes that most animals are near the ceiling of their respiratory effort under the test conditions used. A decrease in respiratory effort could hardly be expected until a substantial reduction in body temperature had been attained.

SUMMARY

Jersey and Red Sindhi-Jersey (F_1) crossbred cows are subjected at regular intervals to a standard test atmosphere at 105° F. with a vapor pressure of 34 mm. Hg (wet bulb 92° F.). The mean response of rectal temperature is less in the crossbred animals than in the Jerseys, whether the comparison be made as young heifers, older heifers, dry cows, or lactating cows. The difference occurs also when comparisons are made between animals at the same level of production.

In heifers, both initial temperature and temperature during exposure are higher in the younger animals, but the rise is similar at all ages. Dry cows show much the same reactions as older heifers. The stage of lactation has little effect on the response of rectal temperature to heat, but the level of production has considerable effect. The rectal temperature during exposure tends to increase fairly steadily with production in Jerseys, but in F_1 crossbreds it quickly reaches a level which does not rise with further increases in production.

There is marked seasonal variation in body temperature response. Repeatability of response is high in heifers when results are sorted by season, but low in lactating cows.

In heifers the respiratory response of the Jerseys to heat is greater than that of the Sindhi-Jersey crossbred animals, but as lactating cows the reverse was found. No difference was seen in dry cows. The contention is confirmed that differences in respiratory rate are the result and not the cause of differences in heat tolerance.

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TECHNICAL NOTES

THE EFFECT OF HEATED FORTIFIED SKIMMILKS UPON THE LIVABILITY OF BOVINE SPERMATOZOA[1,2]

Previous studies at this laboratory (1, 2) have shown that heated fresh pasteurized skimmilk supports satisfactory livability and fertility of bovine spermatozoa. However, a problem in its use has resulted from a relatively new practice in which many creameries fortify skimmilk in preparing it for human consumption. Condensed skimmilk, superheated condensed skimmilk, or nonfat dry milk solids may be added to fresh skimmilk in order to produce a more nutritive, more palatable beverage. Preliminary studies indicated that such practices were undesirable when the skimmilk was to be used as a semen diluent. An experiment was conducted to determine the effect of fortification of skimmilk upon the livability of bovine spermatozoa.

The following milks and combinations of milks were tested: raw skimmilk, condensed skimmilk, superheated condensed skimmilk, raw skimmilk plus condensed skimmilk, and raw skimmilk plus superheated condensed skimmilk. Each of these products was tested at 9.0 and 11.0% nonfat solids. These levels approximate the percentages of nonfat solids usually found in raw or pasteurized skimmilk and fortified skimmilk, respectively.

Raw skimmilk, condensed skimmilk, and superheated condensed skimmilk were obtained

from the University Creamery. The per cent nonfat solids content of each milk was determined by the Mojonnier method. The raw skimmilk contained 9.0% nonfat solids, and the condensed skimmilk and superheated condensed skimmilk contained 29.4 and 27.4% solids, respectively. The fortified skimmilk products were prepared by adding the condensed skimmilks to raw skimmilk in sufficient quantity to raise the nonfat solid content of the raw skimmilk from 9.0 to 11.0%. Aliquots of these products were reduced to 9.0% solids by adding double distilled water; thus, an equal ratio of the constituents was maintained at both solid levels. Condensed skimmilk and superheated condensed skimmilk were reduced to 11.0 and 9.0% nonfat solids by the addition of double distilled water. Unaltered raw skimmilk containing 9.0% nonfat solids served as a control. Raw skimmilk was adjusted to 11.0% solids by adding nonfat dry milk solids.

The diluents were heated to 92° C. and held for 10 minutes. Upon cooling, 1,000 units each of penicillin and streptomycin were added per milliliter of diluent. The concentration of spermatozoa was standardized at 15×10^6 motile spermatozoa per milliliter and the diluted semen was stored for 14 days at 5° C. At the end of the 14-day storage period, the freezing point depression of each diluent was determined by use of a Hortvet cryoscope. Samples used in determining the freezing point depression were taken after the diluted ejaculates were pooled.

The mean livability of spermatozoa during 14 days of storage at 5° C. is presented in Table 1. Each figure represents the mean of seven ejaculates. Analysis of variance showed

TABLE 1

Livability of spermatozoa in various skimmilk products adjusted to 9 and 11% nonfat solids and heated to 92° C. for 10 minutes (mean of 7 ejaculates)

Source of diluter	Nonfat solids	% motile spermatozoa after storage at 5° C. for								
		1 day	2 days	3 days	4 days	6 days	8 days	10 days	12 days	14 days
	(%)									
Raw skimmilk	9	64	61	56	46	36	29	16	10	7
	11[a]	59	51	44	34	24	11	4	0	0
Condensed skimmilk	9	61	59	59	53	36	29	21	13	10
	11	33	23	17	14	4	0	0		
Superheated condensed skimmilk	9	63	53	53	43	30	21	14	3	1
	11	34	26	21	14	6	3	1	0	0
Raw skimmilk plus condensed skimmilk	9	64	60	56	47	36	29	21	11	7
	11	49	34	30	23	17	6	1	0	0
Raw skimmilk plus superheated condensed skimmilk	9	63	57	57	33	37	29	23	10	10
	11	51	46	36	33	20	7	3	0	0

[a] Raw skimmilk plus nonfat dry milk solids.

that increasing the nonfat solids of skimmilk from 9.0 to 11.0% significantly decreased spermatozoan livability (P < 0.01). The differences in livability among diluters were not significant; however, a highly significant diluter-solids level interaction was obtained. Thus, the various diluters responded differently to an equal increase in nonfat solids. In all cases the addition of distilled water to reduce the nonfat solids content from 11.0 to 9.0% resulted in diluents capable of supporting optimum spermatozoan livability.

TABLE 2

The relation of the freezing point depression of various skimmilk diluents to spermatozoan livability (mean of 7 ejaculates)

Diluter[a]	% nonfat solids	Freezing point depression	Mean % motile spermatozoa during 14 days of storage at 5° C.
B	9	-0.592	37.8
A + B	9	-0.583	36.8
A	9	-0.569	36.0
A + C	9	-0.572	35.4
C	9	-0.643	31.3
A + D	11	-0.673	25.4
A + C	11	-0.713	21.7
A + B	11	-0.723	17.8
C	11	-0.793	11.7
B	11	-0.787	10.2

[a] Key: A = Raw skimmilk
B = Condensed skimmilk
C = Superheated condensed skimmilk
D = Nonfat dry milk solids

Variation in spermatozoan survival among the skimmilks adjusted to 11.0% nonfat solids and between the 9.0 and 11.0% skimmilks can be explained in part by differences in the osmotic pressure of the fortified and unfortified products as measured by the freezing point depression (Table 2). A highly significant negative correlation (r = -0.986) was obtained between the freezing point depression of the diluents tested and the mean per cent motility of spermatozoa during 14 days of storage. This suggests that the harmful effect of fortified skimmilk upon spermatozoan livability results from a substance in milk which becomes toxic at elevated osmotic pressure levels.

On the basis of this study, the use of heated fortified skimmilk cannot be recommended as a satisfactory substitute for unaltered fresh milks for diluting bovine spermatozoa unless sufficient quantities of water are added to reduce the nonfat solids content to that of normal skimmilk.

R. G. SAACKE
J. O. ALMQUIST
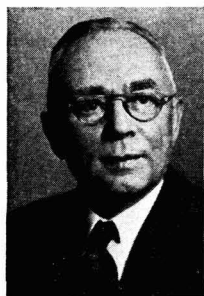S. PATTON
The Pennsylvania State University

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Pioneers in the Dairy Industry

On April 28, 1877, a child was born on a dairy farm in Story County, Iowa, who was destined to pioneer important activities in the dairy field. Among them was his success in persuading the young American Dairy Science Assoc. to publish a journal. After considerable skepticism and some real opposition, the members voted in 1916 to adopt a committee report favoring such a publication, and the man who sponsored the idea, JULIUS HERMAN FRANDSEN, was appointed to be the first editor, a position he held until 1928.

Young Frandsen's early education in a rural school was followed by training at Iowa State College, from which he received the B.S. degree in 1902 and the M.S. degree in 1904. After graduating he served as agricultural chemist at his Alma Mater and later worked for 3 years as a chemist in a commercial plant in Portland, Oregon.

J. H. Frandsen

His real opportunity for a career in the field of dairy education came in 1907, when he was appointed to organize a department of dairying at the University of Idaho. Four years later he became professor of dairy husbandry at the University of Nebraska, where he established a strong department. In 1921 he moved to the field of journalism and for 5 years served as dairy editor of the Capper publications, a group of seven farm papers. In 1926 he returned to educational work and accepted the headship of the Dairy Industry Department at the Massachusetts Agricultural College, now University of Massachusetts, where he remained until his retirement in 1947.

Professor Frandsen has been active in many fields. During World War I he served on President Hoover's Food Administration Committee; he was chairman of the Committee on Dairy Publications for the World's Dairy Congress held in Washington, D. C., in 1923; and in 1931 he was the Massachusetts delegate to the World's Dairy Congress in Denmark. He has been a member of the American Dairy Science Association since 1908. In 1940 the Association presented him with its distinguished service award and a life membership. Professor Frand-

sen has also been given special citations for meritorious contributions to a number of other organizations of which he is a member, including the Massachusetts Milk Inspectors Assoc., the New England Ice Cream Manufacturers Assoc., and the Iowa State Alumni Assoc.

He has been a prolific writer for professional journals on such subjects as the nutritive value of milk products, the importance of disease control in dairy cows, merits of milk pasteurization, and the manufacture of ice cream. In 1915 he collaborated with E. A. MARKHAM to author the first text book on ice cream manufacture. In 1951 he and the late D. H. NELSON published the book, *Ice Cream and Other Frozen Desserts.*

In 1906 Professor Frandsen was married to Matilda C. Madsen, an Iowa State College graduate, who has always shared her husband's interest in educational and civic activities. The Frandsens have three children, Julius H., Jr., general news editor for the United Press in Washington, D. C.; Dana C., in government service at Washington; and Mrs. Dorothy E. Helming of Amherst.

Professor and Mrs. Frandsen live at 92 High Street, Amherst, where they remain active in church and civic affairs. Their hobbies are gardening and photography. Their winters are spent in Florida. As a true pioneer should, Professor Frandsen maintains an active interest in the American Dairy Science Association and in the field of dairy technology through his writings and service as a consultant.

Reid to Do Research at Reading

J. T. REID, professor of animal husbandry at Cornell Univ., has been awarded a Guggenheim Fellowship and a Cornell Univ. College of Agriculture Traveling Fellowship to study abroad on the problem of energy metabolism of cattle. He will spend most of his time at the National Institute for Research in Dairying at the Univ. of Reading, which is considered one of the most outstanding centers of animal nutrition and physiology in Europe. Dr. Reid's research problem will concern the development and application of indirect methods for the measurement of the body composition of live cattle.

Dr. Reid will be accompanied by his wife and five children. They expect to visit Scotland, Wales, France, Denmark, Norway, Sweden, and Holland and will return to the states in July, 1956.

Production of "Mellorine Type" Frozen Desserts

The 1954 production of Mellorine and "Mellorine type" frozen desserts totaled 31,416,000 gal., the Agricultural Marketing Service reports. This is 30% greater than the revised production of 24,207,000 gal. for the preceding year. Mellorine is, as explained in reports for 1953 and 1952, made with fats other than butterfat. The legal and most commonly known name for this product in most states is "Mellorine," and that designation is used in this report.

There were 10 states in which Mellorine was made in 1954, compared with nine for 1953, the additional state in 1954 being Nevada. The production of Mellorine began in Alabama, Arkansas, California, Montana, and Oregon in 1953, and in 1952 or earlier in Texas, Missouri, Oklahoma, and Illinois. It is anticipated that Mellorine will be made in at least one additional state in 1955.

Texas, with 17,635,000 gal., is the leading state in its manufacture and accounted for 56% of the total production in the United States during 1954. Illinois is next in production with 4,681,000 gal., followed by Missouri with 3,017,000 gal.; Oklahoma, 2,927,000 gal.; California, 2,260,000 gal.; Arkansas, 325,000 gal.; Oregon, 295,000 gal.; Alabama, 144,000 gal.; and Montana, 124,000 gal.

Monterey Cheese Definition Amended

The Food and Drug Administration has been petitioned by the California Cheese Assoc. to amend the definition and standard of identity for Monterey cheese. The proposal is that the section title that prescribes the name of the cheese be amended to read "Monterey cheese, Monterey jack cheese," thus making "Monterey jack cheese" a synonym for "Monterey cheese" for the purpose of compliance with requirements of the Federal Food, Drug, and Cosmetic Act.

Kaufmann Joins Staff at Michigan State

O. W. KAUFMANN, assistant professor of food microbiology at the Univ. of Illinois, has accepted a position in the Department of Microbiology and Public Health at Michigan State Univ. Kaufmann, a graduate of the Univ. of Connecticut (B.S. and M.S.), obtained his Ph.D. degree at Purdue Univ. in bacteriology. He joined the Illinois staff in 1950. At Michigan Dr. Kaufmann will have charge of the course work and research in dairy bacteriology.

Dates for Michigan Dairy Conference Announced

The 16th Annual Michigan Dairy Mnfrs. Conference will be held Nov. 2 and 3, 1955, at the Kellogg Center, Michigan State College. Processors of three specialized groups of prod-ucts—market milk; ice cream; and butter, cheese and dried milk—will meet concurrently for the 2-day session. An instructional clinic for nonfat dry milk, ice cream, and market milk will be conducted on the afternoon of Nov. 2.

7th International Grassland Congress

The 7th Intern. Grassland Congress is being held in New Zealand Nov. 6-15, 1956. Anyone interested in attending this congress should notify the Organizing Secretary, CPO Box 1500, Wellington C1, New Zealand, by Nov. 1, 1955. The aim of the congress is to bring together as many as possible of the world's leaders in grassland work and to give them an opportunity to exchange information and ideas on grassland production and problems. The congress is being sponsored by the New Zealand government under the auspices of the New Zealand Grassland Assoc., which has set up an organizing committee to make all the arrangements. The 6th Congress was held in the United States in 1952.

The Congress proper will be held at the Massey Agricultural College. Palmerton North. The College farms are devoted to dairying on the river flats and to sheep and cattle raising on the terraces and hill country. Most types of North Island grassland farming are practiced at or near the College. Adjoining the College are the Dairy Research Institute and the Grassland Division of the Department of Scientific and Industrial Research, which is the major grassland research station in New Zealand.

The papers at the Congress will be at a specialist level and will be presented by persons actively engaged in various aspects of grassland research. The program provides for 13 half-day plenary sessions, with the following titles:

Macro—and micro—climatic factors in pasture production.
Environment and plant improvement.
The pasture grazing animal complex.
The digestion of pasture herbage by the ruminant animal.
Macro—and micro—elements in plant nutrition.
Pasture plant breeding.
Pasture utilization, including conservation
Pasture establishment and management.
Special methods in plant breeding.
Legumes and the nitrogen cycle.
Micro—element deficiency in grazing animals.
Pasture development in difficult environments.
Improvement and management of tropical grassland.

The official language of the Congress will be English. There will be two categories of participants:

Official delegates: official representatives of governments.

Members: scientists and others interested in the establishment, improvement, and utilization of grassland.

Keeney Joins Penn State Staff

P. G. KEENEY accepted an appointment as assistant professor in the Dept. of Dairy Science at Penn State, effective July 1. After 3 years in military service during World War II, Dr. Keeney completed his B.S. degree in dairy manufacturing at the Univ. of Nebraska. He spent 2 years in industry in Minnesota and then did work in dairy technology at Ohio State Univ. for his M.S. degree and at Penn State for his Ph.D. degree. Dr. Keeney comes from a well established dairy family. His father, the late MARK H. KEENEY, a graduate of Penn State in 1915, made a national reputation as a breeder of Holstein cattle. His brother, MARK, is associate professor of dairy husbandry at the Univ. of Maryland, and his brother, DAVIS, is chief chemist for Hauley & Hoops Candy Co. at Newark, N. J. Professor Keeney fills the vacancy left by the retirement of C. D. DAHLE.

Ohio News

A dairy technology student-faculty mixer will be held early in October. This get-together is planned to allow the older students and faculty to entertain all new students at an informal "feed" and to acquaint them with the dairy technology student activities.

An event of Oct. 8 is the employer-employee program. On this day the students entertain their summer employers at a luncheon and attend the Illinois–Ohio State football game.

The 7th Annual Homecoming will be held on Oct. 29 on the Ohio State campus. About 200 alumni, wives, and friends will talk over old times, enjoy a short program, and then watch the Ohio State–Northwestern football game.

The Dept. of Dairy Technology has completed its plans for adult education for the academic year 1955-1956. This includes the 2-week Market Milk and Ice Cream Short Courses to be held on Nov. 7-18 and Jan. 9-20, respectively. These courses are designed for those already employed in the dairy industry who desire specialized training.

Plans for the 23rd Annual Dairy Technology Conference have been announced for Feb. 7-10, 1956. This conference is designed for those who are advanced in the field of dairy processing and covers a wide variety of subjects. Outstanding speakers will bring the latest information to those in attendance.

The 1-week Milk Sanitarians' Short Course will be held March 12-16, 1956. This short course is a cooperative effort between the Ohio Dept. of Health and the Dept. of Dairy Technology and is a basic training course in milk regulations and enforcement for persons employed by public health and regulatory agencies and by dairy plants.

J. H. ERB, vice-president in charge of production of the Borden Co. Midwest Division, has been named chairman of the Education Sub-Committee of the Dairy Industry Committee. This committee concerns itself with matters pertaining to recruiting, training, and developing leaders for the industry.

The 18th annual Ohio State Univ. Milk Marketing Conference has been announced by the Dept. of Agricultural Economics for Sept. 20-21. Topics to be discussed include effect of prices and methods of distribution on consumption, midwestern attitudes on Federal Marketing Orders, and farm bulk tank programs.

Ohio's legislature has had one of the busiest sessions in its history. It considered 14 dairy bills. A law was passed that legalized refrigerated bulk milk dispensers; however, the milk sold must be pasteurized and homogenized. The glass used to serve the milk must be of a 9-oz. size, and the dispensers must not be loaned to restaurants or other outlets for competitive advantage. Another bill which passed into law provided that milk bottles must be filled to within $\frac{1}{4}$ in. of the cap seat instead of $\frac{1}{8}$ in. as previously prescribed.

The Milk Control Commission Bill, one of the most controversial bills introduced this season, did not pass. Other dairy bills that did not pass include a pure food and drug bill, butterfat content labeling bill, a bill to forbid placing non–ice cream items in ice cream cabinets, legalization of a 1/3 qt. bottle, the redefining of imitation ice cream, state-wide compulsory pasteurization of milk, and a bill to establish minimum sanitary standards.

Announce Official Rules for the National Intercollegiate Dairy Cattle Judging Contest

The Production Section of the American Dairy Science Association 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 to officially designate the rules under which it shall be conducted.

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. No member of any team or their coach shall be allowed in the cattle barns or have the privilege of inspecting any cattle on the grounds previous to the contest.

