

# JOURNAL OF DAIRY SCIENCE

*File*

Public Health Service  
Division of Sanitation

## Contents

<i>Type, Body Size and Breeding Efficiency of Ayrshire Cow Families.</i> KENNETH A. TABLER, W. J. TYLER AND GEORGE HYATT, JR. ....	95
<i>Properties of the Colostrum of the Dairy Cow. VI. Creaming and Rate of Flow.</i> E. G. MOODY, G. H. WISE, D. B. PARRISH AND F. W. ATKESON .....	106
<i>Effects of Added Hyaluronidase and Hyaluronic Acid on the Motility of Bull Spermatozoa.</i> JAMES E. JOHNSTON AND JOHN P. MIXNER .....	116
<i>Improved Methods in the Recovery of Lactose Using Ion Exchange Resins.</i> E. D. MCGLOSSON AND J. C. BOYD .....	119
<i>A Comparison of U. S. No. 2 Field-cured Field-baled Alfalfa Hay with Artificially Dried and Ground and Pelleted Alfalfa Hays as a Source of Carotene and Roughage for Holstein Calves.</i> H. D. EATON, C. A. CARPENTER, R. J. CAVERNO, R. E. JOHNSON AND F. I. ELLIOTT .....	124
<i>A Comparison of Dry Sweet Cream Buttermilk and Non-fat Dry Milk Solids in Breadmaking.</i> J. V. REGER, W. B. COMBS, S. T. COULTER AND R. B. KOCH .....	136
<i>B-Vitamin Studies in Calves. I. The Relation of Age of Calf and Levels of Thiamine, Riboflavin and Pantoic Acid Found in the Digestive Tract.</i> E. E. KNODT .....	145
<i>A Comparison of Comfort and Tie-chair Use in the Milking of Dairy Cows.</i> D. PORTERFIELD, GEORGE HYATT, JR., D. P. F. HOUSE .....	149
<i>Type Differences and Blood Antigens in the Jersey Cattle Herd.</i> A. A. DUNLOP .....	154
<i>Abstracts of Literature</i> .....	A11
<i>Association Announcement</i> .....	A17



ANNUAL MEETING  
JUNE 6-8  
KNOXVILLE, TENN.

Vol. XXXIV, No. 2, February, 1951

Published by the  
AMERICAN DAIRY SCIENCE ASSOCIATION

แผนกห้องสมุด กรมปศุศาสตร์  
กระทรวงเกษตรและสหกรณ์

**DEPEND ON**

**Duraglas**  
**MILK BOTTLES**

*for sky-high quality  
and down-to-earth  
economy!*

- Dairies owe it to their operation to prove the true economy arising from the quality in a returnable container.

For uniformity of quality, for year-in-year-out trippage performance, for the durability and attractiveness of Applied Color designs and sales messages, Duraglas dairy containers are unequalled.

And behind the Duraglas name stands the assurance of continued advancement. Nowhere else in the industry are such facilities of research, quality control and marketing experience combined to serve dairies.

From half pints to gallon jugs, it will pay dairies to let Owens-Illinois supply all their container needs.

### Applied Color Labels

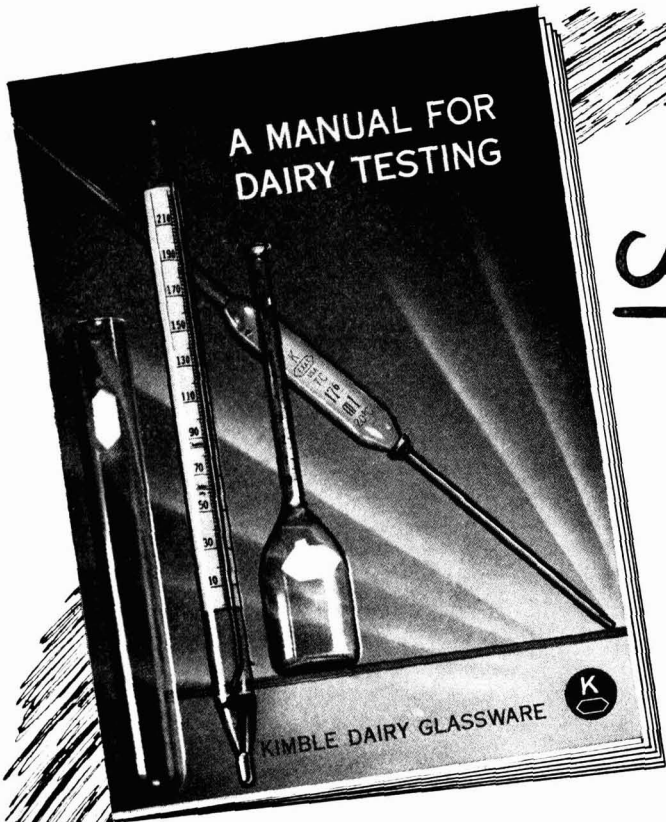
Now that it's time for dairies to sell again, the advertising power of these glass billboards deserves special attention. Owens-Illinois has more than 1500 designs and messages standing ready plus artists skilled in adapting dairies' needs to ACL technique.

# Duraglas Dairy Containers

TRADE MARK REG. U.S. PAT. OFF.

OWENS-ILLINOIS GLASS COMPANY • TOLEDO 1, OHIO • BRANCHES IN PRINCIPAL CITIES

Your advertisement is being read in every State and in 45 Foreign Countries



*Just out!*

**4th edition  
of the  
KIMBLE  
MANUAL**

**—Tells how to test dairy products accurately**

If you sample or test dairy products you'll want the new 4th edition of the famous Kimble Dairy Manual.

Specially prepared after months of research, the Kimble Manual for Dairy Testing gives information on

the Babcock test; how to test milk, cream, butter, cheese, ice cream; tests for milk with sugar or chocolate; eight others. Also pertinent tables and a listing of Federal and State Standards for dairy products.

**KIMBLE GLASS** TOLEDO 1, OHIO  
Division of Owens-Illinois Glass Company



**Send for your free  
copy today**

*MAIL THIS* →

Kimble Glass  
Division of Owens-Illinois Glass Company  
Toledo 1, Ohio

Please send me a *free* copy of the new Kimble Dairy Manual.

Name \_\_\_\_\_

Address \_\_\_\_\_

City \_\_\_\_\_ State \_\_\_\_\_

Your advertisement is being read in every State and in 45 Foreign Countries

# JOURNAL OF DAIRY SCIENCE

OFFICIAL ORGAN OF  
AMERICAN DAIRY SCIENCE ASSOCIATION

Published at  
NORTH QUEEN ST. AND MCGOVERN AVE., LANCASTER, PA.