1. *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.

2. *Reasons.* Contestants shall give oral reasons, two 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 as to 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.

3. *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.

4. *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 consecutive 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 given, 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.

Wisconsin to Hold Dairy Barn Planning Conference

A conference on dairy barn planning will be held at the Univ. of Wisconsin Sept. 29 through Oct. 1, 1955. The program is to include discussions of the following subjects:

The make-up of a well managed herd.
Points in planning new and in remodeling stall and loose-housing systems.
Construction details as related to planning.
C.I.P. pipe-line milking.
Milkhouses for can and bulk cooling.
Materials handling.
Heifer shelters.
Insulation, ventilation, and moisture control in stall barns.
Milking parlors for stall barns and loose-housing systems.
Field trips to stall and loose-housing systems in operation.

Research Award Announced

Competition is now open for the Costantino Gorini Award in Dairy Research. The award, consisting of 100,000 liras (about $160), is given to a candidate who has demonstrated outstanding qualities in research on the production, processing, or uses of milk and dairy products.

Entries should be sent by Dec. 1, 1955, to Instituto Lombardo, Via Brera 28, Milan, Italy. An entry shall consist of a letter of application, supporting reprints of published work, and an account of the research work completed since 1952. The reprints and letter may be written in one of the following laguages: Italian, French, English, Spanish, or German. The account of the work itself must, however, be written in Italian or French.

Industry Promotions

J. A. ROBINS, formerly administrative vice-president of the Stokey–Van Camp Co., Inc., has been made president of Fairmont Foods Co., Omaha, Neb., succeeding D. K. HOWE, who has been made chairman of the board. Mr. Robins was formerly connected with the U. S. Steel Corp. and the General Baking Co. He at one time served as a consultant to the Dept. of Defense in establishing modern business methods and controls for the military.

P. R. OLIVER, former president of American Dairies, which at present is owned by Foremost, has been named vice-president of Foremost with headquarters at Kansas City, Mo., according to GROVER TURNBOW, president. Mr. Oliver, a graduate of the Univ. of Arkansas, has served as president of the Missouri Butter and Cheese Institute and has been on the board of the Intern. Assoc. of Ice Cream Mnfrs.

W. E. SNYDER has been named director of quality control for the Chicago-Central fluid milk and ice cream district of the Borden Co., Chicago. Dr. Snyder joined the Borden Co. in Chicago as laboratory director in 1949. Prior to this time he served on the staff at the universities of Wisconsin and Georgia. He took his undergraduate work at Penn. State and his graduate training at Illinois and Wisconsin.

Finnegan Joins Stein Hall

EUGENE FINNEGAN, former associate professor of dairy manufactures at Virginia Polytechnic Inst., has accepted a position with Stein Hall & Co., Inc., New York. In his new post Finnegan will work closely with sales representatives in the field, lending technical assistance on food production problems. He served on the staff at Massachusetts from 1947 to 1953, receiving his B.S. degree in 1941 and his M.S. in 1950 in dairy manufactures.

The Miracle of Milk

In the Our Industry Today section of the current issue of the Journal is an article written by L. L. RUSOFF by invitation of the Editor that should be made available to every person in the United States. It is an authentic and interesting story of how milk and its products down through the ages have been the principal food in the diet of civilized man.

In this article are ample reasons why more people should use more milk, butter, cheese, and dried and evaporated milk. The story is convincing enough to bring about a shortage of milk solids in this country in a few weeks time. Commercial dairy companies should buy hundreds of reprints of this article for distribution to doctors and dentists, to schools, and other groups who should know about the "miracle of milk."

Dr. Rusoff, who is professor of dairy nutrition at Louisiana State University, is not a stranger to readers of this Journal, for he is a prolific writer of research articles. He is to be commended for the excellent job he has done in preparing this interesting story. He has done a splendid service to the industry.

Completed Theses

M.S. Degree:

W. S. GRIFFITH—Thyroidal activity of dairy bulls. Louisiana State Univ.

L. M. UNDERWOOD—A comparison of *Pennisetum glaucum* (Starr) and *Sorghum vulgare* var. *sudanense* (Tift) for temporary grazing for temporary grazing for lactating dairy cows. Univ. of Georgia.

OUR ASSOCIATION

A message from the president

These Goals We Seek

This is the 50th anniversary year of our Association, and it is natural that the officers are hopeful we shall have an exceptionally outstanding year. However, if this is to be the case, it will require, as Kipling says, "the everlasting teamwork of every bloomin' soul."

Each new administration of a government or organization should have a "platform"—a set of objectives which it hopes to achieve—and this applies to us especially this year if we are to make our GOLDEN JUBILEE YEAR truly noteworthy. Therefore, we propose the following as typical "planks" of our 1955-56 platform:

Increase membership: The never-ending task of membership solicitation is still with us—and is to be given extra effort this year. Every progressive organization needs a constant infusion of new blood—with new ideas and greater enthusiasm. A large number of well-qualified persons in our industry are not now members of our Association. Both the Association and they, themselves, will benefit through their membership.

In all membership activities, careful thought is to be given to the Student Affiliates and to the fact that the future of the industry and of our Association depends upon creating and maintaining interest among this group of young people.

Our membership now is about 1,700. What should our goal be this year—2,000? or 3,000? Neither is out of reach.

Educational improvement: The Association is accepting its rightful role from the standpoint of secondary school recruitment activities and the training of young people for leadership in our industry. Perhaps we have been remiss in not giving enough attention at our annual meeting to the matter of improvement in teaching and to designing a portion of the program especially for the large number of our members who are directly and vitally concerned with teaching—either in the classroom or in the field. The Extension Section has been active in this area, and it is now time that the Association as a whole accept its responsibility in this connection. An Association Education Commitee will have something tangible to report at the annual meeting.

In respect to secondary school recruitment activities, a Recruitment Guide, developed by the Public Relations Committee, is being published in the *Journal*. Also, the Association has already signified its desire to coordinate the efforts of all dairy industry groups in the production of suitable "career opportunity" films.

Public relations and publicity: A further intensification of effort in the field of public relations is an important "plank." The continuing Public Relations Committee has a major charge for this year in respect to publicity—to acquaint persons and agencies outside of our Association with the programs of our group, with its aims and purposes, and with the important role it plays in shaping the destiny of our industry. Is it not time that we allow our "candle to burn brightly"—to announce with pride that our Association, now 50 years of age, is the outstanding educational and scientific organization in the dairy industry, and that such recognition is justly deserved? Perhaps we need to realize ourselves that our members are directing the entire dairy educational program in the United States through classroom and adult programs, and that the research of our members is responsible for the present status of our industry and for its future progress.

The complexity of modern industry requires that we work more closely with trade associations—to recognize that the commercial interests are a vital part of *our* Association. We need to dispel all thoughts that we are "stuffy" and "stand-offish." We must accept the fact that no barrier now exists between education and industry and that the day of isolation of the educator in the "ivy-covered tower" is past.

Internal communication: How important it is that the members of our Association be kept informed at all times of the activities within the organization—if for no other reason than that they are made to realize the work load of the committees and the officers of the various sections and of the Association! One only need examine the January issue of the *Journal* last year to appreciate the large number of committees and the large number of persons who are involved in carrying on the work of the Association. Truly it is a democratic organization, where opportunities for self expression are plentiful and where individuals are dedicated to the principle of improving the welfare of all.

It is urgent as we move forward into the next half century to have each member of our Association realize what it takes in the way of manpower and money to run the Association and how important it is that each member of the Association be willing to give to this cause. Money to finance the Association comes from memberships, *Journal* subscriptions and advertising. It is *everybody's* job to help increase the revenue from *all* of these sources.

"Our" Journal: Another goal this year is to encourage to a greater extent the contribution of our members to the *Journal of Dairy Science* and their general support of it in every way. Perhaps this is the year when valuable research findings, now gathering dust in the researcher's files, should be put into manuscript form and submitted to the Editor for review. This is the

year when greater use should be made of the special section which offers opportunities for personal opinions, for comments on technical matters, for feature articles, and for current news items which would be of interest to the membership. This is no easy job—this getting out each month a magazine which carries the recognition and prestige that is carried by our *Journal*—and the *Journal* Editor and staff cannot do it alone. Here again is a job for all of us.

Program planning: To have effective programs for the sections and for the Association at the annual meeting is the earnest desire of the Program Committee, and to achieve this will necessitate a careful scrutiny of all details —of the papers presented, special features, and the business meetings. "Can the number and quality of submitted papers be controlled?" is a burning question which arises each year, and

"How can the business meetings be more interesting to all members?" is another. What are the answers?

Finally, the over-all goal is to make this, the GOLDEN JUBILEE YEAR, a dramatic climax to the 50 years of progress and achievement of the Association since its inception in 1906. Certainly, this year deserves something out of the ordinary from each of us—and if this is done—then the meeting in Connecticut in June will be a fitting kick-off for the next one-half century which the Association faces.

I. A. GOULD
President

LETTERS TO THE EDITOR

C. F. Doane One of the Founders of A.D.S.A.

In a short note that I wrote about PROFESSOR E. S. GUTHRIE as one of the pioneers in the dairy industry, I mentioned a few individuals as being surviving charter members of the American Dairy Science Association. I wish to correct a serious omission.

PROFESSOR C. F. DOANE not only was a charter member but gave a paper entitled "Opportunities for Experimental Work" at the first meeting at Urbana, Ill., in 1906. At that time he was engaged in research work in the Dairy Division of the USDA, after having served a number of years as professor of dairying at the Univ. of Maryland. Few people have worked in as many different phases of dairy research as did Professor Doane during the 20 years or more from about 1900 to the early 1920's. He is probably best remembered for his outstanding investigations on cheese, encompassing the scientific and practical aspects of cheeses of various types. Especially important were his contributions to the science and practice of Swiss cheese production, and the sound foundation he laid for the development of that industry in America.

With the entrance of the United States into the first World War in 1917, Professor Doane entered commercial research and production in California, where he established Swiss cheese factories and also stimulated the production and consumption of other cheeses, especially cottage cheese, the production of which has grown so greatly during the last 30 years in that part of the country.

About 1925, Professor Doane moved to Oregon, where he established a business of his own. Now 83 years of age, he resides with Mrs. Doane at 1510 N. 21st St., Salem, Ore., where they celebrated their 58th wedding anniversary on June 10, 1955.

J. M. SHERMAN

Data May Be Filed in Library of Congress

Occasionally valuable data are presented by authors in such volume as to make publication in a scientific journal impractical. Such data can be preserved and made available for general use through the Library of Congress. In response to a letter of inquiry, JOHN A. HEATH of the ADI Auxiliary Publications Project, Library of Congress, sent me some essential information which should be of interest to contributors to the *Journal of Dairy Science*.

Certain conditions must be met before data can be submitted to and accepted by the ADI Auxiliary Publications Project:

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WALTER V. PRICE
University of Wisconsin

STUDENT CHAPTER NEWS

A section devoted to the activities of dairy students

A Professor Speaks

The dairy industry needs good leadership in its personnel more than any other quality. Specialized training might be considered less important and scholastic standing and college degrees less significant if the industry could be sure it was getting leadership when it employed new personnel.

Leadership is defined as guiding, directing, or being foremost. The degree of leadership possessed by an individual is determined by heredity, environment, and experience. An individual can do nothing about his heredity but he can do something about his environment and his experience. For example, a student changes his environment markedly when he leaves home to attend college. The extracurricular activities he gets into while in college are largely determined by his interest and effort.

The leader has characteristics that are frequently just the opposite of the nonleader, who may do his job well but seldom goes beyond its immediate requirements.

Leadership training will start the moment a student joins the dairy club and accepts the responsibilities that accompany membership in this organization. The first real test probably will come when a student is appointed to serve on a committee or is given a special assignment. The student who accepts responsibility, explores all possible action on the assignment, and turns in a job well done, will have a good start on the road to leadership.

One does not have to be president to be a leader! Presidents are frequently elected to office as a result of a popularity contest or political coalition. They may or may not be leaders. The Dairy Club stands ready to help all students develop their ability to be leaders.

W. L. SLATTER
The Ohio State University

CHARACTERISTICS OF

The leader	*The nonleader*
Accepts and fulfills obligations to his industry, organizations, and community at large.	Evades obligations whenever possible. Never contributes more than the minimum required.
Thinks of his obligations to his company; is willing to use his initiative in getting the job well done. Puts in extra hours when necessary.	Frequently thinks of the company's obligation to him in terms of 8-hour day, 40-hour week, and vacations with pay.
Accepts responsibility; although he may be afraid of making mistakes, he is so busy trying to do the job he soon learns skills and becomes superior.	Shuns responsibility; being afraid of making mistakes and embarrassing himself, he risks no action.
Does things beyond the line of duty.	May think of giving extra service but seldom does so.

The Miracle of Milk

An important message for people of all ages

L. L. RUSOFF

Dairy Department, Louisiana State University

Centuries ago alchemists in laboratories in many lands secretly tried to discover the "elixir of life." This drink or concoction would make it possible for man to have good health, long life, and perpetual youth and would enable him to perform miraculous feats, such as the prevention and cure of disease. There were those who would have paid fabulous sums for its discovery.

The early Spanish seafarers and explorers searched for this "fountain of youth" in the new world of the Americas. It took many years for man to realize that the elixir of life was close at hand, for cow's milk, recognized all over the world as "nature's most nearly perfect food," has been performing the feats that were expected of the mythical elixir of life. Within the last quarter century science has been peering into a drop of milk and has discovered the "miracle"—for no other food in the world can compare with milk in its outstanding nutritive values.

At present, milk and its products are a daily requirement for the populations in most parts of the world. From the equator, where the Arabs still use camel's milk, to the far North, where the Eskimos and Laplanders use reindeer and caribou milk, this product is the number one food item in the human diet. Although the cow and goat have been bred for milk production and are best adapted as a source of milk for man, other domestic animals are used for this purpose, including the water buffalo and zebus in India and central Asia, the yak in Asia, the llama in South America, the sheep in Asia and Europe, and the mare in Asia.

The milk of these different species contains the same constituents but varies in composition and properties (Table 2). The cow, however, supplies the largest proportion of the milk used by humans. In 1953 it was reported that in 15 specified leading dairy countries approxi-

Reprints of this article for popular distribution will be available from the editor until Nov. 1 at a price of $9.00 per hundred copies.

mately 69 million cows were used for milk production. They produced approximately 335 billion pounds of milk (*37*). Surplus milk products are shipped in immense quantities from home countries for international trade. It is estimated that more than 700 million pounds of cheese a year, 700 million pounds of butter, 500 million pounds of condensed milk and millions of pounds of milk chocolate, milk sugar, casein, and other such products are shipped and exchanged from country to country to maintain the world food supply. The United States produces approximately 37% of the world's milk supply, and dairy products are the largest source of our agricultural income. In 1953 it was estimated that the value of milk and its products totaled $4,370,425,000, or 19% of the gross farm national income. About 46% of the milk used in this country is consumed as fluid milk and cream, 26.7% as butter, and 10.6% as cheese (*37*). How did this great industry come about? When and where did man first begin to use milk and milk products?

Historical Background

The story of milk through the ages is fascinating reading. Just when man began domesticating animals (cattle, sheep, goats, horses) is not definitely known. However, it was a very long time ago, possibly between 8,000 and 5,000 B.C., somewhere in Asia or northeast Africa (*1, 24, 40*).

Thus, the use of milk has been with us even prior to the beginning of civilization. The oldest known civilizations have been revealed by excavations. A mosaic frieze on one of the oldest buildings unearthed at al'Ubaid near Babylon in the Euphrates Valley depicts what is considered to be one of the oldest records of the use of milk. The joint expedition of the British Museum and the Museum of the University of Pennsylvania in 1922 estimated this temple at Ur to be at least 5,000 years old (3,100 B.C.). Woolley, leader of the expedition, in describing the scene states, "On a panel, on one side of a reed built byre, from the door of which two

calves are seen issuing, men seated on low stools are milking cattle; the man sits under the cow's tail milking her from behind; on the other side of the byre two men, clean shaven and wearing the fleece petticoats which in later times seem to survive as the official dress of priests and priest-kings, are pouring milk through a strainer into a vessel set on the ground, while two others are collecting the strained liquid in great stone jars" (68).

Some of the oldest written books are those of the Bible, which is considered by Jewish scholars to be 5,715 years old. The custom of reckoning from creation has been calculated to have taken place in the fall of 3,761 B.C. (21, 63). In the Bible, milk and its products were regarded as highly desirable foods and have been mentioned at least 50 times (14, 63). Palestine is praised approximately 20 times as a "land flowing with milk and honey," for example, in Exodus, 3:8, 3:17, 13:5, 33:3; Leviticus 20:24; Jeremiah, 11:5, 32:22; Ezekiel, 20:6, 20:15, and in ten other places, milk representing the common necessities of life and fertility. Cheese or curdled milk called "laban" was so relished as a food that Abraham offered it to his guests in Genesis 18:8; in I Samuel 17:18, David carried cheese to a captain of thousands, among whom were his brothers. Deborah refers to milk as "a cup of the nobles" in Judges, 5:25, and the abundance which the Israelites will enjoy in Messianic times is pictured in the figure, "the hills of Palestine will flow with milk." There are references to a substance called "hemah" in Isaiah, 49:12 and Job, 20:17; this may be either butter or curds, which were regarded as delicacies, and in Genesis, 19:12, milk was supposed to give whiteness to the teeth.

The Vedic hymns of India, written about 2,000-3,000 B.C., are also among our oldest writings. These stories concerning the Hindu people of prehistoric times indicate that milk and butter were used extensively in the diet at that time (51). The butter was changed into ghee (butter oil) which is still used extensively by the peoples of India and Pakistan. Milk was treated also with the greatest reverence and ceremony as it still is in some countries (15).

Around 2,000-3,000 B.C. various other civilizations—the Egyptian, Greek, and Roman—left many records to indicate that milk, cheese, and butter were commonly used (2). Milk played an important part in sacrificial services and was also considered to have great medicinal value (15).

About 500 B.C. in the eastern countries of Europe, Herodotus wrote that the Libyans and Tartars used mare's milk extensively. Writing about the Auschisae, he states: "They hunt locusts and when taken they are dried in the sun, and after grinding sprinkled them into milk and drank it" (10).

About the middle of the 13th century Marco Polo traveled over central Asia and reported that dairy products and milk, "airan" or cur-

dled milk, butter, and "kumiss," which is fermented mare's milk (milk wine), were much enjoyed. He stated that the Tartars were capable of much endurance and when necessary could remain a month without any food except the milk of a mare and the flesh of animals killed in hunting. He also reported that milk was dried into a kind of paste which, when about to be used, was stirred until it became a liquid and could be drunk (35).

In the 15th century, India used large amounts of dairy products. Not only the people but also hundreds of palace elephants had a diet of "rice and butter," or "milk and vegetables," or "meat, rice, fresh milk, and cheese" (44).

In Caesar's Commentaries the Germans are described as a people who did not practice agriculture; however, the major part of their diet consisted of milk and cheese (4). Cheese was also a regular part of the ration of the Roman army. The oarsmen in ancient vessels and sailors subsisted on cheese.

In the northern countries it was early found that butter would keep for a long time. It is stated that "in early times in Iceland such a quantity of butter was made that having neither earthen vessels nor casks to hold it, fir chests were constructed 30 ft. long and 5 ft. square which were filled every year with salt butter and buried in the ground where it was left until needed" (34).

Thus, it is known that the great food value of milk and its products has been recognized through the centuries.

Milk in Culture and Religion

It has been recorded that people who had practically no domestic animals were in the lowest stages of savage culture. In two continents, America and Australia, prior to the domestication of milk-yielding animals the human mother had to suckle her babies for two or three years or until they were able to walk and partially take care of themselves. This burden along with the other chores which the mother had to perform consequently retarded the growth of population (28). The failure of the Indians in both North and South America to find many animals which they could domesticate was probably one of the reasons they did not develop a high civilization. In contrast, the people inhabiting the European, Asiatic, and African continents, who have always possessed milk-producing animals, early developed flourishing civilizations (15).

In primitive religions, food was considered sacred, especially milk, which had an important part in religious ceremonials at nearly all stages of man's development. The sanctity of the dairy is the chief element in the religion of the Toda tribes of India, and here is where the use of milk in religion reached its climax (15). In the Christian church milk was replaced by wine in the communion service. This was afterwards prohibited by canon law. In the early Christian

church the newly baptized were given milk and honey to taste as symbolizing their regeneration through baptism. This is believed to have been one of the surviving rites of ancient pagan religion (15).

Milk and Health

History has shown that the peoples who have subsisted on diets containing a large proportion of milk and its products were unusually healthy, vigorous, and well developed. Milk users were reported as stronger and longer-lived.

Three separate and different human culture areas have evolved in which the people lived almost exclusively on milk and its products. These are the wiry, fierce Kazak Kirgliz (the Tartars) of Central Asia, the strong, lean Bedouins of Arabia, and the sturdy Bantu tribes (Kaffirs) of South Africa. These peoples, located in different world areas, are racially mixed —Mongolian, White, and Negro, but all use milk, curdled milk, cheese, and fermented milks in large amounts (1, 15, 35).

The pastoral peoples of northern India, who have dairy animals, have been found to be superior in health and strength to those of southern India (30). These "stalwart resolute races of the north of India" subsist on a diet of milk and vegetables, but the "toneless, supine, and poorly developed people" of the south and east live only on cereals (29).

In a study of the health and physique of two tribes, the Masai and Akikuya, living in Kenya, Africa, it was found that the full-grown Masai male was on the average five inches taller and 23 pounds heavier than the full-grown male Kikuyu and had 50 per cent greater muscular strength. This difference was attributed for the most part to their diet—for the weaker, poorly developed Kikuyu tribe subsisted mainly on cereals, roots, and fruit, whereas the diet of the stronger and healthier Masai tribe consisted of milk and meat (47).

Dr. E. V. McCollum of Johns Hopkins University, eminent scientist in the field of nutrition, states, "The people who have achieved, who have become large, strong, vigorous people, who have reduced infant mortality, who have the best trades in the world, who have an appreciation for art, literature, and music, who are progressive in science and every activity of the human intellect are the people who have used liberal amounts of milk and its products" (31).

Thus, the dominant and aggressive peoples of the world have always been those whose nutrition has been the best and has contained milk products. Scientific findings are now giving the reasons why milk is healthful.

Proverbial Phrases

Proverbial phrases in which milk is mentioned are many and are still quoted. From the Bible, "land flowing with milk and honey," denoting an abundance of means, of enjoy-
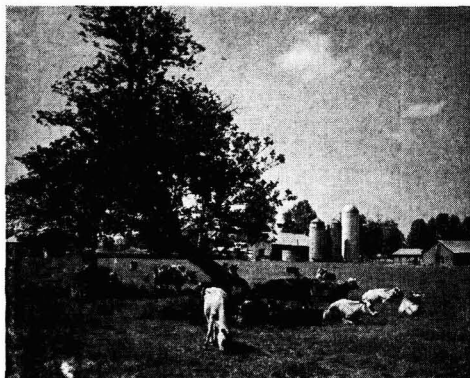

FIG. 1. From herds such as this came the 123.5 billion pounds of milk produced in this country in 1954.

ment; "milk and roses," said of a beautiful pink and white complexion; "milk of human kindness," from Shakespeare, concerning compassion characteristic of human kindness; "spilt milk," anything once misused cannot be recovered; and the "milky way," the galaxy, a way brilliant in appearance or a way leading to heaven (9, 65).

Milk in the New World

There were no cattle in the New World at the time of the discovery of America, for the Indians had no domesticated animals. In his journal Columbus reports, "It was a wonderful thing to see . . . land fit for cattle though they have none . . ." On Columbus' first voyage to America he carried no cattle (27); a certain amount of livestock including four calves and two heifers was taken by him on his second trip, but these were taken to Cuba or South America (19).

Although several Spanish expeditions were conducted to the New World after the discovery of America, that of Coronado, 1540-1542, was the only one that brought cattle to this country; these can be traced as far as Kansas. Part of the expedition's food supply consisted of 150 cattle, from which the present cattle of the Southwest probably originated (46).

The first dairy cattle were brought to the United States during the years 1600-1700 during the colonization of America. These cattle, which served as the foundation of our great dairy industry, represented various breeds and were brought from England, France, the Netherlands, Sweden, and Denmark.

1611—Jamestown Colony. In 1611 the first colonists at Jamestown imported cattle and "at Christmas time, 1618, there were about 300 in the colony" (56).

1620—Plymouth Colony. The Pilgrims did not bring any cattle with them on the first voyage of the Mayflower. Of the 100 men in the Plymouth colony, more than half died that first

winter from malnutrition *(13)*. For three years the colony existed without cattle, and the colonists had a difficult time. In 1624 "the good ship Charity" landed three Devon cows and one bull; after that, severe malnutrition difficulties were greatly reduced.

1624—New Netherlands. In 1624 a group of 30 Dutch families came to this country on the ship "New Netherland." A fort was built at Nut (Governor's) Island. A few months later, 42 more immigrants arrived with 103 head of livestock. In July, 1625, the settlement moved from Nut Island to New Amsterdam (Manhattan, New York). The production of livestock was both profitable and extensive, the black and white Holstein cattle furnishing milk, butter, cheese, meat, and hides. A New Netherland document of 1649 states, "A little hay thrown occasionally to the milk cows is sufficient" *(7)*. New York state was a land ideally suited for raising cattle, and today, New York ranks second among the states in milk production. Wisconsin is first *(37)*.

1629—Massachusetts Bay Colony. In an account of the voyage to Massachusetts Bay Colony in New England in 1629, it is reported that "The Company of New England" sent five ships for further settling of the English plantation. On one of them, the George, "her chief carriage were cattell, 12 mares, 30 kyne (cows), and some goates" *(16)*. Other stock were sent in following years. In 1647 Delaware brought cattle from Sweden, and "large yellow" Danish cattle were imported into New Hampshire *(13, 16)*.

As the early pioneers moved westward they took cattle with them, establishing the dairy cow in all sections of the country. Today, after three centuries, approximately 38 million cattle comprising the major dairy breeds (Jersey, Holstein, Guernsey, Ayrshire, and Brown Swiss) are being used for dairy purposes. This number includes about 25 million cows and heifers, two years old and older; about 13 million younger heifers and heifer calves; and about one million bulls and bull calves *(41)*. Approximately 121 billion pounds of milk were produced in 1953 *(37)*.

How Did Milk Get Its Name?

Since the oldest written records of the human race are in Sanskrit, the word "milk" is derived from the Sanskrit word "mrjati," which means "he strokes," or "he rubs," like the stroking action employed in milking. Our present word milk is derived from the Teutonic words "melki" or "melchan," meaning "to milk" *(54)*. Milk is defined in most dictionaries as a white fluid secreted by female mammals for feeding their young *(63)*.

Phenomena of Milk Secretion

Milk is made from the materials of the blood. It has been estimated that blood makes a complete cycle from the udder to the heart and return in 52 seconds. In a 1,000 lb. cow, approximately 200 lb. of blood pass through the udder in one hour. It requires 150 to 500 lb. of blood for every pound of milk secreted *(6)*. A cow producing 50 lb. of milk per day would require about 16,000 lb., or 8 tons, of blood to be pumped through the udder.

The cells in the mammary gland or secreting tissue in the udder take chemical compounds from the blood for the synthesis of milk. These compounds or their precursors must be present in the feed if they are to get into the blood; therefore, sound nutritional feeding of the dairy cow is important for milk production.

The cow's udder is divided into four quarters, and arteries carry the blood to each quarter. These blood vessels end as small capillaries that surround the milk-making bulbs, or alveoli, like a plastic cover over a cheese. These alveoli look like bunches of grapes and are really tiny udders. Each of these is made up of tiny microscopic milk synthesizing cells. Blood constituents pass from the capillaries to the cells and what is not used for manufacturing milk goes back to the blood circulation. Each alveolus or "grape" produces a fraction of a drop of milk daily.

On comparing the constituents in blood and milk it will be found that milk contains 90 times as much sugar, 13 times as much calcium, 10 times as much phosphorus, nine times as much lipids (fat), five times as much potassium, one-half as much protein and one-seventh as much sodium as blood plasma *(6)*. The milk from millions of these alveoli make up the daily milk production of a cow. The milk manufactured in the cells is deposited in a hollow space in the center of the alveolus, where it is stored until milking time. Milk from each alveolus empties into a small tube, then into larger tubes that empty into the gland cistern, which holds

TABLE 1
Average composition of cow's milk (26, 49)

Constituent	Per cent	
Water	86.90	
Milk fat or butterfat	4.00	Dry matter or total solids
Protein	3.50	
Lactose or milk sugar	4.90	Nonfat milk solids
Ash or minerals	0.70	
	100.00	

TABLE 2
Average composition of milk of various mammals (49)

Species	Fat	Protein	Lactose	Ash	Total solids
	(%)	(%)	(%)	(%)	(%)
Human	3.70	1.63	6.98	0.21	12.57
Cow	4.00	3.50	4.90	0.70	13.10
Goat	4.09	3.71	4.20	0.78	12.86
Ass	1.50	2.10	6.40	0.30	10.30
Mare	1.59	2.69	6.14	0.51	10.96
Camel	5.40	3.00	3.30	0.70	12.39
Ewe	6.18	5.15	4.17	0.93	16.43
Sow	6.77	6.22	4.02	0.97	17.98
Water buffalo	12.46	6.03	3.74	0.89	23.91
Reindeer	18.70	11.10	2.70	1.20	33.70
Whale	22.24	11.95	1.79	1.66	38.14

about a quart of milk. From there the milk goes to the teat cistern and into the streak canal. In milking, the pressure of the descending milk opens the sphincter muscles around the canal and forces the milk out *(6, 43)*.

Chemical Analysis of Milk

Scientists have found that milk contains more than 100 separate chemical components *(26)*. A chemical analysis shows that milk contains water, fat, protein, carbohydrate, and ash. Actually, however, milk is a very complex substance, for the fat of milk is in a state of emulsion with the aqueous solution of minerals and sugar, and the protein is in a colloidal (semi-solution) suspension. Fat-soluble and water-soluble vitamins also are present.

The average composition of cow's milk is shown in Table 1. The constituents of milk minus the water are called dry matter or total solids, and the constituents minus the water and the milk fat are called nonfat milk solids. The quantitative composition of milk varies somewhat, since it is influenced by breed, stage of lactation, season, ration, and environment. The average composition of milk of various mammals (cow, goat, ass, mare, camel, ewe, sow, water buffalo, reindeer, and whale) as compared

to human milk is given in Table 2. It can be seen from the table that human milk is much lower in fat, protein, and mineral content than cow's milk but higher in lactose. These differences are corrected when cow's milk is used in early infant feeding by adding sugar and water in proper concentration. For later infant feeding, cow's milk is more nutritious because of the higher content of fat, protein, minerals, and total solids. Whale and reindeer milks are highest in fat, protein, minerals, and total solid content but lowest in sugar (lactose) concentration of all mammalian milks (Table 2).

The great nutritional value of milk is due to the high quality of its proteins—casein, lactalbumen, and lactoglobulin; to its richness of minerals, particularly calcium and phosphorus; to the easy digestibility of its fat; and to its richness in fat-soluble vitamins—vitamin A and carotene (provitamin A), vitamins D, K, and E, and water-soluble vitamins—thiamine, riboflavin, niacin, and other members of the B complex.

Dairy Products

A variety of dairy products, possessing many of the nutritional qualities of milk, have been prepared by man. Reference has already been

TABLE 3
Dairy products manufactured today (43, 49, 67)

Class		
Milk	Cream	Skimmilk
Creamline	Coffee (40%)	Skimmed milk
Homogenized	Cereal (20%)	Buttermilk (cultured)
Certified	Half and half	Cheese (domestic and foreign)
Soft-curd	Cultured	Cottage cheese
Chocolate	Whipping	Cream cheese
Malted	Sour	Chocolate drink
Canned fresh	Plastic	Nonfat milk solids
Concentrated fresh	Pressurized whipped	Acidophilus milk
Concentrated frozen	Ice cream	Bulgarian and other fermented milks
Condensed	Butter	Frozen desserts
Evaporated	Butter oil	Lactose
Dried or powdered		Casein
Yogurt		Whey
		Dried whey
		Dried buttermilk

FIG. 2. History reveals that milk and its products have played an important part in the development of our civilization.

made to milk products almost as ancient historically as milk itself, such as curdled milk, cheese, butter, and fermented drinks. Today, other dairy products of outstanding nutritive value are manufactured; concentrated forms of milk, buttermilk, or whey, in which some or all of the water is removed by a process of condensation and evaporation (condensed, evaporated, powdered milks, or buttermilks) are available. Various types of cream are made from milk fat; skimmilk and nonfat dry milk solids are to be found on the grocery shelf; ice cream and products like lactose and casein are some of the dairy products developed in more recent years. Table 3 gives a list of the products made from milk, cream, and skimmilk.

What are these afore-mentioned dairy products?

Creamline milk refers to whole milk in which the yellow butterfat rises to the top of the bottle so that a line can be seen between the cream and the rest of the milk.

Homogenized milk is whole milk that has been subjected to high pressure, causing the fat globules to be broken up and resulting in an even distribution of tiny particles of butterfat throughout the milk. There is no appreciable separation of the cream.

Homogenized D milk is homogenized milk to which vitamin D concentrate has been added so that at least 400 U.S.P. units per quart are present.

Certified milk is whole milk produced under rigid standards of cleanliness and certified as such by the Council of the American Association of Medical Milk Commissions.

Soft-curd milk is milk in which the milk proteins have been treated to produce a soft curd, making it valuable in infant feeding.

Chocolate "milk" is a flavored drink generally made of fluid skimmilk or nonfat dry milk solids and water, to which have been added cocoa powder, sugar, a stabilizer, and other materials. Some butterfat may be added. If the product meets the state requirements for milk fat it can be called chocolate milk.

Malted milk is the product made by the combination of whole milk with the liquid separated from a mash of ground barley malt and wheat flour. The mixture is then dried.

Half and half is a mixture of half milk and half coffee cream.

Sour cream is made by inoculating coffee cream with a culture of lactic acid bacteria and ripening it to a low acidity and firm consistency.

Plastic cream is made by passing cream or high testing milk through a cream separator so that the fat content and consistency approach that of butter.

Whey is the portion that remains after the coagulation of the casein when cheese is manufactured. It is high in milk sugar and minerals.

Skimmilk is the product obtained in the separation of cream from milk.

Pressurized whipped cream is sweetened cream to which a gas like nitrous oxide is added under high pressure. Upon release to the air, the cream whips.

Buttermilk is the liquid portion of cream left after fat has been churned into butter. Though it is sometimes used as a beverage it is more often condensed and/or dried for animal feed.

Buttermilk (cultured) is a fermented skimmilk produced with lactic acid bacteria.

Fermented milks are those produced by adding bacteria that ferment or break down lactose (milk sugar) into lactic acid. This is a means of preservation used by the early users of milk. Different types of fermented milks are prepared with special bacterial cultures and by different methods of manufacture.

Acidophilus, yogurt, and bulgarlac are fermented milks prepared with special cultures of bacteria. These drinks, which originated in southern Europe, are supposed to have special therapeutic value.

Cheese is the food product made from the separated curd obtained by coagulating the casein of milk, skimmed milk, or milk enriched with cream. The coagulation is accomplished by means of rennet or other suitable enzyme, by lactic acid fermentation, or by a combination of the two. The curd may be modified by heat, pressure, ripening ferments, special molds, or suitable seasoning to produce the different varieties.

Over 400 different cheeses are recognized (52); however, cheese is usually classified into two general types—hard and soft, as follows:

Hard cheese
  Very hard
    Without gas holes—Cheddar
    With gas holes—Swiss
  Semihard
    Ripened by molds—Roquefort
    Ripened by bacteria—Brick
Soft cheese
  Ripened by mold—Camembert
  Ripened by bacteria—Limberger
  Unripened—Cottage

TABLE 4

Average composition of commonly used dairy products as compared to milk (12)

	Fat	Protein	Lactose	Ash	Water	Calories
	(%)	(%)	(%)	(%)	(%)	(per lb.)
Whole milk	4.0	3.5	4.9	0.7	86.9	310
Skimmilk	0.2	3.5	5.0	0.8	90.5	162
Buttermilk	0.5	3.5	4.6	0.7	90.7	167
Cheese (cottage)	0.8	19.2	4.3	1.7	74.0	459
Cheese (Swiss)	31.3	28.6	1.9	3.3	34.0	1,831
Cheese (Cheddar)	34.5	25.6	1.9	3.3	34.5	1,916
Cheese (cream)	39.9	14.5	1.0	1.9	42.7	1,910
Evaporated milk	7.9	7.0	9.9	1.5	73.7	629
Condensed milk	8.4	8.1	54.8	1.7	27.0	1,484
Dried whole milk	26.7	25.8	38.0	6.0	3.5	2,248
Dried skimmilk	1.0	35.6	52.0	7.9	3.5	1,630
Cream	39.9	14.5	1.0	1.9	42.7	1,910
Butter	81.0	0.6	0.4	2.5	15.5	3,325

Table 4 shows the average composition of some commonly used dairy products as compared to milk.

Nutritional Value of Milk

Thousands of scientific studies (22, 53) in the last quarter century have shown why milk and its products proved to be so essential in the diet of early civilizations. According to our present knowledge of nutrition, six essential elements are necessary for good health and well-being of persons of all ages. These are proteins, substances which build muscle, repair and build new tissues and organs; carbohydrates, such as sugars and starches, which give heat or energy (calories); fats, which supply heat and energy; minerals, which are essential for bone and teeth formation, and for the proper functioning of the body; vitamins, compounds which permit the efficient utilization of the other food nutrients; and water, which acts as a solvent and carrier for the nutrients in the body. In addition to these six basic nutrient classes, there are many specific chemical substances in each class (with the exception of water) that must be obtained to achieve good health and well being. At least 60 nutrient elements have been found to be essential in human nutrition (53).