F. E. NELSON, *Editor*  
Ames, Iowa

*Associate Editors*

F. J. DOAN State College, Penn.	P. R. ELLIKER Corvallis, Ore.	I. A. GOULD Columbus, Ohio	H. A. HERMAN Columbia, Mo.
L. A. MOORE Beltsville, Maryland	W. V. PRICE Madison, Wis.	I. W. RUPEL College Station, Tex.	G. H. WISE Raleigh, N. C.

*Committee on Journal Management*

G. H. WISE, <i>Chairman</i>	
J. K. LOOSLI	F. E. NELSON, <i>ex officio</i>
W. V. PRICE	P. R. ELLSWORTH, <i>ex officio</i>

*Subscriptions.* Price, \$10.00 per volume in North and South America; \$10.50 in all other countries. Prices are net, postpaid. New subscriptions and renewals are entered to begin with the first issue of the current volume. Renewals should be made promptly to avoid a break in the series. Subscriptions should be sent to P. R. Ellsworth, The Ohio State University, Columbus 10, Ohio.

Subscriptions for the British Isles and British Empire, except for Canada and Australia, should be ordered through our agents: Messrs. Bailliere, Tindall and Cox, 7 and 8 Henrietta Streets, Covent Garden, London, W. C. 2, England. Subscriptions for Australia should be sent to our agent: John H. Bryant, Herbert St., St. Leonards, N. S. W., Australia.

*Advertising* copy should be mailed to P. R. Ellsworth, The Ohio State University, Columbus 10, Ohio. Advertising plates or cuts should be mailed direct to the Business Press, Inc., N. Queen St. and McGovern Ave., Lancaster, Pennsylvania.

*Post Office Notices* of undeliverable copies and changes of address should be sent to P. R. Ellsworth at the address above stated.

OFFICERS OF THE ASSOCIATION

R. B. BECKER, <i>President</i> Gainesville, Fla.	P. R. ELLSWORTH, <i>Sec.-Treas.</i> Columbus, Ohio
H. A. BENDIXEN, <i>Vice-President</i> Pullman, Wash.	F. E. NELSON, <i>Journal Editor</i> Ames, Iowa

DIRECTORS

F. J. ARNOLD Ames, Iowa	P. R. ELLIKER Corvallis, Ore.	J. H. ERB Columbus, Ohio	H. B. HENDERSON Athens, Ga.
L. A. MOORE Beltsville, Maryland	G. M. TROUT East Lansing, Mich.	C. W. TURNER Columbia, Mo.	

Entered as second-class matter April 13, 1934, at the postoffice at Lancaster, Pa. under the act of March 3, 1879.



**USING OUR BEAN TO BEST ADVANTAGE!**



Nature's a pretty grand gal and we're among her most enthusiastic rooters. On our Puerto Rico experimental plantation, we do everything possible to coax the world's finest vanilla beans out of her . . .

But it takes a solid assist from science before the true potentialities of the vanilla bean can be realized. (Here's where Michael magic comes in!) By artistic blending of selected vanilla crops from Puerto Rico, Mexico and Madagascar, we balance the qualities and characteristics of each and achieve a high degree of uniformity. By adding vanillin (derived from tropical spices), we reinforce the fragile delicacy of true vanilla flavor and aroma and capture that piquant subtlety which is the secret of Mixevan's superior appetite appeal.

We suggest you try your own experiment. Make up a batch of vanilla ice cream, using Michael's Mixevan. Taste the difference!

*America's Favorite*

**Michael's  
Mixevan**

The Powdered Vanilla with the Locked-In Flavor

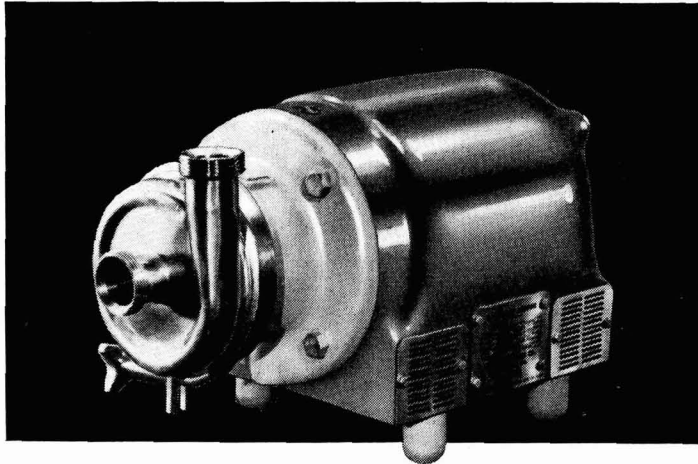
The same painstaking "know-how" goes into other fine Michael's products for special uses:

**TRUE VANILLA SUGAR**  
for custom made ice cream  
**TWIXT** for popular-priced ice cream  
**CREST** for use in novelties

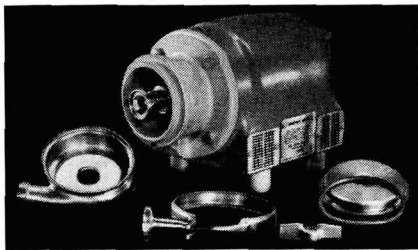
**DAVID MICHAEL & CO., Incorporated**  
*Half a Century in the Flavoring Field*  
**3743-63 D STREET • PHILADELPHIA 24, PA.**

For further information and prices see our representative or write direct

Your advertisement is being read in every State and in 45 Foreign Countries



## Look at the New FlexFlo Sanitary Pump



The new Flexflo disassembles easily for cleaning—is just as readily reassembled.

**Send for Information.** Ask your Cherry-Burrell representative or write direct to general offices for more information on the new Flexflo.

Flexflo pumps—long the leading sanitary pumps on the market—now offer still more advanced design features that mean higher efficiencies, greater sanitation, easier cleaning and less maintenance. The new Flexflo involute pump body enables them to pump the same volume with less horsepower. They are equipped with the new type sanitary motor. This pump meets 3A Code standards. Capacities to 70,000 lbs. per hour.

### 6 IMPORTANT IMPROVEMENTS

- 1 New Balanced Sanitary Rotary Seal—longer lasting.
- 2 Heavier Construction—less danger of crushing or distortion. Better gasketing—seals with less pressure.
- 3 All Welded—no soldering.
- 4 More Sanitary Stainless Steel Construction—easy to clean—no sharp corners or hard-to-reach surfaces.
- 5 New Sanitary Motors—totally enclosed and splash-proof.
- 6 Higher Efficiencies.

#### CHERRY-BURRELL CORPORATION

General Sales and Executive Office:

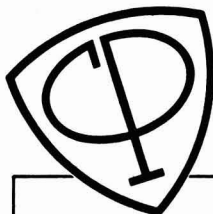
427 W. Randolph Street, Chicago 6, Ill.