Protein value. In the protein class, eight essential amino acids, the chemical compounds that contain nitrogen, are required in the human diet. The protein foods that contain these essential amino acids are called high quality proteins and are of animal origin. The proteins in milk supply all the essential "building stones" to make muscle, blood, skin, hair, and hormones.

According to recent nutritional surveys in the United States, protein is one of the nutrients usually deficient in our diet (5, 17, 36, 45, 53, 58, 59, 60, 61, 69). In growing children, expectant and nursing mothers, and athletes in training, in whom new tissue and larger muscles

are being rapidly built, milk proteins are the best source of nutrients. To furnish the amount of protein found in one quart of milk would require five large eggs, 5½ oz. of liver, 6⅓ oz. of fish, 5 oz. of dried beans, 5¾ oz. of beef, or 16 slices of bread (64, 67). Dr. P. C. Jeans, professor of pediatrics, Iowa State College (20), states, "In meeting the protein requirements, one should place emphasis on the value of milk. A quart of milk daily supplies most of the protein needs of the young child and half the need at the beginning of adolescence. Such a quantity of milk contributes more protein to the diet than any other single food."

Mineral value. In the mineral class, 13 essential mineral elements are necessary to maintain good health and vitality. These are the bone-forming elements—calcium, phosphorus, magnesium, and manganese; the blood-forming elements—iron, copper, and cobalt; and a miscellaneous group—sodium and chlorine (common salt), potassium, iodine, sulfur, and zinc. The ash in milk contains these essential minerals, especially calcium and phosphorus. Milk is low in iron, but other foods in the diet usually supply a sufficient amount.

For nonmilk users calcium is the nutrient most likely to be lacking in diets today (3, 5, 25, 32, 36, 38, 39, 50, 53, 55, 61, 69). This mineral is the chemical element found in oyster shells, chalk, and limestone. More calcium is needed by our bodies than any other mineral. This vital mineral is essential for strong bones and good teeth, for preventing rickets, for blood clotting, for stimulating the heart, and for the regulation of muscle-nerve responses. Milk is the most important source of food calcium for people of all ages. It is the only food that is a practical source of the total daily requirement for calcium. The recommended daily calcium requirements for people of all ages is given in Table 5. One quart of milk or its equivalent supplies one gram (1/28 oz.) of cal-

cium. Although vegetables, fish, and fruits contain this vital element, it would be impossible to eat these foods in the amounts needed to supply the daily calcium requirement (see Table 6). It has been estimated that one would have to consume approximately 28 oranges, 6¾ lb. of cabbage, 7¼ lb. of carrots, 27 lb. of potatoes, or 39 eggs to obtain the equivalent amount of calcium in one quart of milk *(64, 67)*.

Infants obtain their calcium from mother's or cow's milk. Children need calcium for bone and tooth growth, especially from 11 to 15 years of age, when the long bones grow fast. In periods of pregnancy and lactation, the amount of calcium for the new infant is obtained from the mother's body. If there is an inadequate supply of calcium in the daily diet during this period, the calcium will be withdrawn from the mother's bones and teeth, and a weakened condition will result. Since 1940 there has been a substantial increase in the number of early marriages. Two-fifths of the teen-age brides had a child in 1950, and for women under 20 there was a 33⅓% increase in the number of births between 1940 and 1950 *(33)*. Dietary surveys have indicated that the teen-age group and young women do not consume enough cal-

FIG. 3. Modern dairy plants are planned and equipped to process milk in a sanitary and efficient manner.

cium for optimal storage reserve, especially during pregnancy and lactation—the periods of stress. This results in a great strain on the mother who is trying to recover from mineral undernutrition *(23, 25, 55)*. Therefore, it is important that young women maintain adequate nutrition so that their bones will be fully mineralized when pregnancy occurs.

The calcium requirement of those past middle age is now considered to be double what was formerly recommended. Instead of one pint of milk daily, older people should drink a quart or more of milk for calcium retention in bones and teeth (see Tables 5 and 6).

It has been pointed out that in older people, particularly women, the daily calcium requirement sometimes is not met because of a reduced intake of food resulting from lessened activity *(38, 39, 50, 62)*. These people frequently experience loss of teeth and osteoperosis (loss of minerals in the bones), and hip and vertebra fractures are common *(62)*. Dr. Genevieve Stearns *(55)* of the Iowa State Medical School reports, "It has been observed that the digestive juice in the stomach tends to decrease in old age so that it is not surprising that the absorption of calcium, phosphorus, and magnesium is not efficient in older people. Their requirements should resemble those of young children. A gram or more of calcium daily, taken from milk, seems highly desirable for the elderly adult . . . As the mean age of our population increases we are concerned with postponement of senescence. Maintenance of a well mineralized skeleton throughout adult life may well be a factor in the maintenance of physical vigor into old age."

Recent studies *(11)* with radio-active calcium in experimental animals have shown that there is a constant exchange between the calcium in the blood and the calcium in the bones. Therefore, it is imperative that all people obtain their calcium requirement daily so that their skeletal system will not become weakened as a result of the diffusion of calcium out of the

TABLE 5

Recommended daily calcium requirements for people of all ages (8, 55, 66)

	(Grams)[a]
Nursing mothers	2.0
Pregnant women Toddlers Children Adolescents Young adults The aged	1.5
Adults (All not included above)	1.0

[a] 1 gram = 1/28 oz.

TABLE 6

Some food sources of calcium (64, 66)

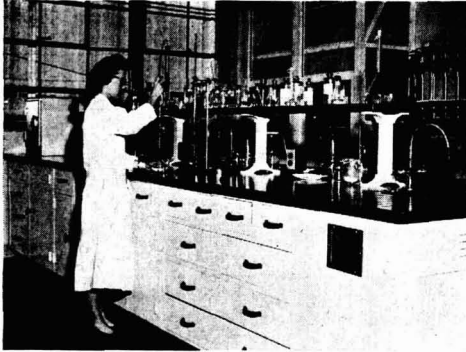| Food	Serving	Grams
Milk	1 qt. (4 cups)	1.151
Milk	1 cup	0.283
Skimmilk	1 cup	0.283
Buttermilk	1 cup	0.283
Cheese (American or Swiss)	1 oz. slice	0.247
Ice cream	1/6 qt.	0.132
Cottage cheese	1 oz.	0.025
Cabbage, outer green leaves cooked	1/2 cup	0.214
Cabbage, fresh cooked	1/2 cup	0.023
Kale, cooked	1/2 cup	0.225
String beans, cooked	1/2 cup	0.065
Carrots, cooked	2/3 cup	0.022
Celery, raw	2 large stalks	0.036
Leaf lettuce	1 large leaf	0.006
Salmon, canned	1/2 cup	0.097
Oysters	6 medium	0.068
Cucumbers	1 large	0.024

FIG. 4. Research plays an important part in the development of the dairy industry.

bones. The average lifetime of the American people reached a new high of 68.4 years at the half century *(57)*. The Bureau of Census reported that there were 12,759,000 persons at ages 65 and over in July, 1951 *(33)*. This means that there are more people past middle age than ever before. Therefore, these nutritional findings should be considered seriously by this group if they desire to continue to remain active and to enjoy good health.

Vitamin value. In the vitamin class are the fat-soluble vitamins: A, D, E, and K; and the water-soluble group: ascorbic acid (C) and the B family—thiamine, riboflavin, niacin, pyridoxine, para amino benzoic acid, pantothenic acid, biotin, choline, folic acid, inositol, and cobalamine (B_{12}). Many more B vitamins have been discovered, and their function in human nutrition is being studied. Vitamins are essen-

TABLE 7

Per cent of National Research Council's recommended daily allowances of calcium, riboflavin, and protein provided by a quart of milk (67)

Group	Calcium	Riboflavin	Protein
Children:			
1-3 years	115	168	86
4-6 years	115	140	68
7-9 years	115	112	57
Girls:			
10-12 years	96	93	49
13-15 years	89	84	43
16-20 years	82	67	46
Boys:			
10-12 years	96	93	49
13-15 years	82	80	40
16-20 years	82	67	34
Women:			
Age 25 years[a]	144	120	62
During pregnancy	77	84	43
During lactation	58	67	34
Men:			
Age 25 years[a]	144	105	53

[a] Woman or man, active, in good health, normally vigorous, and living in temperate climates.

tial for the efficient utilization of other nutrients.

All of the fat-soluble and water-soluble vitamins are found in milk *(26)*. The level of ascorbic acid (vitamin C) is reduced when milk is pasteurized; however, citrus fruits are excellent sources of this vitamin and should be included in the daily diet. Of all recognized vitamins, riboflavin is most deficient in our diets, and vitamin A is next *(5, 36, 39, 45, 50, 53, 58, 59, 60, 61, 69)*. Milk is a rich source of these two vitamins.

Riboflavin, one of the water-soluble B complex vitamins, is needed to prevent unhealthy skin conditions and dimness of vision. It is indispensable for growth, health, and vigor, as it is part of an enzyme system present in all living cells. A continuous supply of this important vitamin must be available from childhood through adulthood in order to remain young in appearance. A quart of milk or its equivalent in other dairy products will provide the daily requirement of riboflavin for people of all ages (Tables 7 and 8). To obtain the amount of riboflavin furnished by one quart of milk one would have to eat 3/4 lb. of cheese, 12 eggs, 1/5 lb. of liver, 1½ lb. of dried navy beans, 2½ lb. of lean beef, or 1½ lb. of greens *(64, 67)*.

Vitamin A is a fat-soluble vitamin. It is necessary for normal growth and well being; it prevents "night blindness" (the inability to see in dim light) and promotes normal vision; it also builds up resistance to disease. This vitamin or its precursor, carotene, is present in large amounts in butter, cream, and other products containing milk fat, as well as in fresh green and yellow vegetables.

Health-Promoting Nutrients in Dairy Products

The Food and Nutrition Board of the National Research Council has published recommended daily dietary allowances for the maintenance of health in the United States *(8)*. Based on their recommendations, the percentage of the nutrients usually found to be deficient in the U. S. diet—calcium, riboflavin, and protein—as provided by one quart of milk is shown in Table 7. The amounts of health-promoting nutrients in dairy products are given in Table 8. These products are excellent sources of calcium, riboflavin, protein, vitamin A, and energy and should be included in the daily diet.

Many people who do not show pronounced symptoms of malnutrition are suffering from "hidden hunger" resulting from the lack of the most important nutrients in the diet—vitamins and minerals. Malnutrition results not only from insufficient food but from improper food.

Milk and Weight Reduction

Many people are overweight; that is because they consume more calories than are needed. The secret in taking off pounds safely and still

TABLE 8

Health promoting nutrients in dairy products (67)

Product	Quantity	Calcium	Ribo-flavin	Protein	Vitamin A	Energy
		(mg.)[a]	*(mg.)*	*(g.)*[b]	*(I.U.)*[c]	*(cal.)*
Whole milk	1 cup (½ pt.)	288	0.42	8.5	390	166
Buttermilk	1 cup	288	0.43	8.5	10	86
Skimmilk	1 cup	303	0.44	8.6	10	87
Chocolate milk	1 cup	272	0.40	8.0	230	185
Malted milk	1 cup	364	0.56	12.4	680	281
Cheese (cottage)	1 oz. (2 T.[d])	27	0.09	5.5	10	27
Cheese (Cheddar)	1 oz. slice	206	0.12	7.9	400	113
Cheese (cream)	1 oz. (2 T.)	19	0.06	2.6	410	106
Evaporated milk	½ cup	306	0.45	8.8	500	174
Condensed milk (sweetened)	½ cup	418	0.60	12.4	660	490
Nonfat dry milk solids	3 T.	292	0.44	8.0	10	81
Ice cream	1/7 qt. brick	100	0.15	3.2	420	167
Cream, light (20%)	1 T.	15	0.02	0.4	120	30
Butter	1 pat (½ T.)	1	trace	0	230	50

[a] 1 mg. = 1/28,000 oz.
[b] 1 g. = 1/28 oz.
[c] I. U. = international units.
[d] T = tablespoon

feel "in the pink" is to watch the diet and choose foods that supply all the nutrients required for good health and are at the same time low in total calories. Dairy products fit into this category. In Table 8 it will be seen that the usual servings of dairy products—skimmilk, buttermilk, cheese, nonfat dry milk solids—are low in calories but high in calcium, riboflavin, protein, and vitamin A and are therefore ideal nonfattening foods. Butter is high in vitamin A content and is not fattening in itself. Increased weight occurs only when the total calories consumed are more than the body requires. One should not "cut out" health-giving foods because they contain a little fat but should "cut down" instead. Dairy products can and should be included in every reducing diet. Every person, even when reducing, needs high quality protein, minerals and vitamins. To reduce and remain in good health, one should drink milk and eat cheese, ice cream, and other dairy products.

Dairy Products Are Cheap

Agricultural economists, nutritionists, and home economists are all agreed that milk and its products are the most economical buy *(42, 53, 64, 67)*. Dairy products have risen less in price during the past 5 years than meats, beverages, fruits, and vegetables *(67)*. In a recent survey, the U. S. Department of Agriculture pointed out that for the money spent, dairy products offer excellent returns in proteins of high quality, calcium, riboflavin, and other minerals and vitamins. It has been estimated that the cost of the nutritional elements in a quart of milk would be nearly 60 cents if duplicated with other foods *(48)*. If milk were used for no other reason than to supply the daily calcium

requirement, it would be worth double its price. The other nutrients are bonuses.

Summary

Milk as a food has been used by man for thousands of years. It has been obtained from a variety of animals and has reached varied positions of importance in different cultures or even different periods in the same culture.

Milk and its products are a "must" in the daily diet. Milk is superior to any other food for muscle and bone building and for maintenance. It is an important and economic source of proteins of good quality because it contains all of the essential amino acids, the "building stones" of proteins so necessary for growth and maintenance. It contributes to the energy need; it contains mineral elements, particularly calcium and phosphorus, needed for strong bones and good teeth; and, most important, it supplies fat-soluble vitamins such as vitamin A for good growth and normal vision and the B-complex vitamins so essential for youthful appearance.

A quart of milk is a quart of health—that is the miracle of milk.

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A Guide for
Secondary School Recruitment Programs
In the Dairy Industry[1]

The Public Relations Committee
The American Dairy Science Association
1955

The dairy industry is currently furnishing employment for about one-seventh of the nation's population. The total number thus employed approximates the number of people living on farms throughout the United States. In addition to furnishing employment for many people, the industry provides a wide range of occupations. In the farm production of milk and in the processing (manufacturing) and distribution of dairy products, people in practically all walks of life are needed. In view of these facts, it should be clear to all concerned that to insure competent leadership throughout the industry in the years to come, many young persons must constantly be attracted to this area and trained in it. To this end, there is the continuous need of acquainting outstanding young people with the different phases of dairying and the career opportunities they offer.

The need for a youth education and promotion program in the dairy industry is urgent. Much is being done these days to help secondary school students select vocations into which they should go for technical training and lifetime occupations. The situation is one in which there is strong competition between large industries in recruiting the most promising young men and women. Wholehearted support by the dairy industry to a recruitment program at this time is doubly important for the reason that while the demand for well-trained young people is growing, the number electing to enroll in dairy curricula in many colleges and universities is on the decrease.

The effectiveness of a secondary counseling and recruitment program depends largely upon activity at the local level. Plans, literature, promotional material, and speakers are available at the state and national levels, but these will be of little importance unless the local organizations and/or persons assume the responsibility for developing, activating, and maintaining an effective local program.

One of the first needs of a counseling and recruitment program is to make certain that the persons who are to be involved have a full understanding of its purpose and of the educational and professional areas to which the program applies. A clear delineation of the two main areas of the dairy industry should be given along with the respective background requirements of the secondary school students who are contemplating entering into one of these areas. The following information is applicable:

Dairy Farm Production or Dairy Husbandry:[2] This is the farm or the basic agricultural phase of the dairy industry and involves such aspects as dairy farm management; dairy cattle feeding, breeding, health, and management; milk production practices; crop planning and production; land utilization and soils. A young person considering this field should have a farm background. The curriculum in the progressive Land Grant Colleges and Universities in this field is wide in scope and highly flexible. Such a curriculum insures the student of a broad education encompassing areas other than agriculture, such as liberal arts, physical and social sciences, business administration, and economics.

Job opportunities for the dairy husbandry graduate involve dairy herd management, dairy farm management, agricultural extension service at county, state, or federal level, vocational agricultural teaching (if program supplemented with suitable education courses), teaching and research in college, university, and federal agencies (graduate degrees usually required), agricultural sales representatives for a variety of commercial enterprises, fieldmen for dairy plants and milk producer organizations, farm inspectors for Health Departments.

Dairy Technology, Dairy Manufacturing:[3] This is the industrial phase of the dairy industry and pertains specifically to the procurement, handling, processing, manufacturing, sales, and distribution of milk and its products. The dairy husbandry and dairy technology fields overlap at the point of milk production on the farm. A farm background is not essential for the major portion of this field—a fact which must be

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[1] This is not to be reproduced in part or in whole without credit to The American Dairy Science Association.

[2] Certain dairy departments in which dairy husbandry is taught bear the name ''Dairy Science.'' This term has a broad meaning and applies equally to both areas of the dairy industry.

[3] Some of the major departments in the dairy products area are called ''Dairy Industry.'' This term is probably more descriptive of the subject matter covered than ''Dairy Technology,'' but is somewhat all-inclusive in its full meaning.

stressed in discussing dairy industry opportunities with secondary school students and faculties in the larger cities where "agriculture" and "dairying" are mistakenly assumed to be synonymous with "farming."

The principal goal of the University program in this area, as is the case also for the area of dairy husbandry, is to give the young persons involved a broad education. It should be emphasized that the dairy products area is basically a profession and the course program in a progressive department is designed to fit the graduate for industry leadership—provided he has the desire and ability. The program is well founded in science—because the past, present, and future success of the industry is based on scientific knowledge and because training in science is an ideal means of developing mental discipline. The progressive curriculum has a sufficient number of well-designed courses in dairy processing and manufacturing and, also, makes possible educational training in the areas of liberal arts, social sciences, business administration, and engineering. Flexibility of the curriculum permits each student to have a high degree of freedom in selecting courses which will suit his specific needs and abilities.

Graduates from the dairy products areas of universities and colleges are found in raw product procurement or field work; dairy plant operations (dairy plant production, supervisors, and managers); dairy plant management and executive positions; dairy engineering (equipment research, design, and sales, plant lay-out and production planning); laboratory product control (laboratory supervisors, directors, quality control specialists); dairy products and supplies, merchandising and sales; public and industrial relations and trade association positions; public health departments; teaching, research and extension work in universities and federal agencies (additional advanced studies usually required).

In all—emphasis should be placed on the fact that the university program in dairying is not designed simply for vocational training—but that it encompasses a broad educational area in which the student will be fitted to take his rightful place in a large, challenging, dynamic, stable industry.

The remainder of this manual consists of suggestions for a program for counseling and recruiting secondary school students. It is recognized that the suggestions incorporated will not all be used in any one recruitment program or at any one time. The local program will use such of the suggestions as are applicable to the particular situation. For example, the sections of the manual apply differently when a recruitment program is just being organized than when one has been active for a period of time. Also, a program in which the dairy processing and manufacturing phase of the industry is to be featured may be entirely different from one designed for emphasizing the dairy husbandry aspect.

Suggestions for an Effective Secondary School Recruitment Program for the Dairy Industry

I. ORGANIZATION.

A. Organization for an effective program involves close cooperation between state and local groups.

1. *State level.* The key to a successful program in any state is the dairy department of the state Land Grant institution. The department is both the leader and the coordinator of the entire program of the state. It would be expected to have suitable literature for dissemination; to set up details of scholarship programs with particular reference to standards and administration; to supply speakers for local and state meetings for discussing career opportunities in the dairy industry; to encourage local groups to establish active counseling and recruitment programs, and to assist them in developing and maintaining such programs; to serve as a communications center in maintaining full understanding between all interested groups within the state; and to encourage and bring into being an advisory "education" committee at the state level representing the commercial interests.

Another major need at the state level is an advisory "education" committee which represents the dominant dairy organization of the state concerned with the particular area of dairying being promoted. For example, the state Dairy Products Association would be the organization concerned with educational programs in dairy products. This advisory committee should meet frequently with the representative of the dairy department (not less than three times per year) to review and offer suggestions in respect to such matters as secondary school recruitment, curriculum adjustments, and the industry's treatment of university-trained men. Through this committee, funds may be obtained for financing certain phases of the recruitment program. The committee should have a rather

large membership with representation from each segment of the state in order to permit sectionalizing the state and maintaining proper communications throughout the entire area.

2. *Local level.* The success of an effective program is dependent, also, upon an aggressive group at the local level. The group may be at the county or city level. The county producers association or the local dairy technology society may serve as the organizing body.

The heart of the local program is a committee which may be designated as the "Education Committee." Too often when it is said, "Let George do it," the job is not done. The approach to a worthwhile program at a local level is to have an active education committee which will be charged with the responsibility for the program.

Care must be used in selecting the committee to make certain that the members are, themselves, aware of the work of the committee and are willing to dedicate themselves to it. "Enthusiasm begets enthusiasm," and positivism on the part of the committee is needed if the young persons contacted are to be convinced that the dairy industry has a place for them.

The committee should be established with an eye to continuity of the program. A complete change of the committee each year may be disastrous to a good program. The advice given is: Appoint some good men and keep them on the job. A rotational system for replacing committee members may be considered.

The local education committee should work closely with the dairy department of the state institution at all times in respect to such matters as contacting interested students, dissemination of literature, local meetings, and secondary school career days.

II. PROMOTIONAL AIDS AND POSSIBILITIES.

A. *Promotional literature.* Promotional litterature of various kinds can be used effectively—and the dairy department of the state university is expected to be the source of this material. Literature pertaining specifically to the respective state is thought to be more effective as recruiting aids than literature of a more general nature. However, literature which is produced at the national level is valuable as supplementary material. An attractive illustrated printed brochure is preferred to mimeograph material as the principal informational piece for distributing in the state.

Brochures should include a brief statement concerning the scope of the dairy industry and the areas of main interest within it; information of general interest about the institution and specifically about the dairy department, its facilities, the courses of training offered, and student activities. Stress should be placed on the opportunities afforded the young man who is willing to accept the challenge offered by the dairy industry.

The distribution of promotional material is an important consideration. Secondary school seniors are being flooded with such material and, because of this, special effort should be made to get the material into the hands of those outstanding students who are most likely to be interested in dairy industry careers.

Secondary school career days attended by dairy industry representatives, dairy industry celebrations, and special dairy events afford opportunities for many good contacts. For economical, general distribution it appears that the most logical persons through which the material might be channeled are secondary school counselors, vocational teachers, school principals, 4-H and F.F.A. counselors, and chairmen of the local education committees.

Another literature item which may be used is the poster—suitable for placing on bulletin boards. Attractive posters, with illustrations and drawings, but little writing, may be used to advantage in distribution to secondary schools, clubs, and posting in dairy plants. These may well be produced at the national level but with space so that the state distributing agency may be identified. The dairy departments should supply such posters if possible.

B. *Scholarships.* An effective scholarship program may serve as a core for successful local secondary school counseling and recruitment. A large number of scholarships is available at every college and university, and many worthy students need financial help to go to college. Most scholarships, however, are restricted to a special subject matter field, and the number available to dairy students is comparatively low.

One type of scholarship now being made available to dairy students is awarded either in one lump sum or distributed over one academic year. The

stipends range from $100 to $300, and may be awarded *(a)* to secondary school graduates to attract them into the dairy program at the state university, or *(b)* to students at higher levels in the university to encourage them to continue in the dairy curriculum or to reward them for work well done. It is this type of scholarship which is used largely in the secondary school recruitment activity.

Another type of scholarship is of a continuing design, in which the recipient is awarded a definite sum payable over a 4-year period. This type of scholarship is often established by a company or an individual in a given area. In some cases, the recipient is given a commitment that employment will be offered him each summer during his schooling period if he desires to work for the donor. This scholarship program has great promotional possibilities and insures for greater excellence of selection of the recipient.

Opinions vary as to how large scholarships should be. Some favor a fairly large number of small scholarships, whereas others favor a few large ones. Scholarships of $100 to $200 serve to attract students but they are not large enough to be of much help to the needy student. Scholarships should probably pay matriculation costs and miscellaneous fees.

The main objective of a scholarship program used for recruitment is to attract secondary students to the dairy industry. The size of the scholarship (within limits) is of less importance than the promotional effort put forth at the local level.

The procedure and care used in selecting the recipients is a major consideration in determining the type of scholarship program to have and the size of the scholarship. One procedure which may be used where a wide area is involved follows: *(a)* The interested secondary school student fills out an application blank (supplied by the dairy department of the state university or college) and refers this to the high school principal or counselor; *(b)* the secondary school official completes the blank giving his evaluation of the applicant, the applicant's standing in the senior class, the size of the senior class, and results of aptitude tests, and then mails the blank to the department at the university; and *(c)* at a designated time (late in April or early in May), the applicant meets for the personal interview and scholarship examination. The interviewing committee consists of several members of the local dairy organization sponsoring the scholarship and representatives of the university's dairy department. Each committee member is given the opportunity to evaluate each applicant. The examination is the standard written high school scholarship examination supplied by the state department of education in the fields of science, mathematics, and English, and is administered by a representative of the dairy department. The results of the evaluation are compiled in the dairy department and after consultation with the local committee chairman, the selection of the scholarship recipient is made. With this procedure, the major evaluation points considered are personal interview, high school report, and written examination.

The success of this procedure in selecting students who will do satisfactory work at the university is dependent upon *(a)* having a large number of applicants and selecting the best, *(b)* maintaining rigid and reasonably high standards in respect to all of the points considered in the evaluation, and *(c)* selecting a recipient who has the necessary adaptability and motivation. The general tendency is to give a well-meaning applicant the benefit of the doubt and to hope that he will do better at the university level than his high school record indcates he will do. When a questionable case is involved, the scholarship may be held in abeyance until after the student has demonstrated his ability to master the university program. In any case, the payment of the scholarship in installments over the first year is desirable, since it permits close supervision of the student.

Another procedure for selecting a scholarship is applicable where a limited area, or perhaps where only one high school, is involved. In this case, the selection is made by the principal, the county superintendent of schools, or a committee of the area high school representatives. This method is excellent and places the responsibility at the local level.

Generally, it is well to award the scholarship to the recipient with two stipulations: *(a)* that he remain in the dairy curriculum during the effective period of the scholarship and exhibit keen interest in the dairy industry, and *(b)* that he maintain at least average grades each quarter or semester. Failure to fulfill either of these requirements should result in immediate cancellation of the scholarship.

C. *Films and slides*. Moving picture films are being used extensively in vocational guidance work, particularly in secondary schools. They rate high as effective mass communication media, and the number of suitable films being made available is on the increase. In film libraries today, however, there are few films especially designed to acquaint the viewer with the many phases of the dairy industry and the career opportunities which the industry affords.

The objective of any films which may be produced should be to dramatize and popularize the dairy industry, and to suggest the many career opportunities and challenges which the industry affords young people. For most effectiveness a film should be specific and not too long. For example, two films are needed to cover the dairy field adequately, one designed for the farm production phase and the other for the processing (manufacturing) and marketing (distribution) phase.

Some production and processing films are now available which portray fairly well the production, handling, processing, and manufacturing of milk and milk products. These are helpful in meetings for secondary school and civic groups.

One approach to the subject of visual aid material is the development of appropriate slide strip sequences. This is an inexpensive means of telling the career story effectively.

I. SOME APPROACHES TO COUNSELING AND RECRUITMENT.

A. *Personal contact methods*.

1. *Individual personal contacts*. This involves a personal discussion between an individual in the dairy industry and some prospective secondary school student. This might involve an alumnus of the university, a dairy plant employee, a fieldman, or maybe a neighbor's son. Just a word, accompanied by a leaflet on dairying, will plant the seed; a follow-up may bring the harvest. It has been found that about 80% of students entering a specific area of a university do so because of a personal contact.

2. *Producer or plant letters*. A personal letter from the manager of a dairy plant or of a producers association to his producers is a means by which information regarding career opportunities in dairying may be disseminated. This may be accompanied by a leaflet describing opportunities for young people in the dairy industry.

Another possibility is a letter from the plant manager to his employees calling attention to the career opportunities for their own children.

B. *By local organizations*.

1. *Literature distribution*. A local organization through its education committee may distribute appropriate literature to secondary schools in the area and may see that use is made of it. Also, such literature is placed in the hands of any secondary school student who has evidenced interest in the field. Use of civic groups is also a possibility.

2. *Informing secondary school superintendents, principals, and counselors, and counselors for FFA and 4-H groups*. This may involve either a personal conversation with individuals or an organized meeting, at which time a presentation concerning the dairy industry is made to a group of representatives by a staff member of the university or a qualified local industryman. This meeting may be a dinner arranged by the local dairy industry. If this is done, there are three points to keep in mind: *(a)* have some key industry leaders present to mingle with the counselors. The counselors should realize that this job of recruitment is of concern to top management. In other words, this selling of the industry and its opportunities is a "man's" and not a "boy's" job. *(b)* Have a good dinner in good environment. The money is well spent. *(c)* Have a short informative program designed to arouse the interest of those present, and allow adequate time for questions. Avoid over-selling or self-promotion.

3. *Secondary school and civic group contacts*. Definite annual programs involving contacts at each high school are effective. Discussions with civic groups and meetings with secondary school groups at the local high school career days offer possibilities.

4. *Scholarship programs*. Every active dairy trade or professional organization has the possibility of developing a suitable scholarship program. Also, individual companies may develop such programs. The establishment of scholarship programs in memory of a deceased dairy leader offers a wonderful opportunity to have something of lasting value to perpetuate the beliefs and philosophies of a friend and associate.

C. *Through FFA and 4-H Clubs.* Many dairy industry leaders of tomorrow are members of FFA and 4-H Clubs today. For this reason every effort possible should be made to give aid and assistance to the activities of these two nation-wide organizations.

Owing to the fact that the 4-H Club program is directed by the agricultural extension service, the dairy industry through extension dairymen has always been well represented in the conduct of 4-H Dairy Club programs. FFA Clubs, to a lesser extent, because of organizational set-up, also have received a great deal of assistance from extension dairymen. In addition, by virtue of being high school students, FFA boys are reached through high school counseling and recruitment programs.

In addition to the contact being made by college and university representatives, the following are ways dairy industry organizations may support these two programs and thereby direct the interest of outstanding young people: 1. Provide program material for club meetings. Motion picture films about dairying might be included. 2. Assist club members in obtaining projects. This is a natural for state, district, or county breed associations. 3. Assist club members with exhibits and demonstrations. Plant fieldmen can do the best job here. 4. Arrange and conduct educational tours—judging contests and demonstrations. Take the farm boys to town to visit dairy plants and the town boys to the country to visit farms. 5. Conduct essay contests designed to inform and interest contestants in different phases of the dairy industry. 6. Provide suitable recognition for top performance and/or achievements in FFA and 4-H activities.

In many states, if not all, the FFA and 4-H Club programs are being supported by purebred breed organizations and by dairy industry organizations. To coordinate the assistance now being given, however, and to provide for an extension or expansion of the program, there is need for a memorandum of understanding or outline of plans whereby cooperation might be effected at the state level.

D. *Dairy plant and dairy farm open house.* "Seeing is believing"—and having organized "open houses" in the modern dairy plants and at modern dairy farms for adults and high school students is effective in promoting the dairy industry. Suitable hand-out literature setting forth the major branches or divisions of the dairy enterprise and raising questions about career opportunities are excellent supplements for such visits.

E. *Working experience for students.* "Experience is the best teacher"—and the hiring of high school and beginning university students by the dairy plant or by the dairy farm offers a means of acquainting these young people with the dairy industry. This may be a "selling" proposition; it may also be (and frequently is) an "unselling" one. In this employment, it should be the objective to give the student an opportunity to gain a fair appraisal of the dairy industry and what it may hold for him. He should not be "spoon-fed"; he should be shown "the facts of life"—that there is no easy way to success but that opportunities abound on every hand for the right man. However, to use these promising students without purpose and as cheap labor—and with no attempt on management's or supervision's part to counsel with them—will cloud the vision of these young men and often cause them to look to areas other than dairying for their educational and professional future.

F. *Career day at the university.* The crux of a year's high school counseling and recruitment program in any state may well be a dairy high school career day held at the university or college. This is a special day at which high school groups will be entertained on the campus and will be given an insight into the dairy industry. The success of this depends upon an efficient state-wide organization where all groups are working together with the view of having good attendance at the university.

The local dairy groups may supply transportation and arouse the interest among the secondary school students. Early planning with the high school counselors is essential in order to have proper representation. Whether to invite all secondary school prospects or only the upperclassmen is a decision which will need to be made to suit the state situation. Usually, inclusion of both junior and senior groups should be considered.

Great care needs to be used in developing the program at the university. It should be short, it should not be "stuffy" or too formal. Inclusion of some of the university students on the program is desirable. "Over-selling" may be disastrous—the main purpose is to arouse interest. Sufficient time should be allowed for the visitors to just "browse" about the campus without a completely "regimented" campus tour.

Example of letter from the plant manager to the milk producers regarding career opportunities in dairy technology, follows.

A similar letter may be prepared by the manager to go to each of his employees to solicit their support in interesting members of their families.

Dear .... :

Since you are a milk producer you are probably as concerned as I regarding the future leadership of the dairy industry. It is for this reason I am addressing this personal letter to you.

At present, a great shortage of trained men exists in our industry, especially in the processing and manufacturing branch, and career opportunities for young qualified men are excellent. A major task those of us in the industry have is to see that young men now in high school are made aware of these opportunities and encouraged to investigate the educational possibilities in this area.

In this connection, I am enclosing a leaflet which describes the field of dairy technology, the industrial phase of dairying, for which an educational program is designed at *(name of school)*. Will you please put this in the hands of a young man who is considering a university education so that he may be made aware of this challenging field of study.

If the young man would like to visit our plant, he would be most welcome. In fact, I would be happy to discuss the entire matter with him.

Thank you for your cooperation.

Very sincerely yours,

Plant Manager

Enc.

A Career Awaits in

DAIRY TECHNOLOGY

The Industrial Phase of the Dairy Industry
Science - Plant Operations - Management - Engineering - Sales

Dairying is one of the nation's largest and most stable industries—and the dairy technology phase deals basically with the procurement, processing, manufacturing, distribution, sales, and merchandising of milk and milk products. The field is wide and varied in scope, the opportunities for qualified men numerous and excellent. **Farm background is not essential.**

The broad four-year educational program of *(Name of school)* leads to the degree of Bachelor of Science in dairy technology. Excellent modern facilities are available for instruction —among the finest in the United States.

Dairy technology graduates are found in dairy plant operations, business management and executive positions, technical control, engineering for equipment and plant design and operation, product and equipment sales, field work with milk producers, public relations and trade association activities, public health, research, and teaching.

Scholarships for superior high school male graduates are available on a competitive basis.

For information consult the High School counselor or principal, the local dairy organizations, or write to the Department of Dairy Technology, College of Agriculture
(Name and address of school)

SCHOLARSHIPS

In Dairy Technology at (Space for school name)

Scholarships have been established for *(Name of state)* high school graduates to study dairy technology at *(School name)*. These scholarships become available with the opening of the autumn quarter and carry stipends of $150 to $300.

Each award will be made on the basis of a written examination, a personal interview, and an evaluation of the candidate's background, character, personality, and high school academic and extracurricular record. Proficiency in science and English is desired, and only male students will be considered. **Farm background is not essential.**

THE FIELD OF DAIRY TECHNOLOGY

Science—Plant Operations—Management—Engineering—Sales

Dairying is one of the nation's largest and most stable industries. Dairy technology is the industrial phase of the Industry and includes procurement, processing, manufacturing, distribution, merchandising, and sales of milk and milk products. Job opportunities exist for dairy technology graduates in plant operations, management, technical control, dairy engineering, product and equipment sales, field work with producers, public relations, public health, research, and teaching.

(Space for school name) has excellent modern facilities for the study of dairy technology and the Department is ranked high among the departments in the United States. A graduate in dairy technology receives a broad four-year university education, the degree of Bachelor of Science in dairy technology, and is virtually certain of a job in his chosen field. While attending college, he has an opportunity to earn part of his expenses and to obtain experience through summer work in the industry.

For information consult the High School counselor, the local dairy organizations, or write to the Department of Dairy Technology, College of Agriculture (Name and address of university or college)

JOURNAL OF DAIRY SCIENCE

ABSTRACTS OF LITERATURE

W. O. Nelson, Abstract Editor

ANIMAL DISEASES

598. Industrial molybdenosis of grazing cattle. J. C. Buxton, S. B. Loughborough, and R. Allcroft, Min. of Agr., Vet. Lab., Weybridge. Vet. Record, 67, 15: 273. 1955.

It is known that an excess of molybdenum in herbage causes a scouring disease of cattle called "teart." The increased molybdenum intake limits liver copper storage and can produce severe clinical symptoms of hypocuprosis. Recent work has indicated that molybdenum limits copper storage in liver of sheep only when inadequate inorganic sulfate is present in the ration. The unthriftiness and scouring caused by a high molybdenum intake can be helped by increasing the copper intake of the animals.

Evidence is presented showing contamination of herbage by molybdenum compounds in the escaping smoke and fumes of a metal alloys factory caused severe diarrhea and loss of condition in cattle. Affected cattle had low blood copper values and feeding of copper sulfate both prevented and cured the disorder.

R. P. Niedermeier

599. Effect of high and low vacuum milking machines on udder health and milk removal. R. Porter, D. D. Miller, and S. R. Skaggs, N. M. Agr. Expt. Sta., State College. Bull. 394. 1955.