Milk and Food Plant Equipment and Supplies

FACTORIES, WAREHOUSES, BRANCHES, OFFICES OR DISTRIBUTORS  
AT YOUR SERVICE IN 56 CITIES

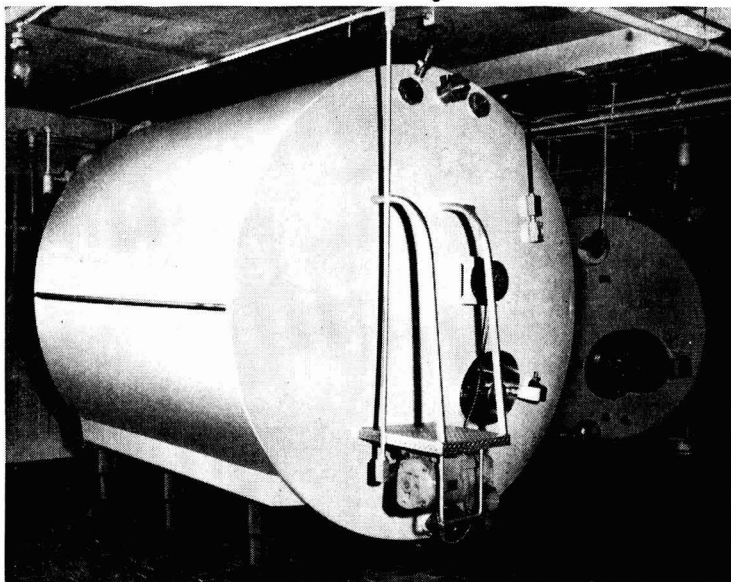


*trendmaker in a great industry*



# COLD HOLD

## *Storage Tank*



Here's the last word in cylindrical storage tanks designed to provide the efficiency and economy of combining COOLING and STORAGE in one compact unit.

The refrigerated section is built integrally into the bottom and sides of the tank. Its double stainless steel welded wall section with a smooth inside surface makes thorough

cleaning quick and easy. Outer wall is scientifically designed and fabricated for ample strength. Highest refrigerating efficiency is obtained with a completely flooded ammonia system.

CP Cold Hold Storage Tanks are offered in sizes from 1,000 to 5,000 gallons.

Write today for complete details.



**THE *Creamery Package* MFG. COMPANY**

**General and Export Offices: 1243 W. Washington Blvd., Chicago 7, Illinois**

Atlanta • Boston • Buffalo • Chicago • Dallas • Denver • Houston • Kansas City, Mo. • Los Angeles • Minneapolis • Nashville • New York • Omaha • Philadelphia • Portland, Ore. • St. Louis • Salt Lake City • San Francisco • Seattle • Toledo, Ohio • Waterloo, Ia.

**CREAMERY PACKAGE MFG. CO. OF CANADA, LTD.**  
267 King St., West, Toronto 2, Ontario

**THE CREAMERY PACKAGE MFG. COMPANY, LTD.**  
Avery House, Clerkenwell Green, London, England

DAIRY & FOOD  
PROCESSING  
EQUIPMENT  
•  
SUPPLIES  
•  
REFRIGERATION

# Johnson & Johnson CONTRIBUTES AGAIN TO QUALITY MILK PRODUCTION

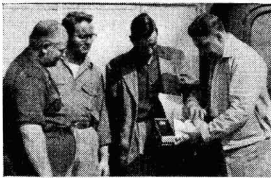
## With New, Improved Formula **RAPID-FLO**

Trade Mark of Johnson & Johnson

### Filter Disks and New Factory-Sealed Carton

The better, safer filtration reported by milk producers everywhere, plus the obvious sanitary advantages of the new Factory-Sealed Carton, have earned immediate and enthusiastic response from Dairy Industry leaders, including dairy farmers, haulers, fieldmen, plant managers and health authorities.

### Dairymen Everywhere Endorse New Formula RAPID-FLO Filter Disks in the New Factory-Sealed Carton

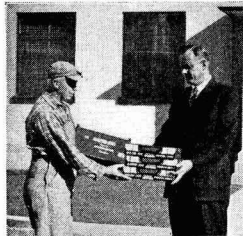


"Boy, it's a lot easier to handle and will surely keep the disks clean." Carlos R. Leffler, Puritan Dairy, Lebanon, Pa.

"This is what we have been looking for, for years." N. S. Rissler, Puritan Dairy, Lebanon, Pa.

#### THE "REAL McCOY"

"The new package is easy to handle and doesn't take up as much room in the cab as the old one did. My patrons think this new package is the real McCoy and they like the self-closing lid on dispenser." Arnold Edwards, Milk Hauler, Deerfield Creamery Co., Deerfield, Wisconsin.

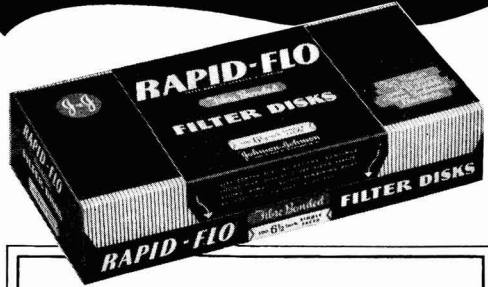


#### "JUST THE TICKET"

"My experiences with Rapid-Flo filter disks have proven their ability to do an exceptional job of filtering milk for their users. The new method of packaging has made it more attractive to producers in that they are not only convenient but provide protection for disks yet to be used. Milk haulers are also pleased with the new



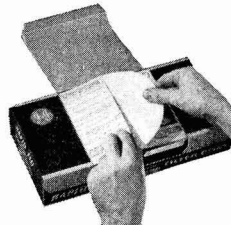
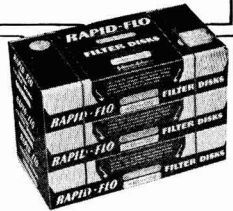
style of package. The Rapid-Flo metal dispenser box is just the ticket for proper storage of disks on producers' farms." Gene Florence, Field Supervisor, Dairyland Cooperative Assoc., Juneau, Wisconsin.



### THE ORIGINAL FIBRE-BONDED RAPID-FLO FILTER DISKS NOW MADE BETTER—SAFER, MORE RETENTIVE

Rapid-Flo, the first and best FIBRE-BONDED filter disk, has been improved with a new formula developed after several months of laboratory research and farm testing. More retentive, Rapid-Flo now removes finer microscopic extraneous matter than ever, providing an even more reliable Farm Sediment Check-Up, urged by milk authorities as an effective aid to Quality Milk Production.

The new Factory-Sealed Carton protects filter disks from factory to farmer. 100 Disks to a Carton. Can also be obtained in the 300 put-up. 3 cartons of 100 disks tied together as illustrated.