A reversal trial was conducted, in which all cows were milked alternate 90-day periods with high and low vacuum milking machines. Of the 44 cows on test, 29 showed a positive bromthymol blue test at some time during the experiment, and only 3 showed flaky milk on the strip cup. These 3 cows all had histories of former mastitis. The results of mastitis studies of these particular cows did not reveal any difference between the two different machines used. Observations of all the cows showed leucocyte and total counts of the foremilk were not dependent on the machines in use. Individual yield was also unaffected by the type of machine, although a slight drop in production was noticed for 2 or 3 days after the machines were switched. Hard milking cows were milked more completely and faster by the high vacuum machine. Operators preferred high vacuum milkers, since they did not drop off the cows too readily.   R. W. Hunt

600. Control mastitis in dairy cattle. J. O. Schnautz, Ore. Agr. Expt. Sta., Corvallis. Bull. 545. 1954.

A series of questions and answers to describe mastitis and effective control measures.

R. W. Hunt

601. Two years of collective mastitis campaign in cooperative dairies with practical control of penicillin-induced starter failures by use of penicillinase or penicillin-resistant starter. (English text). H. C. Hovmand, A. Jepsen, and A. J. Overby, Royal Vet. and Agr. Coll., Copenhagen. Bull. 47. 1955.

A collective treatment of about 200 dairy herds for *S. agalactiae* mastitis resulted in elimination of 93% of the cases during a period of two years. This result was obtained by monthly inspection and treatment with penicillin. About 69% of the originally infected herds were cured between the first and the second inspection. After five months about 85% were cured.

The failure to completely eliminate the *S. agalactiae* mastitis was largely due to lack of cooperation on the part of some of the owners.

Serious incidences of yeast mastitis occurred in 12 of the penicillin-treated cows. The cows recovered only slowly from such an attack and several quarters were lost. No effective cure was found.

Mastitis due to streptococcus of groups L and Ca was difficult to cure. One treatment with penicillin appeared effective but reoccurrence of new cases was frequent after 7-8 months.

There did not seem to be any correlation between the amount of penicillin administered and the amount excreted with the milk. One cow excreted as little as 2.7% penicillin after one day whereas another excreted 84%. The average excretion was about 40%.

The penicillin-containing milk was successfully made into butter and cheese by the use of penicillinase. A mixed strain starter was successfully adapted to resist 1.0 unit of penicillin for the same purpose.   T. Kristoffersen

602. On the probability of total eradication of brucellosis in Denmark (English text). P. A. Bruhn, Royal Vet. and Agr. Coll., Copenhagen. Bull. 45. 1955.

The author's calculations based on the actual results obtained on the island of Boruholm show that brucellosis probably will be completely eradicated in Denmark by 1961.

T. Kristoffersen

603. Spandemaelksundersoegelser og besaetningsundersoegelser i mastitisdiagnostiken.

(**Examination of can milk samples and herd milk samples in mastitis diagnostics**). (**English summary**). O. KLASTRUP and P. L. PEDERSEN, Royal Vet. and Agr. Coll., Copenhagen, Bull. 50. 1950.

Comparative examinations of the milk from the individual quarters of 2,450 cows and of pooled can milk samples indicated that the latter method was well suited for diagnostic and following control of mastitis.

<div align="right">T. Kristoffersen</div>

BOOK REVIEWS

604. Methods of Biochemical Analysis, Volume II. Edited by DAVID GLICK. Interscience Publishers. 470 pp. $9.50. 1955.

The titles, authors, and number of pages devoted to the various chapters follow: Analysis of Steroids by Infrared Spectrometry, by Harris Rosenkrantz, (56); Chemical Determination of Adrenaline and Noradrenaline in Body Fluids and Tissues, by Harold Persky, (27); Lipide Analysis, by Warren M. Sperry, (31); Measurement of Lipoxidase Activity, by Ralph T. Holman, (9); Assay of Compounds with Folic Acid Activity, by Thomas H. Jukes, (33); Determination of Vitamin E, by Robert W. Lehman, (37); Methods for Determination of Coenzyme A, by G. David Novelli, (27); Assay of Proteolytic Enzymes, by Neil C. Davis and Emil L. Smith, (45); Determination of Glutathione, by J. W. Patterson and Arnold Lazarow, (21); Determination of Serum Glycoproteins, by Richard J. Winzler, (35); New Color Reactions for the Determination of Sugars in Polysaccharides, by Zacharias Dische, (47); Recent Developments in Techniques for Terminal and Sequence Studies in Peptides and Proteins, by H. Fraenkel-Conrat, J. Ieuan Harris, and A. L. Levy, (68); Spectrophotometric Assay of Cytochrome c Oxidase, by Lucile Smith, (9); Author Index, (20); Subject Index, (15).

This volume is one of a series which emphasizes methodology and instrumentation in the field of biochemical analysis. "Topics to be included are chemical, physical, microbiological, and, if necessary, animal assays, as well as basic techniques and instrumentation for the determination of enzymes, vitamins, hormones, lipids, carbohydrates, proteins and their products, minerals, antimetabolites, etc."

The presentation of the experimental details is such that the laboratory worker is furnished with the complete information required to carry out the analyses. "Those invited to prepare the respective chapters are scientists who have either originated the methods—or have had intimate personal experience with them."

The success of this commendable venture depends upon the wise selection of the most generally useful techniques and procedures. The regrettable duplication of effort often involving the same authors is readily apparent when recent treatises of a related nature are examined.

Some long range planning by such organizations as the American Society for Biological Chemists might partially circumvent this difficulty in the future and conserve space on already crowded bookshelves.

The number of obvious errors is not large but it would be less confusing if the editors and authors could agree on the manner of spelling such words as lipid (lipide). R. G. Hansen

605. Pilot Plant Techniques of Submerged Fermentation. Special English Edition of Rendiconti Instituto Superiore di Sanita. Vol. 17 Interscience Publishers, Inc. New York, N. Y. $8.10. 1955.

This book is a series of 12 papers from the International Research Centre for Chemical Microbiology in Rome. It is primarily concerned with the technical aspects of submerged fermentations with special emphasis on aeration studies and fermenter design. The authors state: "A number of technical innovations, some of industrial applicability, have been described; they have been subjected to extended practical tests and it is hoped and believed that they will serve to improve both the industrial and laboratory technique of submerged fermentations."

In the first paper aeration is discussed; an amperometric method for continuous measurement of oxygen concentration in agitated aqueous solutions is presented and compared with the conventional manometric methods. The effect of volume of solution, shape of container, and type of agitation on the rate of oxygen diffusion is discussed.

Papers 2 and 3 describe fermenter assemblies using vortex and sparger aeration systems; detailed drawings are given for both laboratory and pilot plant models. Paper 4 gives instructions for making a pressurized stuffing box designed to eliminate contamination. Paper 5 shows a typical pilot plant layout for submerged fermentations. Paper 6 outlines plans for the construction of a rotary shaker in which steel balls are used in place of universal joints; this shaker is described as stable, durable, easy to construct and almost noiseless in operation. Paper 7 presents a simple laboratory method for the qualitative and quantitative evaluation of the activity of antifoam agents. The importance of dispersion, the nature of the carrier, and the differences between baffled and free agitation systems from the point of view of foam inhibition are discussed.

Papers 8 and 9 are concerned with the submerged fermentation of *Escherichia coli* and *Penicillium chrysogenum*, Thom. The interrelation of protein and polynucleotide synthesis is shown in the former. In the latter the effect of mechanical agitation on mycelial formation and autolysis is discussed. Paper 10 describes a new soil fungus for which the genus name *Romanoa* has been suggested. The type species is *Romanoa terricola;* it has antibacterial ac-

tivity against *Bacillus subtilis* and *Micrococcus pyogenes* in agar culture but not in broth culture filtrates. Papers 11 and 12 deal mainly with the genetics of *Penicillium chrysogenum*. A simple technique is given for constituting heterokaryons between mutant strains with complementary nutritional requirements in *Penicillium chrysogenum*, Thom. A procedure is also given whereby it is possible to confirm the process of "parasexual" recombination in this imperfect fungus.

This book is well organized and well written. Directions are given in a straightforward manner. There are 250 pages of text and over 100 pages of graphs, diagrams, and plates, some of which are in color. Each chapter is written in English and is summarized in English, Italian, French, and German. A portion of each chapter is devoted to discussion and adequate references are given in each instance. This book should be of value to anyone interested in submerged fermentations either on a laboratory or commercial production scale. It is not primarily a textbook but anyone interested in bio-engineering would find this a valuable source of technical information as the engineering aspects of submerged fermentation are emphasized.

O. W. Kaufmann

606. Dairy Cattle—Selection, Feeding, and Management—4th Ed. W. W. Yapp and W. B. Nevens. John Wiley and Sons, Inc., 420 pp. $4.76. 1955.

This edition has been completely rewritten and brought abreast of new developments in the many areas having a bearing upon dairy farming and dairy cattle management.

Twenty-nine chapters are grouped into 9 parts as follows: 1. Dairy Farming Contributes to National Welfare, 2. Selecting and Breeding Dairy Cattle, 3. Feeding Dairy Cattle, 4. Managing Dairy Cattle, 5. Financial Aspects of Dairy Farming, 6. Milk Secretion, Care and Merchandizing of Milk, 7. Producing Roughages for Dairy Cattle, 8. Providing Good Buildings for Dairy Cattle, 9. Looking into the Future. The authors competently discuss the subjects indicated in the part and chapter headings.

The text is illustrated with 115 figures, chiefly photographs, 51 tables, 2 full-page pedigrees and two pages carrying 4 reproductions in color. Each chapter is well documented with references and concludes with a series of review questions.

In the preface the authors state "—our aim has been to point out principles which students and dairy farmers may use as guides rather than to give explicit directions for doing every job, keeping in mind the fact that new machines, new types of buildings, and new pieces of equipment may alter the manner in which the various operations are carried out. The principles governing good care and management of dairy cattle, however, remain the same as

they were at the time the book was first issued in 1926." This objective has been well achieved. Discussion and elaboration of the various subjects is well balanced.

The text has been well edited. The style used is consistently followed. The vocabulary and sentence structure would appear to require some effort on the part of 9th grade students, for example, to master and fully comprehend. For those who seek to master the subject matter presented, the book should prove to be a reliable and helpful guide.   I. W. Rupel

607. Holtbare Milch. (Nonperishable Milk) M. E. Schulz. Verlag Hans Carl, Nurnberg, Germany. 194 pp.

Much of our present knowledge about sterile milk has been summarized in the 194 pages of this book. Sterile milk is considered an international problem and a means of encouraging greater milk consumption in the temperate and tropical regions of the world. Tables reveal a steady increase in the production of sterile milk in certain European countries.

Technical information is conveyed under the heading of the following chapters: Tests of Milk to Determine Its Suitability for the Manufacture of Sterile Milk; Fundamentals of the Manufacture of a Good Sterile Milk; Retorts, Continuous Sterilizers and Aseptic Canners; Packaging of Sterile Milk; Technical Control of Sterile Milk; Changes Occurring as the Result of Sterilization and Storage; Pasteurized Milk Versus Sterilized Milk; Sterile Dairy Drinks; Fermented Milk; Sterile Reconstituted Milk; Hydrogen Peroxide—Catalase Treated Milk; Sterile Buffalo, Ewe, and Goat Milk.

Considerable attention was given to milk sterilized in retorts of various types and illustrations are given of the type of equipment available. The author was quite aware, however, of the advantages of continuous sterilization and aseptic canning and cites a number of advantages of such systems.

In reading this book one gains interesting impressions of the problem of milk production and distribution in many parts of the world. Sterile milk does appear to hold promise of increasing milk consumption. The author points out that the flavor of pasteurized milk is better than that of sterile milk, but in tropical lands where milk has to be boiled this is not a serious problem. Even in Western Germany 20% of the bottled milk sold is sterile milk. This indicates that people realize the advantages of a nonperishable milk and may be educated in its consumption.   J. Tobias

608. Analysis of Insecticides and Acaricides. F. A. Gunther and R. C. Blinn. Interscience Publishers, Inc., New York. 696 pp. $14.00. 1955.

The book consists of three sections divided into 15 chapters. Section 1 deals with problems

encountered in securing quantitative residue data, Section 2 with problems in analyzing technical grade materials and formulated products, and Section 3 with sampling, measuring, processing, cleanup, and analytical methods. An appendix includes detailed ultraviolet and infrared spectra. Emphasizing plants, plant parts, and soils as substrates, rather than animal products, the authors have selected procedures based on their proved general utility and reliability either in their own laboratories or upon their general acceptance in similar residue or regulatory laboratories. Some of the methods and modifications thereof are published for the first time. Procedures of highly specialized and limited applications are not included, but references to most of them are cited and their utility is mentioned to orient the analyst who has need for a method of restricted utility. Detailed composition and residue analytical procedures in current use and which are known to be reliable by the authors are given for 90 insecticides and acaricides.

In a field which is changing rapidly, this work cannot be expected to be nor was it intended to be complete even at the time of publication, much less 10 years hence. This hardly limits its value, however, for it should be a worthwhile reference and guide for many years to come. It attempts to initiate standards in a realm where standardization at best has been haphazard. As such, it is a work which most insecticide analysts can ill afford to be without.

N. Gannon

609. Handbook of Food and Agriculture. Edited by FRED C. BLANCK. Reinhold Publishing Corporation. 1955.

Twenty-six chapters and appendix; 1039 pages with numerous tables and figures. Each chapter is written by a specialist in the field covered and is followed by a comprehensive literature citation. Some of the chapters of especial interest to workers in dairy industry are: 7. Enzymes, 8. Oxidative Rancidity and Antioxidants, 9. The Essential Nutrients, 10. Storage of Agricultural Raw Products, 11. Food Preservation, 12. Effect of Canning and Dehydration on the Nutritive Values, 13. Food Spoilage and Deterioration, 15. Dairy Products, by Byron H. Webb, 16. Vegetable Fats and Oils, 21. Food Engineering, 22. Food Packaging, 23. Food Quality and Quality Control, 24. Disposal of Food Plant Wastes, 25. Chemicals in Foods, 26. Food, Drug, and Cosmetics Act. The Appendix of 109 pages consists of five parts: 1. Food Laws, II. Nutrition, III. Special Food Agencies, IV. Research Groups, V. Miscellaneous Information. It is the hope of the editor "that the Handbook of Food and Agriculture will earn a useful place on the desk of every worker who has to do with foods . . . We have particularly designed this book to meet the needs of the small laboratory or factory with a limited number of reference books."

L. M. Dorsey

CHEESE

610. Factors involved in the control of gelatinous curd defects of cottage cheese. E. B. COLLINS, Univ. of Calif., Davis. J. Milk Food Technol., **18**, 7: 169. 1955.

Prolonged incubation of cottage cheese inoculated with *P. fragi, P. viscosa,* and *A. metalcaligenes* caused surface spoilage at initial pH values as low as 4.6 and temperatures as low as 3.5° C.

Fruitiness is a common defect that limits the storage life of cottage cheese and is primarily due to the growth of *P. fragi.*

Violet red bile agar was a satisfactory culture medium for detecting the three species.

H. H. Weiser

611. Increased sales with tear-tape packages. ANON. Milk Prod. J., **46**, 7: 17. 1955.

Tear-tape packaging of rindless Cheddar cheese has resulted in a 16% increase in sales for an Oregon creamery. The ease with which the package opens seems to be popular with consumers.

The tear-tape wrapper has been used successfully for one-half, one, two, and five lb. packages. The packaging is accomplished by machine, thus saving time and labor costs.

J. J. Janzen

CONDENSED AND DRIED MILKS; BY-PRODUCTS

612. Evaluating the heat treatment of skim-milk in the manufacture of nonfat dry milk solids. D. W. MERRILL, Dept. of Dairy and Food Ind., Univ. of Wis., Madison. Milk Prod. J., **46**, 7: 56. 1955.

The use to which nonfat dry milk solids is to be put dictates the preheat treatment used in its manufacture. A method of determining the heat treatment given milk is important because of the wide range of heat treatments required for nonfat dry milk solids for different uses. Some of the more notable effects of heat on milk include changes in flavor, serum proteins, and curd tension.

The most used index of heat treatment is the % serum protein denaturation. A calculation of the % serum protein denaturation requires a determination or knowledge of both the total serum protein content and the denatured serum protein content. The total serum protein content of the milk is determined periodically on the raw milk. Unless the raw milk samples are analyzed for every batch, the assumption that the serum protein content is constant from day to day and milk to milk must be made. The undenatured serum protein content of the sample is determined by the Harland-Ashworth test.

An exact estimate of heat treatment from a determination of the serum protein denaturation would require: (1) that identical heat treatments cause the same % serum protein

denaturation in all milk, (2) the total serum protein content of the milk be determined on each sample or that the assumed serum protein content be constant and correct.

The limitation of such serum protein denaturation determinations is that neither of these conditions are met exactly.

Heating milk reduces both the rennet curd tension and the pepsin-HCl curd tension. These tests have shown some indication of being useful in evaluating heat treatment. The rennet curd tension method was found to be more sensitive to heat treatment than the pepsin-HCl method.

J. J. Janzen

613. Using nonfat dry milk solids in home prepared foods. L. G. MAHARG and M. MANGEL, Mo. Agr. Expt. Sta., Columbia. Bull. 632. 1954.

This bulletin contains several recipes which include soups, main dishes, breads, desserts, and milk drinks. All recipes call for the inclusion of nonfat dry milk solids.   R. W. Hunt

DAIRY BACTERIOLOGY

614. 1954 Summary of disease outbreaks. C. C. DAUER and G. SYLVESTER, Public Health Service, Washington, D. C. Public Health Rpts., 70, 6: 536. 1955.

In 1954, disease outbreaks in the United States attributed to foods were as follows: dairy products, 8 outbreaks (100 cases) ; water, 7 outbreaks (452 cases), and other food products, 230 outbreaks (11,421 cases). Dairy products were involved in less than 4% of all disease outbreaks reported.

Raw milk was responsible for one outbreak of brucellosis and typhoid fever. In one outbreak of typhoid fever raw milk was suspected but definite proof was lacking. Milk also was involved in an outbreak of shigellosis; an investigation indicated that the milk probably was contaminated by an assistant cafeteria worker. Milk apparently was responsible for several cases of tuberculosis in children; 80% of the herd supplying the milk showed a positive tuberculin reaction.

Cheese was involved in an outbreak of *Salmonella* food poisoning in one hospital. Cream cheese containing Gram positive cocci was responsible for an outbreak of gastroenteritis. Cheese was also suspected in one staphylococcal food poisoning outbreak.

Ice cream mix which was improperly held for several hours prior to freezing was responsible for 100 cases of staphylococcal food poisoning. Ice cream was also involved in another staphylococcal food poisoning; in this instance the product was contaminated by a chef who harbored *Staphylococcus aureus* in his throat and on his hands.

Eggnog, prepared by a carrier of *Salmonella typhimurium,* caused food poisoning at a large institution.   O. W. Kaufmann

615. Bactericidal effectiveness of iodophor detergent sanitizers. W. S. MUELLER, Univ. of Mass., Amherst. J. Milk Food Technol., 18, 6: 144. 1955.

The bactericidal properties of iodine liquid and iodine powder were comparable. Available iodine in 25 p.p.m. incorporated in the iodine detergent sanitizers was equal to 100 p.p.m. of available chlorine in killing *E. coli, S. typhosa, M. pyogenes* var. *aureus,* and *P. aeruginosa* in the presence of hard water (500 p.p.m. $CaCO_3$), 1% whole milk and 1% dish wash soil.

When the iodine was reduced to 12.5 p.p.m. and compared to chlorine under similar conditions, *E. coli* and *S. typhosa* were destroyed.

It appears that iodine detergent-sanitizers can be used for sanitizing food utensils, if field tests substantiate results of the laboratory performance tests.   H. H. Weiser

616. Bacterial counts of milk as affected by inconspicuous deterioration in milking machine teat-cup liners. T. J. CLAYDON, Kan. State Coll., Manhattan. J. Milk Food Technol., 18, 6: 160. 1955.

Bacterial counts in milk were higher when used teat-cup liners were tested as compared to new liner units.

A high contamination in the liners was necessary to cause an appreciable increase in the microbial content of the milk. When the bacterial count was low, there was very little difference in the microbial count of the milk. Bacterial accumulation increased more rapidly in used liners than in new liners.

H. H. Weiser

617. Application of the coliform test to pasteurized milk and cream. J. M. FRAYER, Vt. Agr. Expt. Sta., Burlington. Bull. 578. 1955.

No relationship exists between the coliform count of a particular sample of raw milk and of the same milk after pasteurization. The use of as many as 5 tubes inoculated with 1 ml. of sample does not indicate whether several colonies or a single coliform organism is contained in the milk. Coliform tests on single bottles of pasteurized milk or cream have virtue, in that positive results indicate carelessness somewhere along the line. The points of carelessness may be traced by making routine line tests at as many points as possible between the pasteurizer and the closed bottle. However, coliform tests should be used only as an indication and should not be considered in the absolute sense. A total of 782 samples of pasteurized milk was plated on violet-red-bile agar and duplicate sets on desoxycholate agar. Colony counts indicated that either agar is satisfactory for the enumeration of coliform bacteria.   R. W. Hunt

618. Relation of foam to the presence of coliform. H. B. SIEGMUND, Hendler Cry. Co., Baltimore, Md. Ice Cream Field, 65, 4: 64. 1955.

The results of a cooperative study are given. Two methods of pasteurization were employed, viz. 165° F. for 30 min. and a HTST system in which the mix was heated to 240° F.

In both types of pasteurization plating of foam immediately after pasteurization gave no coliform organism. When the foam was held for 24 to 48 hr. before plating, positive results were obtained with both types of pasteurization.

It is concluded that foam should be eliminated before any heating process occurs, since the presence of foam has been shown to contribute to inefficient pasteurization.   W. C. Cole

DAIRY CHEMISTRY

619. An instrumental method for measuring the degree of reversion and rancidity of edible oils. S. S. CHANG and F. A. KUMMEROW, Univ. of Ill., Urbana. J. Am. Oil Chem., **32**: 341. 1955.

This test is reported to provide values for rancid fats which can be correlated to flavor judgments. The test measures the volatile carbonyl compounds which are diffused from 100 g. of oil after a stream of nitrogen has passed through the sample. The carbonyl compounds are converted into their 2,4-dinitrophenylhydrazones and then measured by the absorption of the wine-red color of the quinoidal ions at 480 mμ. From the absorbence obtained, a carbonyl index is calculated and assigned to the oil.
                                    R. W. Hunt

620. Stability of ascorbic acid in citrus concentrates during storage. J. R. MARSHALL, K. M. HAYES, C. R. FELLERS, and C. W. DUBOIS. Quick Frozen Foods, **17**, 12: 50. 1955.

The ascorbic acid in citrus concentrates is stable. When stored at temperatures between 10 and 75° F. the product deteriorates physically and chemically very rapidly, whereas the loss of ascorbic acid is approximately 4%. The rate of deterioration of citrus concentrate is a function of temperature. Concentrate held at a temperature of 0 to –20° F. is inactive chemically and the ascorbic acid loss is 2%.

There is no loss of ascorbic acid in the concentration of the fruit solids. The deterioration of the product cannot be correlated with the loss of ascorbic acid.

Variations in the amounts of ascorbic acid in frozen concentrated citrus as delivered to the consumer are affected more by varieties of fruit and by conditions under which the fruit was grown than by any losses incurred in distribution.   L. M. Dorsey

621. Determination of total solids by modifications of the Mojonnier test. W. G. JENNINGS and N. P. TARASSUK, Univ. of Calif., Davis. Milk Dealer, **44**, 10: 46-47. 1955.

The Mojonnier test is modified to permit: (1) Cutting the heating period from 10 to 2 min., (2) Eliminating the transfer to cooling desiccator, and (3) Cutting the cooling period

from 5 to 1½ minutes. Preliminary studies indicate the traditional Mojonnier accuracy is preserved for a drying period of 2 min., the largest deviation from the results of the standard Mojonnier procedure was 0.03% T.S. When the drying period was shortened to 1½ min. or lengthened to 2½ min., only 1 sample out of 15 showed a deviation greater than ±0.05% T.S. Construction details of the modified test are given.   C. J. Babcock

DAIRY ENGINEERING

622. Two effects do work of 4 in new 6-stage steam-jet evaporator. A. V. GEMMILL, SR. Food Eng., **27**, 7: 48. 1955.

A new type double-effect steam evaporator has various outstanding features. A triple-stage operation is accomplished in each of two effects by a patented system of vertical baffles. This gives efficiency almost equal to that of the usual quadruple effect evaporator with smaller size and less investment. The unit is automatically controlled and operation is exceptionally efficient and economical. Low temperatures of heating mediums and high velocity of product through evaporator result in a finished product with no cooked flavor. The unit is designed for concentration of 8.8% skimmilk to 42% solids and is manufactured in capacities of 4,400 to 26,000 lb/hr.   T. J. Claydon

623. The Roswell pasteurizing process. R. O. TARDIFF, Breyer Ice Cream Co., Philadelphia, Pa. Ice Cream Field, **65**, 3: 36. 1955.

The author describes in some detail the Roswell pasteurizer installation at one of their plants. He concludes "The Roswell equipment is flexible and lends itself to various forms of installation."   W. C. Cole

624. Concrete floors. W. OHMAN, Portland Cement Assoc., Lansing, Mich. Milk Plant Monthly, **44**, 7: 33. 1955.

The best concrete floor for a milk plant is one with a heavy-duty industrial finish. It can be finished smooth and slip-resistant by incorporating such materials as carborundum or emery in the mix. The main factors that govern its strength and durability are the proportion of water to cement in the mix, the characteristics of the aggregate, bonding of the wearing surface to the base slab, placing and finishing, and curing. The water-cement ratio affects the strength and durability of concrete more than any other single factor. The less water used per sack of cement the better the cement-water paste and the stronger, more durable concrete. Methods of laying, troweling, and curing are discussed.   C. J. Babcock

625. Aluminum corrosion control in refrigeration service. R. L. HADLEY, General Electric Co. Refrig. Eng., **63**, 6: 65. 1955.

Aluminum has many of the physical and chemical properties of an ideal material for

refrigeration systems. Resistance to corrosion is a doubly important property since it must maintain a clean, bright appearance and the ability to retain refrigerant for many years.

Much is known about the protection of aluminum by its adherent oxide film and the chemical conditions under which the film may fail. The highly reactive nature of a clean aluminum surface results in susceptibility to corrosion by electrolytic cells activated by contact with chemical concentration variations, differential aeration, nobler metals, or traces of chemical compounds which promote failure of the protective film. All of the unfavorable environmental factors can appear in refrigeration service.

General corrosion, which is detrimental to appearance, may not represent as serious a hazard as the presence of isolated, perforating pits resulting from concentrated corrosion of small areas.

Moisture condensing from the atmosphere permits continuous formation of electrolytes for corrosive attack. Hazards are greatest during periods of defrosting and in gradient areas where condensate does not freeze. It can promote the operation of corrosion processes arising from such factors as unsealed crevices, contact with copper components in the system, the use of insulating or structural parts from which impurities can be leached, improper joining techniques, and atmospheric contamination.

Good design and careful control of manufacturing and finishing are requirements for assuring maximum resistance of aluminum to corrosion. L. M. Dorsey

626. New trailer refrigeration system. ANON. Ice Cream Field, **65**, 4: 37. 1955.

A completely automatic and thermostatically controlled trailer refrigeration system which operates off a live trailer axle is described. It is claimed it will maintain any specified temperature from −20° to +50° F. throughout the entire trip. W. C. Cole

DAIRY PLANT MANAGEMENT AND ECONOMICS

627. Report on quantity discounts. ANON. Milk Dealer, **44**, 10: 137. 1955.

A query of a national cross-section of milk dealers showed that 35% of the replying dairies have some form of discount plan in operation. Of the dairies answering, 15% give discounts through ½ gal. and gal. jug prices. Of the remainder, some have tried limited quantity discount plans and discontinued them, while others have never had a plan in operation. Most of the replying dairies using quantity discounts did not include by-products in their discount rates. The results obtained by various plans are discussed. C. J. Babcock

628. Market reaction to gallon jugs. S. JOHNSON, Univ. of Conn., Storrs. Milk Plant Monthly, **44**, 7: 18. 1955.

Research to date does not prove that gal. jugs hurt milk consumption but casts doubt on claims that they help consumption materially. Probably the most important factors affecting the consumption of fluid milk are its price, consumer incomes, and the availability of a high-quality product. Estimated savings as compared with qt. bottles are: (1) 0.1¢ per qt. on container cost, assuming an average deposit of 22½¢ per gal. jug, 45 trips per gal. jug, and 30 trips per qt. bottle, (2) 0.3¢ per qt. on cost of bottle caps, assuming an inexpensive cap for the gal. jugs, and (3) loss of 0.1¢ per qt. on the filling operation. These items total 0.3¢ per qt. saving for gal. jugs on container and processing costs. The majority of people in industry believe that store customers prefer the ½-gal. paper package over the gal. jugs and that consumer demand is dominated by the pricing of the package rather than the desirability of the packages. C. J. Babcock

629. Costs of accidents. E. B. KELLOGG, Milk Ind. Foundation, Washington, D. C. Sou. Dairy Prod. J., **57**, 2: 140. 1955.

In 1952, fluid milk company employees were injured at work at the rate of 23.1 injuries for each million man-hr. The av. for all industry was 10.1. The most hazardous jobs in decreasing order of rate were bottling and casing, cold room, loading, delivery, milk processing operations, and plant and equipment maintenance. An accident cost includes higher insurance rates, cost of damaged equipment and product, loss of work time, cost of time of supervisory personnel, and losses of employee and public goodwill. A $100 loss represents the av. profit on the sales about 30,000 qt. of milk. Accidents may be decreased by the use of information such as "Accident Prevention in the Fluid Milk Industry" published by Milk Industry Foundation, keeping records of personal injuries and vehicle accidents, setting up a safety committee and keeping safety before everyone. F. W. Bennett

630. How to get better results from your sales contests. D. BURGER, Carnation Co., Houston, Tex. Sou. Dairy Prod. J., **57**, 2: 44. 1955.

Travel and merchandise are excellent prizes for sales contests because they lend themselves to showmanship. Recommended features of such contests are an enthusiastic kick-off meeting including the contestants and their wives, representatives of the concerns selling the articles offered as prizes, lively entertainment, and liberal publicity; mailing pieces to arouse enthusiasm of the wives, liberal information of the progress of the contest to customers, and full publicity about the winners of the contest. The cooperation of other companies in giving minor prizes consisting of some of their merchandise and in promoting the contest will broaden and increase interest. F. W. Bennett

631. Office responsibility for greater and more profitable sales. B. G. NICHOLS, American Dairies, Inc., Kansas City, Mo. Sou. Dairy Prod. J., **57**, 6: 37. 1955.

An office is a service organization which coordinates all departments of the company. It records and interprets the financial history of the company up to the moment and even predicts some of the future. Included should be all usable cost records, comparisons of sales volumes, comparisons of individual and composite tests, statements of returned products, and products unaccounted for. An efficient routine system of reporting should be developed which will avoid duplication and useless reports. The office force should get along pleasantly with people in all departments and with the public and provide as much needed information as possible without the necessity of specific requests.   F. W. Bennett

632. Management's responsibility for greater and more profitable sales. G. W. BECKMAN, Beatrice Foods Co., St. Louis, Mo. Sou. Dairy Prod. J., **57**, 6: 36. 1955.

The manager's responsibility for sales includes evaluating and controlling administrative costs, establishment of sound policies or programs for personnel, regular job evaluation, safety, maintenance, purchasing, sales and public relations and providing and assuring compliance with rigid product standards. Altogether, any manager should keep his organization alert, active and profit-minded.
                                          F. W. Bennett

633. How to plan a refrigerated fleet. W. J. THOMPSON, Carnation Company, Los Angeles, Calif. Milk Plant Monthly, **44**, 7: 15. 1955.

Modern refrigerated retail and wholesale trucks, equipped with self-contained refrigeration, cost up to $4,500 and $6,200, respectively. Combination ice cream and wholesale milk bodies cost about $6,850. The cost of refrigerated trucks is considerably less when refrigerated with an ammonia hook-up from a central system.

Nearly all of the wholesale trucks and about 30% of the retail trucks operated by the Carnation Company are refrigerated. The desired temperature of any truck body is governed by the plate solution. Minus 8° or –12° F. plates are used in the ice cream bodies and +22° and +26° in the milk bodies, depending on the ambient temperature where the trucks are required to work. Two 1-h.p. units are installed in 1,000-gal. ice cream bodies, a 1½-h.p. unit in 12½-ft. wholesale milk bodies, a 2-h.p. unit in 15-ft. wholesale milk bodies, and two 3-h.p. units are used to charge the 10 large plates in 35- to 40-ft. ice cream vans. In retail trucks a heavy duty ¾-h.p. or 1-h.p. semi-sealed unit is used in connection with two 30″ × 60″ × 2⅝″ holdover plates or one large 36″ × 66″ × 2⅝″ plate.

The actual cost of refrigerating trucks with Freon averages from $300 to $500 more than refrigerating with ammonia. The cost differential is large because of the Freon compressor units, the tubing, etc., and the cost of the Freon gas. The greatest advantage of using self-contained Freon units is greater flexibility. Where trucks are stationed permanently at one plant, the ammonia system for refrigeration is the most satisfactory. A standardization of design and dimensional requirements of truck bodies would be an important step ahead for the dairy industry.   C. J. Babcock

634. Personnel practices of dairies in Alabama. E. STEELE, W. R. MYLES, and S. C. McINTYRE. Sou. Dairy Prod. J., **57**, 3: 24. 1955.

Thirty-seven dairies make greater use of training programs, records of exit interviews, suggestion plans, and outside consultants than do 464 industrial plants surveyed in Alabama. The other programs are more widely used by the industrial plants. These include a safety program used by 78% of the dairies, interviews of persons leaving employ 71%, a procedure for employees to air grievances 61%, unionization in part or in whole 35%, personnel research 25%, a full-time personnel manager and a personnel department 22%, and posting vacancies 19%. Eighty-four % of the dairies have a group insurance plan, 71% furnish some health and medical treatment, 19% have a pension plan other than social security, 16% have a credit union, 3% furnish some type of food service, and 3% rent houses to their employees.
                                          F. W. Bennett

635. Personnel practices of dairies in Alabama. E. STEELE, W. R. MYLES, and S. C. McINTYRE. Sou. Dairy Prod. J., **57**, 1: 26. 1955.

Twenty-two % of 37 dairy plants surveyed in Alabama and 34% of 464 industrial plants of all types in the state have full-time personnel workers. Thirty-five % of the dairies and 43% of all plants are unionized. Ninety-seven % of the dairy plants interview applicants when hiring, 89% request references, 69% use an application blank, 54% use hiring ages ranging from minima of 16-25 yr. to maxima of 29-65 yr. and 19% use tests. Routemen in most plants are on a commission basis and other workers are paid by the hr. Eighty-three % consider seniority, 67% use incentive plans of pay, 26% work more than one shift, 22% have a job evaluation program, 16% have a profit sharing program and 15% use a systematic employee rating system. The smaller av. size of dairy plants than that of other plants probably accounts for less use of tests, less job evaluation, and less employee rating systematically. Technological characteristics of the dairy industry explain a lower proportion using multiple shifts.   F. W. Bennett

636. **Personnel practices of dairies in Alabama.** E. Steele, W. R. Myles, and S. C. McIntyre. Sou. Dairy Prod. J., **57**, 4: 26. 1955.

The four key factors affecting personnel practices of Alabama dairies are company structure, personnel specialization, size of plant, and unionization. The first of these appears to have the greatest impact and the last the least impact. All the factors usually influence policy in the same direction.   F. W. Bennett

637. **A study of milk packaging costs.** F. V. Solzan, Edward B. McClain Co., Memphis, Tenn. Sou. Dairy Prod. J., **57**, 2: 28. 1955.

On the basis of quarterly reports from 60 plants, the author concludes that paper packaging of milk costs 1¢ more and paper selling and delivery costs 1.2¢ less per "cost factored qt." than packaging in glass. Cost for each type was lowest when only one type of package was used.   F. W. Bennett

638. **We doubled inspection performance with a caser.** A. Geiss, Bowman Dairy Co., Chicago. Food Eng., **27**, 6: 97. 1955.

The installation of an automatic caser for gal. jugs of milk at Bowman Dairy Co.'s River Forest, Ill., plant produced several benefits. Men freed from the heavy manual packing job now have more time for final quality inspection of the filled jugs. Breakages have been reduced since there is less bumping of jugs with the mechanical unit. The caser is pneumatically operated and packs four 1-gal. jugs to a case.
      T. J. Claydon

639. **Impact of the dairy industry on foreign relations.** J. H. Stambaugh. Sou. Dairy Prod. J., **57**, 1: 70. 1955.

More American investment abroad, greater convertibility of currency, freer trade, and increased sharing of technical knowledge are essentials to better foreign relations and the maintenance of other nations as members of the democratic world.   F. W. Bennett

640. **How to get better results from your soliciting crew.** D. Burger, Carnation Co., Houston, Tex. Sou. Dairy Prod. J., **57**, 1: 25. 1955.

A soliciting crew which has been properly selected, trained, and supervised will afford valuable sales training for prospective route salesmen and will add new customers at a low and measurable cost. Every Carnation routeman does at least one week of soliciting before operating a retail route. This practice eliminates applicants who are not likely to be good solicitors. A 3-day training program consists of realistic training in selling, handling objections, and actual solicitation under supervision. New crews compare experiences after the first day of soliciting. Four to six men may work satisfactorily from one truck. Duplicate calls or omissions should be avoided systematically. Some common problems may be solved by

putting lagging men to work first, setting goals to be reached before coffee breaks, approval, or disapproval of customers by routemen, and aid in solving personal problems.

Carefully selected women solicitors have been successful in Houston. An age range of 30 to 35 yr. has been most satisfactory. Training is given in groups with men. Particular attention has been given to newcomers and to those moving into new homes in soliciting.
      F. W. Bennett

641. **Outdoor vending machines.** W. Cantley, Land O'Lakes Creameries, Minneapolis, Minn. Sou. Dairy Prod. J., **57**, 5: 27. 1955.