New Sanitary Dispenser—Made exclusively to hold the new Factory-Sealed Carton. Sturdy metal construction. Specially designed sloping lid closes automatically. Ask your supplier for this convenient sanitary dispenser.



The package is easy to carry, easy to open—closes with a touch of your finger-tip.

DAIRY FILTERS DEPARTMENT FILTER PRODUCTS DIVISION

# Johnson & Johnson

4949 WEST 65TH STREET CHICAGO 38, ILLINOIS

# JOURNAL OF DAIRY SCIENCE

VOLUME XXXIV

FEBRUARY, 1951

NUMBER 2

## TYPE, BODY SIZE AND BREEDING EFFICIENCY OF AYRSHIRE COW FAMILIES<sup>1</sup>

KENNETH A. TABLER<sup>2</sup>, W. J. TYLER AND GEORGE HYATT, JR.  
*West Virginia University, Morgantown*

The breeding of better dairy cattle involves selection for a number of desirable characteristics including type, size and breeding efficiency. Each of these may be related to production; however, the correlation between production and type is estimated to be low (6, 17). Nevertheless, the number of cows being classified for type is increasing because of the higher prices that breeders are willing to pay for the animals with higher type ratings. There is some evidence that live weight influences milk-energy yield as reported by Gaines (4, 5). Breeding efficiency also has some relationship to production since the lifetime production of a cow depends considerably upon the frequency with which she freshens. Therefore, most dairymen want cows that calve regularly in order to get the increased milk flow resulting from the physiological stimulation accompanying freshening as well as the additional returns from a larger calf population.

The purpose of this investigation was to compare the type, body size (as measured by height at withers and weight) and breeding performance of 19 Ayrshire cow families in the Reymann Memorial Herd to determine if the differences between cow families were significant. All the female descendants of each of the cows which were the foundation of the herd were considered as a cow family. The number of descendants of each foundation cow varied because the individuals in some generations had more female offspring than others. This, plus the differences in relationship of the sires to these families, tends to make the members of some families more closely related than others. Therefore, when comparing cow families, it is necessary to know the average genetic relationship between members of the same family. This must be corrected if selection is to be practiced between related families.

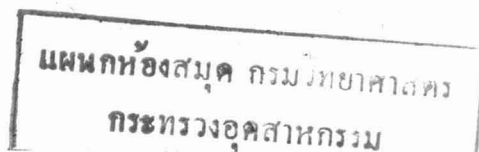
### REVIEW OF LITERATURE

A study of cow families by Bartlett and Margolin (1) showed that certain cow families transmit genetic factors for superior size (as measured by height at withers) and type. They also stated that the sires determined the conception rate of progeny in a family more than did the foundation cow. However, the number of cows in some of the families that they studied was small.

Received for publication May 25, 1950.

<sup>1</sup> Published with the approval of the Director of the West Virginia Agricultural Experiment Station as Scientific Paper No. 425.

<sup>2</sup> Present address: Department of Dairy Science, University of Illinois, Urbana.



In the Yellow Franconian breed, Kab (8) found that through several generations 35 families had high fertility and 11 families had low fertility. From the analysis of the fertility of the daughters of 22 bulls, he believed that the existing variations had a genetic basis.

Seath *et al.* (12) reported on some cow families that varied from 40.6 to 92.3 per cent in breeding efficiency and averaged 63.3 per cent. These percentages were based on one service per conception as 100 per cent. They indicated that breeding efficiency was inherited and should be given consideration when practicing selection.

Data on breeding efficiency of cow families in the University of Nebraska herd was studied by Trimberger and Davis (16). This included from four to seven families in each of the four major dairy cattle breeds. Among these 221 cows in 20 different families, they found significantly high and low fertility based on services per conception. Likewise, there were considerable differences in the conception rates among the daughters of 19 sires.

Several different methods of measuring the reproductive efficiency of dairy cattle have been proposed. Williams (18) adopted 2 yr. as the ideal age for a heifer to calve, which requires conception at 15 mo. if this ideal is to be attained. Every month thereafter was designated as a "breeding month" with 12 of them being considered as the ideal calving interval. He found the average number of breeding months required to produce a calf by dividing the total breeding months by the number of calves born. This was converted to a percentage by dividing 12 by the computed average number of breeding months per calf dropped. Williams and Carpenter (19), used this method to analyze the reproductive efficiency of a Guernsey herd that produced a calf for each 28.7 breeding months, which is only 41.8 per cent of the ideal.

Spielman and Jones (14) determined the reproductive efficiency for groups of dairy animals that were descendants of different foundation cows. They used a numerical expression based on the assumption that to be 100 per cent a heifer should be bred at a certain age, depending upon the breed, and that she should drop a calf every 12 mo. thereafter. The desired calving interval of 12 mo. credited each cow with twelve 100 per cent months per pregnancy. Any portion of the calving interval in excess of 12 mo. was considered as possessing 0 per cent reproductive efficiency. Cows removed from the herd before calving and known to be pregnant were allowed one 100 per cent month for each month of pregnancy. The number of 100 per cent months divided by the total number of reproductive months gave the per cent reproductive efficiency. This method evaluated incomplete reproductive cycles, while the one Williams used did not because he excluded such things as abortion. These workers concluded that a marked difference in reproductive efficiency existed between their various cow groups as well as between breeds.

#### EXPERIMENTAL PROCEDURE

The data for this study included the type, body size and breeding performance records of 19 cow families in the Reymann Memorial Herd of Ayrshires.

There were a total of 401 cows in these 19 families; however, the records on some of the animals were incomplete. The management of this herd was described in an earlier publication (15).

Two members of the West Virginia University Dairy Department scored the cows for type from 1935 to 1941. The herd first was classified officially by inspectors of the Ayrshire Breeders' Association in 1942 and since then, has been classified for type at least twice yearly. The first after-calving type ratings of 282 cows collected from these two sources were used, regardless of whether this was the cow's highest or lowest.

Data collected and analyzed on size at 1 mo. after first calving included the weights of 253 cows and the measurements for height at withers of 212 cows. The growth data of Bowling and Putnam (3) shows that the curves for weight and height at withers begin to level out at 26 to 30 mo. of age, about the time when most heifers in the Reymann Herd drop their first calves. A study by Morgan and Davis (10) reveals that Ayrshire cows seemed to reach a stabilized weight about 30 days after calving. According to Putnam and Henderson (11) the increase in body weight due to the second pregnancy is less than 2 per cent of the body weight up to the fifth month of gestation.

The per cent reproductive efficiency was determined for each of the 401 cows by calculating the number of months from first breeding as a heifer to 3 mo. past the last calving date, or 3 mo. past the next calculated calving date, provided the cow was in the last one-half of her gestation period before being culled from the herd. Then the number of 100 per cent months (one for each month of pregnancy plus three for each calf dropped) was divided by the total number of reproductive months, thus obtaining the per cent reproductive efficiency. These percentages then were converted into angles according to Snedecor (13) before being analyzed. This method differs from Spielman and Jones by not penalizing heifers that were not bred to freshen at a given age, because heifers often were left open until such a time that breeding would enable them to freshen in the fall months. Moreover, at the time these calculations were made, some cows were soon to freshen and they were given the full 9 mo. credit for gestation and the 3 mo. thereafter, thus completing their twelve 100 per cent months.

The length of service period was used as an additional criteria of breeding efficiency. This was measured by the interval between first service and conception, and the interval from calving to first service in terms of days. Bowling *et al.* (2) analyzed the breeding performance in this same herd and found that more services per conception were necessary when older sires were mated to heifers than when younger bulls were used. Rather than adjust for the age of the service sires, it seemed advisable to omit the breeding record on the number of days from first service to conception of virgin heifers. Thus, the first record on days from calving to first service and the second record on days from first service to conception were used. The number of services per conception also was analyzed, using the conception figures for the second breeding period.

Both families and subgroups (cows in the same family and by the same sire)

were analyzed by the analysis of variance (13) to test the significance of differences between families, between sires and between families within sires. The variations in the characteristics were divided into: (a) differences between families, (b) differences within families, (c) differences between sire-family subgroups which, in addition, were divided into: differences between sires and differences between families within sires, and (d) differences within subgroups.

#### RESULTS AND DISCUSSION

*Type.* The average type ratings of cows in each of the 19 cow families in this study are listed in table 1. These averages were based on a rating of excel-

TABLE 1  
*The number of cows and their average type rating for 19 cow families  
in the Keymann Memorial Herd*

Family	No. of cows	Average rating
A	9	86.4
B	16	85.0
C	17	84.9
D	23	84.2
E	19	84.1
F	30	83.8
G	21	83.5
H	9	83.1
I	13	82.9
J	7	82.5
K	7	82.5
L	12	82.1
M	9	81.9
N	9	81.9
O	26	81.7
P	6	81.6
Q	27	81.0
R	9	80.8
S	13	80.6

lent = 92.5, very good = 87.5, good plus = 82.5, good = 77.5, and fair = 72.5. Seventeen cow family averages ranged between 80.6 and 84.9 or an average of good plus. Two families, A and B, averaged 85.0 or more (very good).

The differences in the average type ratings of the 19 cow families were small, yet the variation between families was significantly larger than that within families (table 2). The differences between the daughters by different bulls were highly significant. These sire differences include both genetic and environmental variations. Environment here would include not only the yearly variations that may have occurred, but also differences between inspectors, as each sire's daughters were contemporaries and tended to be classified by a different inspector. The differences between families within sires, which is a measure of the variation between families after the sire differences have been removed, were not significant. Therefore, the significant differences in type found between these cow families are a result of both environmental changes and variation between the sires that were a part of the family structure. These results agree with the observations of Bartlett and Margolin (1).



TABLE 2

*Analysis of variance of type and body size on 19 cow families in the Reymann Memorial Herd*

Source of Variation	Type		Ht. at withers		Wt.	
	Degrees of freedom	Mean square	Degrees of freedom	Mean square	Degrees of freedom	Mean square
Between families	18	34.3*	18	24.5*	18	14,263
Within families	263	19.6	193	14.2	234	9,760
Between subgroups	197	23.8	137	14.7	171	10,592
Between sires	32	60.1**	17	22.2	26	22,718**
Between families within sires	165	16.7	120	13.6	145	8,418
Within subgroups	84	12.9	74	15.8	81	9,003
No. of cows in study	282		212		253	

\*\* P &lt; 0.01 Highly significant

\* P &lt; 0.05 &gt; 0.01 Significant

*Body size.* Data on the average body size as measured by height at withers and weight, taken at 1 mo. after first calving for the 19 cow families are given in table 3. The differences between families for wither height were statistically significant while the sire differences were not, as shown in table 2. These family differences may be due partly to changes in environment. The variation remaining after the sire effects were eliminated from the family differences was not significant. Consequently, the sires that were used on these cow families and environment seem to be largely responsible for differences in height at withers.

The variation between the family averages of body weight was quite large;

TABLE 3

*The number of cows and their average height at withers and weight taken at 1 mo. after first calving for 19 cow families in the Reymann Memorial Herd*

Family	No. of cows	Ht. at withers	No. of cows	Wt.
		(cm.)		(lb.)
B	12	128.3	15	1002
Q	16	128.1	18	1008
M	7	126.7	8	967
G	26	126.3	27	983
F	19	126.1	27	998
L	11	126.1	14	954
N	9	126.0	10	955
P	5	125.9	4	1001
S	12	125.8	14	1021
D	21	125.7	22	990
H	5	125.7	6	985
O	11	125.6	18	961
C	12	125.1	14	944
J	5	125.0	7	981
E	13	124.2	15	962
R	10	124.1	11	943
I	6	123.7	7	937
K	3	122.8	6	963
A	9	121.9	10	865

however, these differences were not significant, as may be seen in table 2. The differences between sires were highly significant, but again the variations may be due to environmental causes, as well as genetic differences between sires. The between-family within-sire variation was not significant.

*Breeding efficiency.* The average per cent reproductive efficiency for the 19 cow families in the Reymann herd are shown in table 4 and range from 77 per cent in the R family to 89 per cent in the O family. Table 5 lists the number of services per conception for the second reproductive cycle and the length of calving interval for each of the 19 cow families.

TABLE 4

*The number of cows and their average per cent reproductive efficiency for 19 cow families in the Reymann Memorial Herd*

Family	No. of cows	Reproductive efficiency
		(%)
O	31	89
E	24	89
Q	29	88
I	17	87
H	12	87
K	10	87
F	45	86
M	14	86
L	25	85
B	22	85
A	13	85
G	37	84
S	19	84
J	13	84
D	35	83
N	14	83
C	18	81
P	11	81
R	12	77

There was a considerable amount of variation between families in each of these three criteria of breeding efficiency; nevertheless, these differences were not large enough to be significant (table 6). For services per conception and the average number of days from first breeding to conception, there was more variation within families than between families. Although the differences between sires were highly significant, the service sire to which these cows were mated may have made these differences larger or smaller. The inherent reproductive ability of these cows cannot be separated from the effect of the service sire. Thus, the service sire and environment may have been responsible for some of the differences between the daughters of different sires.