Twenty-six outdoor milk vendors are operated and serviced by 2 drivers and a relief driver. Space rental is $\frac{1}{2}$¢/$\frac{1}{2}$ gal. including sufficient attention to see that the machines are operating properly and reporting need for serviceman. The av. daily volume is 147$\frac{1}{2}$ gal. including Sundays and holidays. Dependable equipment which maintains a temperature of 33-38° F. is used. Locations are selected for availability of change, clean, spacious, well lighted surroundings; clean, courteous attendants; open as near 24 hr/day as possible and in the center of a population of 4,000 within a mile. Need for this type of outlet is believed to be the principal inducement to customers. A few machines cannot be operated economically. One hundred $\frac{1}{2}$-gal/day should be the minimum volume after 6 mo. of operation. Sales through vending machines are entirely plus business.   F. W. Bennett

642. **Plastic bodies for refrigerated trucks.** Anon. Milk Prod. J., **46**, 7: 22. 1955.

Advantages claimed for plastic bodies for refrigerated trucks are: (1) permits construction of a more sanitary vehicle, (2) reduces costs of maintenance of bodies, (3) helps to reduce refrigeration costs by 50% because these materials act as insulators, (4) lighter weight, (5) does not require repainting since the color is impregnated into the material and will not corrode or fade.   J. J. Janzen

FEEDS AND FEEDING

643. **Some factors affecting the stability of carotene in mixed foods.** A. W. Halverson and C. M. Hendrick, S. D. Agr. Expt. Sta., Brookings. Tech. Bull. 14. 1955.

Carotene storage losses were observed in a corn-soybean basic diet during a 30-day, 37° C. storage period. The experiment included diet modifications as well as different carotene supplements. Diet modifications were made by adding separately any one of the following common feeds: 15% of oats, wheat bran plus middlings, dried buttermilk, meat scraps or fish meal, 1 and 4% ground limestone, 1% steamed bone meal, and 2% urea or trace minerals.

The additions caused carotene losses in many instances with greater losses being experienced

with oil supplements than with alfalfa. However, since oil carotene was more stable than that of alfalfa in unmodified diets, the extra losses induced in oil supplemented diets by modification caused an equalization of losses among supplements. Meat scraps, when used at low levels in the basal diet plus alfalfa mixtures, increased carotene stability. Comparison of losses among freshly mixed diets and those aged 2, 4, and 6 mo. before the stability test period showed that losses were not greatly affected by aging in the basal diet or when meat scraps or normal trace mineral levels were present. However, losses increased markedly in the high trace mineral diets following aging. The age-induced loss generally approached maximum at 2 mo.   R. W. Hunt

GENETICS AND BREEDING

644. Storage of boar and stallion spermatozoa in glycine–egg yolk medium. A. ROY, A.R.C. Unit of Reproduction, An. Research Sta., Cambridge. Vet. Record, **67**, 18: 330. 1955.

Previously (Nature **174**: 746. 1954.), Roy and Bishop reported that an extender made up of equal parts of 3% glycine and egg yolk improved survival of bovine spermatozoa stored at 4° C. This report presents the results on the use of glycine–egg yolk extenders for the storage of boar and stallion spermatozoa. Survival of boar sperm was better in glycine extenders than in citrate extenders, with best results obtained with the 4.5% glycine–egg yolk extender. Glycine improved motility after storage when boar semen was used directly or when it was centrifuged to remove seminal plasma, and then resuspended with a higher concentration of semen. Boar semen in 4.5% glycine–egg yolk extender was successfully stored at −79° C. with the addition of glycerol. Sperm motility was also improved when glycerol was added to the extended semen stored at 4° C. Stallion semen responded similarly to boar semen.
                                        R. P. Niedermeier

645. The evaluation of bull semen. M. W. H. BISHOP and J. L. HANCOCK, An. Research Sta., Cambridge. Vet. Record, **67**, 20: 363. 1955.

This paper reports the results of 11 different semen evaluation tests applied to samples of semen collected at 4 different A.I. centers. A total of 121 samples of semen was examined from 76 bulls of 7 breeds. Conception based upon a 90-day nonreturn rate was used as the fertility index. Relationships between the different test characteristics and fertility were studied statistically.

Results suggest that the most useful measures of the fertility of bull semen are the number and % of living spermatozoa present and their physical activity. Tables are presented showing interrelationships of semen characteristics studied.   R. P. Niedermeier

HERD MANAGEMENT

646. The use of a self-feeding trench silo for dairy cattle. M. RONNING, Okla. Agr. Expt. Sta., Stillwater. Mimeo. Cir. M-262. 1954.

A diagram is provided, showing the construction of a trench silo.   R. W. Hunt

ICE CREAM

647. Low-fat dairy foods. C. D. DAHLE, Pa. State Univ., University Park. Ice Cream Field, **65**, 5: 78. 1955.

The author gives in outline form a compilation of pertinent data relative to ice milk and related products.   W. C. Cole

648. What food dealers want from ice cream makers. ANON. Ice Cream Field, **65**, 5: 30. 1955.

The food store is the outlet through which the ice cream manufacturer sells more than half of his output, and ice cream is a source of volume and profit to the food store. The results of a recent survey are summarized. At 5 super markets in Cleveland, Ohio, it was found that ice cream contributed 0.70% to total sales and 1.2% to total store profit.

It is claimed that price alone no longer is the prime consideration of purchase. The manufacturer should supply consistently good quality products attractively packaged. Variety of flavors with different sized packages are important.

Prompt, efficient service and good merchandizing assistance are expected by food stores. The manufacturer who does the best job of helping the retailer can expect to sell more ice cream.   W. C. Cole

649. A study of industry trends based on a nationwide survey by Ice Cream Field, Ice Cream Field, **65**, 2: 35. 1955.

The results of this survey are presented by charts and discussion. Data are given for 1953 and 1954 with estimates for 1955.

The estimate for 1955 production is: bulk 34.86%, packaged 48.70%, and novelties 16.44 %. Of all packaged ice cream in 1954, 48.87% was in ½-gal. cartons, 39.17% in pt., 8.95% in qt., 1.85% in ½-pt., and 1.16% in gal.

Sales of ice cream by outlets for 1954 were found to be: food stores 50.34%, drug stores 12.59%, restaurants 11.85%, confectioners 7.16 %, company-owned stores 6.33%, home delivery 3.69%, street vending 1.95%, and all others 6.11%. Figures are also given for promotion plans and advertising media.

The survey also showed a decrease in the price of ice cream during 1954. It was estimated that of the total 1955 sales, ice cream would show a percentage decrease, mellorine a percentage increase with ice milk remaining about the same.   W. C. Cole

650. Stabilizers and HTST. G. D. Sperry, Kelco Co., San Diego, Calif. Ice Cream Field, **65**, 6: 80. 1955.

Stabilizer incorporation in ice cream mixes where HTST pasteurization is practiced presents some problems not necessarily encountered where the batch pasteurization method is employed. Dissolving the stabilizer in water before adding it to the balance of the mix is common practice with HTST pasteurization. Various methods of preparing pre-solutions of stabilizers are considered depending upon the type of equipment available.

Where dry addition is desired, the stabilizer should be readily dispersible in the cold mix ingredients; and it should be soluble at low temperatures so as to assure complete solution in the HTST system. If all mix ingredients, except the stabilizer are liquid, the author recommends a high speed recirculation system for stabilizer incorporation. Satisfactory results were obtained with this method of incorporation using several different types of stabilizers.

It is concluded that somewhat greater efficient action of the stabilizer is likely if it is added as a pre-solution. If dry addition is used, the stabilizer should be allowed to hydrate for at least 15 min. or the mix should be preheated.

W. C. Cole

651. Research yields way to boost skimmilk solids. J. J. Sampey, Abbotts Dairies, Inc., and C. E. Neubeck, Rohm and Haas Co., Philadelphia, Pa. Ice Cream Field, **65**, 3: 24. 1955.

It is claimed that with a butterfat content of 10% or less, ice cream should contain 13% to 14% milk-solids-not-fat. The likelihood of developing sandiness under these conditions is great, unless preventive measures are taken. Enzyme hydrolysis of lactose is given as one method of preventing sandiness.

Data are presented to show that at 40° C. yeast lactase is active at pH 6.0 to 6.5, whereas at lower temperatures the pH is less critical. The optimum temp. for hydrolysis periods of approximately 15 min. is 45° C., whereas 40° C. is best for periods of 30 min. or longer. Hydrolysis of 15% to 20% of the lactose in skimmilk concentrates is considered adequate in most cases.

The lactose in sweetened condensed milk can be hydrolyzed, but at a slower rate than where the lactose concentration is higher. Hydrolysis to the desired degree can take place at low temperature, i.e. 4° C. during storage for the required length of time.   W. C. Cole

652. Good sherbets. G. M. Illes, A. E. Illes Co., Dallas, Tex. Ice Cream Field, **65**, 5: 50. 1955.

The author gives in some detail the major factors contributing to the manufacture of good sherbet.   W. C. Cole

653. Making a good soft-serve mix. S. J. Werbin, Stein, Hall & Co., New York, N. Y. Ice Cream Field, **65**, 5: 60. 1955.

As compared with regular mixes the author recommends relatively high milk-solids-not-fat, somewhat lower sugar content with slightly lower stabilizer but higher emulsifier contents for soft-serve mixes. Glycerol monostearate type emulsifiers are less likely to contribute to "buttering" of the mix in the freezer than the polyoxyethylene type.   W. C. Cole

654. Problems of the soft-serve operator. IV. Common defects of soft-serve products. P. E. Piper. Ice Cream Rev., **38**, 12: 66. 1955.

The author discusses the factors responsible for some of the common flavor and body defects which may be encountered with soft-serve products. This information should prove of value to every soft-serve operator and to mix suppliers as well.   W. J. Caulfield

MILK AND CREAM

655. Quality milk from bulk milk tanks. R. P. March and M. Rulison, N. Y. State Agr. Expt. Sta., Geneva. Farm Research, **21**: 2. 1955.

In February 1955, 700 farms in New York State were handling their milk under the bulk system. This constitutes a ten-fold growth since 1952. This study was undertaken to determine the best method of operating bulk tanks. It was found that in order to assure thorough mixing of the butterfat, the tank agitator should be turned on at least 3 min. before the butterfat sample is taken. Mixing warm milk with milk which has been previously cooled, is not likely to result in rancid milk, nor is the agitation which the milk receives in the tank a cause of rancid flavor. Slow cooling of milk with continuous agitation produces some churning. If a large portion of milk in the tank becomes frozen, cream flakes are apt to develop.

R. W. Hunt

656. A market survey of the fat content of fluid dairy products and cottage cheese. T. V. Armstrong and I. A. Gould, Ohio State University, Columbus. Milk Dealer, **44**, 10: 43. 1955.

Data are presented to show that the tests of 70% of the samples on both regular pasteurized and homogenized milk agreed within ±0.1% with the stated standard fat, 18.9% varied within the limits of ±0.2%, and the remainder, or 11.1%, fell within the limits of ±0.4%. The tests of fat in coffee cream indicated that 30.5% of the samples tested the same as the standard, with 22% testing lower and 47.5% testing higher than the standard. With whipping cream 15% of the tests agreed with the standard, whereas 35% tested lower and 50% tested higher. The tests of cottage cheese showed that the fat content of the large curd cheese varied between the limits of 2.22% and 4.19%, and

of the small curd variety between 2.83% and 6.92%. These results predicate the necessity for each plant to have an active control program designed for maintaining uniformity and accuracy of the composition of the products.

C. J. Babcock

657. Bulk pick-up butterfat sampling. H. J. PRESTON, USDA. Milk Plant Monthly, **44**, 7: 27. 1955.

A study designed to evaluate the present butterfat sampling procedures used in the bulk handling of milk showed that milk in farm bulk tanks when agitated for a minimum of two min. was blended adequately. Differences in tests between selected positions in the farm tanks usually were not significantly larger than the difference in tests between duplicate samples from a single position. A sample for testing purposes would be more representative if it consisted of several small samples from different positions rather than one large sample from one position.

For refrigerated samples the composite tests averaged 0.028% lower than the av. of fresh samples for the comparable 10-day period. The refrigerated composite was 0.038% less than the av. of the fresh samples for the comparable 15-day period. For nonrefrigerated samples, the tests of composites averaged 0.065 and 0.064% less than the av. of fresh samples for the comparable 10- and 15-day periods, respectively. The composite test of producers with an av. butterfat test of 4.67% was 0.09% below the av. of the fresh tests for the 10-day period. For producers with an av. test of 3.27% the composite test was only 0.016% below the av. of the fresh test for the 10-day period. Data are presented showing comparison between butterfat test results from fresh, 10-day, and 15-day composites, and between refrigerated and nonrefrigerated composite samples.

C. J. Babcock

658. The manufacture of sour cream. E. S. GUTHRIE, Cornell Univ., Ithaca, N. Y. Sou. Dairy Prod., J., **57**, 2: 26. 1955.

Sour cream of good quality may be made by standardizing the cream to 18% fat, pasteurizing at 165° F. for a minimum of 10 min. to a maximum of 30 min. for raw cream, homogenizing with a single valve twice at 165° F. and 2,500 lb/sq in., cooling to 72° F., adding 1 pt. of young starter to 10 gal. of cream, adding 5 ml. of rennet in 15 ml. of cold water per 100 gal. of cream while being stirred, drawing into ripening containers, holding at 72° F. for 15 hr., chilling without agitation to 35-40° F. and stirring with about 10 cycles of the agitator just before distributing into consumer packages.

F. W. Bennett

659. Whole can cream filter solves sediment testing problem. ANON. Milk Prod. J., **46**, 7: 20. 1955.

A whole can cream filter has been developed through the joint action of the Food and Drug Liaison Committee of the American Butter Institute and the Food and Drug Administration.

The unit operates by sucking cream from the can, pumping it into a tulle filter head and then discharging it into a receiving pan or tank. If collected sediment is so great, or of such a nature to demand rejection, a valve on the tank is opened and the cream flows to a waste outlet. The tulle disc is then sent to the farmer or buyer who shipped the cream for observation and study.

This method has resulted in great saving in time and labor. With wider acceptance of these cream filters a definite improvement in the quality of cream offered for sale by both producers and cream buyer are expected. Wherever tried to date, such has been the case.

J. J. Janzen

NUTRITIVE VALUE OF DAIRY PRODUCTS

660. Milk as a food throughout life. M. H. IRWIN, Wis. Agr. Expt. Sta., Madison. Bull. 447. 1954.

This is a well written, informative publication which tells why we should drink milk and what portion of our nutritive needs are provided by this food.

R. W. Hunt

661. Low sodium milk usage spreads. T. D. LEATHERS. Milk Plant Monthly, **44**, 7: 21. 1955.

Many persons suffering from certain heart ailments, hypertension, and kidney disorders, who in the past had to give up milk, now are able to drink a new fresh, fluid milk called "Lo-Sodium" milk. The process by which the milk is made is known as ion-exchange and the product contains about 50 mg. of sodium as compared to 500 mg. in unaltered milk.

C. J. Babcock

662. What vitamin D fortification means to the dairy industry. K. G. WECKEL, Univ. of Wis., Madison. Sou. Dairy Prod. J., **57**, 6: 40. 1955.

Studies made 25 yr. ago, when vitamin D milk first was made available, indicated the incidence level of rickets to be 50-90%. Vitamin D milk is largely responsible for the extreme rarity of rickets and a tremendous reduction in dental caries today. The development of this product was one of the most important developments which has brought nation-wide attention to milk as a food. The methods of producing vitamin D milk involving the feeding of irradiated yeast or irradiating the milk itself have practically been displaced by the addition of vitamin D concentrates in sterile canned milk. About 30-50% of the bottled milk in smaller communities and 90% in the larger metropolitan areas is fortified with vitamin D.

F. W. Bennett

SANITATION AND CLEANSING

663. Cleaning stainless steel sanitary lines in place. C. G. FORTNEY, M. P. BAKER, and E. W. BIRD, Iowa Agr. Expt. Sta., Ames. J. Milk Food Technol., **18**, 6: 150. 1955.

A comparison of sanitization procedures was made on pipelines cleaned in place vs. dismantling and brushing. Different temperatures, velocities, and cleanser compositions were studied. The microbial content of the milk from CIP lines was comparable to that from hand-cleaned lines.

Bacteriological results from CIP lines (150° F. or higher for 20 min.) were lower than those of HC lines and showed no coliform contamination.

A velocity of 7 ft/sec at 130° F. for 10 min. showed better results on internal surfaces of cold milk lines than a velocity of 2 ft/sec at the same temperature.

Temperature in CIP procedures has more effect upon cleaning efficiency than either time or velocity, when the recirculating time is 20 min. or longer.

Cleaner A containing the highest concentration of polyphosphate in solution gave the best physical cleanliness. Cleaner D, a chelated caustic, gave results comparable to A.

H. H. Weiser

664. How benefits outweigh the problems in cleaning pipelines in place. A. V. GEMMILL, SR. Food. Eng., **27**, 6: 59. 1955.

The CIP cleaning of 800 ft. of pipeline in a 3-story milk plant presented various problems. The article describes details of the system developed to accomplish the task satisfactorily.

T. J. Claydon

665. Fly and insect control. II. Protecting cows in the barns of dairy farms. ANON. Milk Prod. J., **46**, 7: 28. 1955.

Two general methods of insect control often recommended are: (1) elimination of such breeding places as the manure piles, and (2) the systematic killing of large numbers of flies.

The first method concerns itself with the outside surroundings. The use of DDT is recommended as long as it gives satisfactory control. When it fails, the use of lindane, chlordane, methoxychlor, or toxaphene is suggested. Once the outside breeding places have been minimized attention should be focused on mass killing inside the barn.

The second control measure includes the use of residual sprays and space sprays. The residual spray may be used when animals are not present and precautions are taken not to get materials into feed troughs. Suitable space sprays, "Pyrenone" (combination of piperonyl butoxide and pyrethrum), which are nontoxic to man, animals and animal products, are available.

The space spray has been found to be effective. Spraying cows as they enter the barn or just prior to milking while in the barn are suggested procedures. Some farms are equipped so that cattle receive a spray on their way to the watering trough. Such treatments often have lasting effects up to 10 days.

J. J. Janzen

666. Experiences with a new type of dairy waste treatment. K. L. SCHULZE, Green Bay, Wis. Sou. Dairy Prod. J., **58**, 2: 28. 1955.

A 2-stage cavitator system of handling dairy wastes is described. With an aeration tank capacity of 6,000 gal., air supply of 36 c.f.m. or 52,000 cu. ft/day, two 5 h.p. motors, a raw feed rate of 5-8.3 g.p.m. continuously, clarifier capacity of 1,100 gal., pumpage of 7,200-12,000 gal/day and biochemical oxygen demand (B.O.D.) load of 58-170 lb/day, the B.O.D. removal ranged from 98.7% to 93.6%. The sludge return was 1-2 g.p.m. and the sludge removal 100-200 gal/day. Operation and maintenance require about 0.5 hr/day and no trained personnel.

F. W. Bennett

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