*Components of Variance.* It is pertinent to this study to have an estimate of the sire, family, sire  $\times$  family interaction and error components of variance in order to evaluate the relative importance of each. Because of the disproportionate subclass numbers, mean unbiased estimates of the components of variance in these data were obtained by equating the sample sums of squares to the expected

TABLE 5

The number of cows and their average number of days from calving to breeding, breeding to conception and services per conception for the 19 cow families in the Keymann Memorial Herd

Family	No. of cows	Days from calving to breeding	Days from breeding to conception	Services / conception
S	16	91	30	1.6
P	11	84	39	1.6
J	11	89	28	1.8
E	24	82	30	1.9
G	30	92	38	2.0
A	13	94	46	2.0
R	10	98	37	2.0
C	17	97	36	2.1
Q	28	88	50	2.3
O	29	84	48	2.4
B	19	98	56	2.4
K	8	84	49	2.4
F	44	91	61	2.5
L	25	98	50	2.5
I	15	79	61	2.5
D	30	86	53	2.6
N	14	94	70	2.7
M	14	94	72	3.1
H	11	87	73	3.1

sums of squares as outlined by Henderson (7). The model assumed was for a two-way factorial classification and is

$$y_{ijk} = \bar{x} + f_i + s_j + fs_{ij} + e_{ijk}$$

where  $y_{ijk}$  is the observed value of a characteristic on one of several cows in the subgroup of the  $i^{\text{th}}$  family and the  $j^{\text{th}}$  sire.  $\bar{x}$  is the mean of all observed values,  $f_i$  and  $s_j$  are the amounts by which the  $i^{\text{th}}$  family and the  $j^{\text{th}}$  sire are above or below the mean respectively.  $fs_{ij}$  is the interaction between family and sire and  $e_{ijk}$  is the error associated with the  $y_{ijk}$ . This model assumes that  $E(f_i) = E(s_j) = E(fs_{ij}) = E(e_{ijk}) = 0$  and that  $E(f_i)^2 = \sigma^2_s$ ,  $E(s_j)^2 = \sigma^2_s$ ,  $E(fs_{ij})^2 = \sigma^2_{fs}$  and  $E(e_{ijk})^2 = \sigma^2_e$ .

The expected sums of squares are given in table 7. Estimates of the variance components for each of the characteristics studied are listed in table 8. Some of the components are negative. These could well be the consequence of sampling error as small negative estimates are to be expected frequently if the true value of the variance is nearly zero and if the sampling variance is large.

From table 8, it may be seen that the family component of variance accounted for only a small portion of the total variation. For the interval from first breeding to conception and for services per conception  $\sigma^2_f$  was negative indicating that its effect is very nearly zero. All the estimates of  $\sigma^2_s$  were real, and, except for wither height, explained considerably more of the variation than  $\sigma^2_f$ . In this case,  $\sigma^2_f$  was 1.13 and  $\sigma^2_s$  was 0.84. Since these are both small, this could be a consequence of sampling error. On four of the seven characteristics studied, the family  $\times$  sire component was negative. The error variance was in every instance more than twice as large as any of the other components.

TABLE 6  
*Analysis of variance of reproductive efficiency, services per conception and length of service period for 19 cow families in the Reymann Memorial Herd*

Source of variation	Per cent reproductive efficiency		Services/conception		Days from calving to first breeding		Days from first breeding to conception	
	Degrees of freedom	Mean square	Degrees of freedom	Mean square	Degrees of freedom	Mean square	Degrees of freedom	Mean square
Between families .....	18	163	18	2.6	18	621	18	3236
Within families .....	382	134	350	3.0	350	548	350	4187
Between subgroups .....	269	152	255	3.2	255	593	255	4532
Between sires .....	45	411**	44	7.7**	44	991**	44	9026**
Between families within sires .....	224	100	211	2.2	211	511	211	3595
Within subgroups .....	131	101	113	2.5	113	457	113	3257
No. of cows in study .....	401		369		369		369	

\*\* P < 0.01 Highly significant

TABLE 7  
Components of variance in the expected sum of squares

S.S	$\sigma^2_F$	$\sigma^2_S$	$\sigma^2_{FS}$	$\sigma^2_E$
Family	$\frac{p}{n..} \frac{\sum_{i=1}^{p^2} n_{i.}}{n..}$	$\frac{p}{i=1} \frac{\sum_{j=1}^{p^2} n_{.j}}{n..}$	$\frac{q}{i=1} \frac{\sum_{j=1}^{p^2} n_{ij}}{n..}$	$p-1$
Sire	$\frac{q}{j=1} \frac{\sum_{i=1}^{p^2} n_{.i}}{n..}$	$\frac{q}{j=1} \frac{\sum_{i=1}^{p^2} n_{.j}}{n..}$	$\frac{p}{i=1} \frac{\sum_{j=1}^{p^2} n_{ij}}{n..}$	$q-1$
Family x Sire	$-\frac{q}{j=1} \frac{\sum_{i=1}^{p^2} n_{ij}}{n..} + \frac{p}{i=1} \frac{\sum_{j=1}^{p^2} n_{.i}}{n..}$	$-\frac{p}{i=1} \frac{\sum_{j=1}^{p^2} n_{ij}}{n..} + \frac{q}{j=1} \frac{\sum_{i=1}^{p^2} n_{.j}}{n..}$	$\frac{q}{i=1} \frac{\sum_{j=1}^{p^2} n_{ij}}{n..} - \frac{p}{j=1} \frac{\sum_{i=1}^{p^2} n_{.i}}{n..} - \frac{q}{i=1} \frac{\sum_{j=1}^{p^2} n_{ij}}{n..} + \frac{p}{i=1} \frac{\sum_{j=1}^{p^2} n_{.i}}{n..}$	$pq-m-p-q+3$
Error				1

TABLE 8  
Estimates of the components of variance on the seven characteristics studied in the 19 cow families in the Reymann Memorial Herd

Component	Type	Wither ht.	Wt.	Per cent reproductive efficiency	First breeding to conception	Calving to first breeding	Services / conception
	Estimate	Estimate	Estimate	Estimate	Estimate	Estimate	Estimate
Family	0.94	1.13	397.92	1.68	-56.57	2.81	-0.009
Sire	5.30	0.84	1603.77	36.84	680.76	60.91	0.68
Family x Sire	1.73	-2.54	-796.86	-2.51	288.86	33.76	-0.16
Error	12.88	15.77	9002.71	100.79	3283.29	457.21	2.47

These components give some indication of the magnitude of the effects of sire, family, sire by family interaction and error in determining the total variation. From them, it is evident that sire differences are larger than family differences; however, this sire variation would include environmental variation as well as genetic differences. Also, the progeny of a sire are more closely related than the individuals of a family.

*Relationship.* The average coefficient of relationship ( $r$ ) was 15 per cent between members of the same family. For selection between families to be very effective,  $r$  must be large (9). This low relationship may partially explain the small amount of variance accounted for by families. In addition, any relationship between families reduces the utility of family averages as an aid in selection. Using sires on two or more families would bring this about aside from any relationship of foundation animals. However, after a few generations, the relationship of individuals solely through the foundation cow of their respective families would be almost negligible.

Artificial insemination may aid in the development of unrelated families in the same herd, whereas before in a one or two sire herd this was not possible. Selection between unrelated families in different herds presents the problem of evaluating differences in herd environment.

Generally, cow families have been considered as they chance to have been developed with little or no effort on the part of the breeders to increase the amount of genetic variance between families by building up the relationship within the family. Therefore, cow families are not sufficiently differentiated as such to demand a great deal of consideration in a selection program.

#### SUMMARY

Data on type, body size, breeding efficiency and length of service period of 19 cow families were analyzed by the analysis of variance. Estimates of the variance components were obtained by equating the sample values to the expected sums of squares.

The family averages for weight, breeding efficiency and length of service period varied considerably. These differences were not significant, however, with  $0.2\%$  accounting for only a small portion of the total variation. The significant differences between families for type and height at withers could be partially attributed to environment and to the sires that were used on these cow families. Except for wither height, the sire variance was somewhat larger than that attributed to the family influence. The error variance explained more than half the total variation in every characteristic studied.

The low relationship between members of the same cow family may account for the small amount of variance attributed to family differences. It appears that under present breeding practices the genetic variance between families is too small for family selection of type, body size and breeding efficiency to be very effective.

## ACKNOWLEDGMENTS

The authors wish to thank H. O. Henderson, Head, Dairy Department, West Virginia University for his assistance in the preparation of this manuscript and to R. W. Touchberry, Department of Dairy Science, University of Illinois, for suggesting the method used in solving for the components of variance.

## REFERENCES

- (1) BARTLETT, J. W., AND MARGOLIN, S. A. Comparison of Inbreeding and Outbreeding in Holstein-Friesian Cattle. N. J. Agr. Expt. Sta. Bull. 712. 1944.
- (2) BOWLING, G. A., PUTNAM, D. N., AND ROSS, R. H. Age as a Factor Influencing Breeding Efficiency in a Dairy Herd. J. Dairy Sci., 23: 1171-1176. 1940.
- (3) BOWLING, G. A., AND PUTNAM, D. N. Watch your Dairy Cattle Grow. W. Va. Agr. Expt. Sta. Cir. 79. 1943.
- (4) GAINES, W. L. Live Weight and Milk-energy Yield in Holstein Cows. J. Dairy Sci., 23: 259-265. 1940.
- (5) GAINES, W. L. Live Weight of Cow at Various Stages of Lactation in Relation to Milk-energy Yield. J. Dairy Sci., 24: 795-797. 1941.
- (6) GOWEN, J. W. Conformation and Milk Yield in the Light of the Personal Equation of the Cattle Judge. Maine Agr. Expt. Sta. Bull. 314. 1923.
- (7) HENDERSON, C. R. Estimation of General, Specific, and Maternal Combining Abilities in Crosses Among Inbred Lines of Swine. Unpublished Ph.D. Thesis. Iowa State College, Ames. 1948.
- (8) KAB, E. Untersuchugen uber die Verebung von Fruchtbarkeit und unfruchtbarkeit beim gelben Frankenvieh. Dissertation, Inst. Tierz. univ Muenchen. Abs. in Imp. Bur. Genetics Animal Breeding Abs. 6, 3, p. 200. Sept., 1938.
- (9) LUSH, J. L. *Animal Breeding Plans*. 3rd ed. Iowa State College Press, Ames. 1945.
- (10) MORGAN, R. F., AND DAVIS, H. P. The Effect of Pregnancy and Parturition on the Weight of Dairy Cows. Neb. Agr. Expt. Sta. Research Bull. 82. 1936.
- (11) PUTNAM, D. N., AND HENDERSON, H. O. The Effect of Pregnancy on the Body Weight of Dairy Cows. J. Dairy Sci., 29: 657-661. 1946.
- (12) SEATH, D. M., STAPLES, C. H., AND NEASHAM, E. W. A Study of Breeding Records in Dairy Herds. La. Expt. Sta. Bull. 370. 1943.
- (13) SNEDECOR, G. W. *Statistical Methods*. 4th ed. Iowa State College Press, Ames. 1946.
- (14) SPIELMAN, ARLESS, AND JONES, I. R. The Reproductive Efficiency of Dairy Cattle. J. Dairy Sci., 22: 329-334. 1939.
- (15) TABLER, K. A., TYLER, W. J., AND HYATT, G., JR. Differences in Milk and Butterfat Production and Test of Ayrshire Cow Families. J. Dairy Sci., 33: 449-456. 1950.
- (16) TRIMBERGER, G. W., AND DAVIS, H. P. Predictability of Breeding Efficiency in Dairy Cattle from their Previous Conception Rate and from their Heredity. J. Dairy Sci., 28: 659-669. 1945.
- (17) TYLER, W. J., AND HYATT, GEORGE, JR. The Heritability of Official Type Ratings and the Correlation between Type Ratings and Butterfat Production of Ayrshire Cows. J. Dairy Sci., 31: 63-70. 1948.
- (18) WILLIAMS, W. L. A Standard for Measuring the Reproductive and Dairying Efficiency of Dairy Cattle. Cornell Vet., 9: 204-213. 1919.
- (19) WILLIAMS, W. L., AND CARPENTER, C. M. Annual Report N. Y. State Vet. College at Cornell Univ., p. 90. 1917-1918.

PROPERTIES OF THE COLOSTRUM OF THE DAIRY COW.  
VI. CREAMING AND RATE OF FLOW<sup>1</sup>

E. G. MOODY,<sup>2</sup> G. H. WISE,<sup>3</sup> D. B. PARRISH AND F. W. ATKESON

*Kansas Agricultural Experiment Station, Manhattan*

Though colostrum is a variable product (1, 2, 7, 11), it normally has certain chemical, physical and immunological characteristics (12, 13) that distinguish it from milk secreted later in lactation. Inasmuch as general observations have revealed that creaming and viscosity of colostrum are different from those of normal milk, the possibility of a relationship of these properties to the concentrations of certain nutritive constituents in the mammary secretions was considered. Thus, the objectives of the investigations reported herein were (a) to study the variability of creaming and of rate of flow (reciprocal indicator of viscosity) in colostrum secretions produced under the range of conditions normally encountered in a dairy herd and (b) to determine the relationship of the two aforementioned properties of colostrum to other characteristics.

EXPERIMENTAL METHODS

*Source of samples.* Most of the data were collected concurrently with those from other colostrum studies, 1945-47 (10, 11). The samples available for observation represented mammary products secreted during the initial stages of 120 lactations (first to seventh) of Holsteins, Jerseys, Ayrshires and Guernseys.

During the terminal 4 to 8 wk. of gestation, the experimental animals received either a typical winter ration (grain concentrate mixture, Atlas sorgo silage and hay) or a summer ration, which was pasture either alone or in lieu of part of the foregoing roughages. Some of the cattle received supplemental vitamin A and/or vitamin E during the prepartal period, but these constituents apparently did not affect the specific physical characteristics under consideration. All animals were subjected to routine herd management.

*Collection and storage of samples.* In order to obtain representative samples of colostrum secretions, new-born calves were prevented from nursing and the mammary glands were evacuated (twice daily) as completely as possible by either hand or machine milking. The first milking usually was completed within 4 hr. postpartum; subsequent milkings were at the regular morning and evening periods. The mammary products were weighed, carefully mixed and sampled at each milking during the first 4 days and the seventh day. The portions used for measurements of cream volume, rate of flow and leucocyte count were stored at about 10° C. for 24 hr. to obtain a standard uniform temperature. When

Received for publication July 22, 1950.

<sup>1</sup> Contribution no. 193, Department of Dairy Husbandry and no. 407, Department of Chemistry.

<sup>2</sup> Present address: Department of Dairy Husbandry, Purdue University, Lafayette, Indiana.

<sup>3</sup> Present address: Department of Animal Industry, N. Carolina State College, Raleigh.



determinations of other properties (11) were made, a part of the original sample either was analyzed immediately or was stored at 4° C. for analyses later.

*Properties of colostrum determined.* (a) *Creaming.* The sample, stored at 10° C. for 24 hr., was mixed uniformly by pouring from one container into another four times, after which 200 ml. were placed into a graduated cylinder and maintained at 10° C. for an additional 48 hr. The depth of the upper layer (expressed as per cent) developed during this period was recorded as the *cream volume* or extent of creaming (fig. 1). When demarcation was not discernible, the cream volume was listed as 100 per cent.

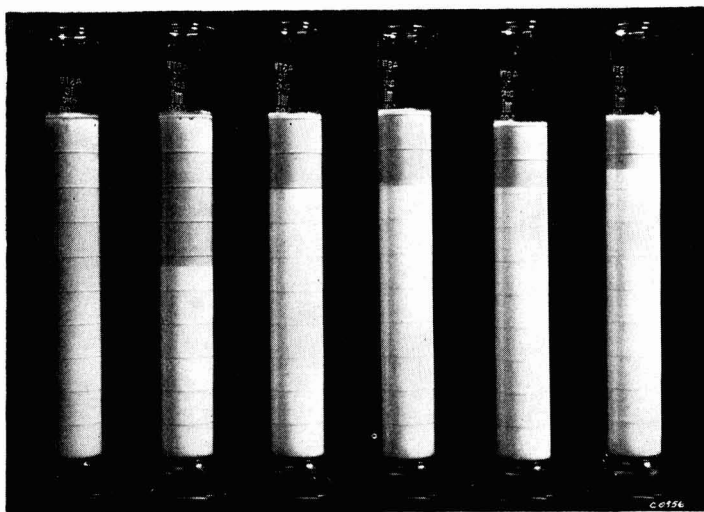


FIG. 1. Measurement of creaming properties of the first six consecutive postpartum milkings as illustrated by samples from one cow. (Sudan III added for photographic purposes).

(b) *Rate of flow.* This value, expressed in milliliters per minute, was computed from the time required for 188 ml. of a 200-ml. sample (12 ml. remained in receptacle) at 10° C. to flow through a 1.97-mm. orifice of a Borden Body-Flow meter (9). Thus determined, rate of flow is a reciprocal expression of the relative viscosity.

(c) *Other properties and constituents.* All samples were inspected to determine the presence of red to brown substances, apparently blood constituents.

The number of leucocytes in the samples of the first, third and eighth milkings from thirteen cows was estimated by Breed's microscopic method (6), using Pappenheim's stain (3) to facilitate identification of white corpuscles. No differentiation of types of leucocytes was made.

The standard Babcock procedure for testing whole milk was used to determine the fat content of the samples.

The methods used in the determination of proteins (10) and of specific gravity, solids-not-fat, lactose and ash (11) have been described.

## RESULTS

*Transitional changes in cream volume, rate of flow, occurrence of blood and number of leucocytes.* Changes in cream volume of mammary secretions during the transition from colostrum to normal milk are illustrated in fig. 2. The aver-

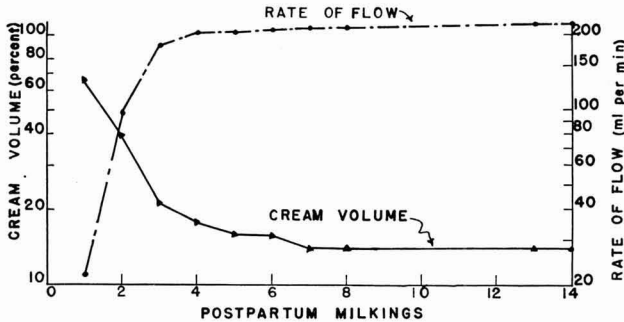


FIG. 2. Average cream volume and rate of flow of mammary secretions collected during the transition from colostrum to milk.

age cream volume decreased rapidly, from 66 per cent to 21 per cent, during the first three milkings; thereafter, the values gradually declined to a level of about 14 per cent. This trend was observed in samples from about four-fifths of the cows; in the remaining, however, the cream volume was greater in colostrum from a milking after the first. In every series of transitional samples the cream volume in at least one of the collections during the colostrum period exceeded subsequent values in normal milk.

The average rate of flow increased rapidly from the first to third milking, after which there was little change (fig. 2). In only two instances were exceptions to this trend observed. Apparently, the rate-of-flow values of the transitional mammary secretions had plateaued before the thirteenth or fourteenth milkings.

Frequently, blood constituents concentrated at the bottom of the samples, but occasionally there was a tendency for the pigmented material to aggregate immediately below the cream layer. In some samples, however, the particles remained diffused throughout.

This pigmented material was detected in varying amounts in at least one sample of colostrum from one-half of the cows; the frequency of occurrence and the apparent concentration usually did not decrease perceptibly until after the third milking. No relationship between the presence of blood constituents and the magnitude of either cream volume or rate of flow was evident.

An abundance of leucocytes has been listed as one of the principal biological characteristics of colostrum (5). The average numbers of leucocytes in colostrum secretions from 13 cows were 5,600,000, 1,600,000, and 350,000 per milliliter for the first, third and eighth milkings, respectively. Although these changes during the transitional stages followed the same general downward trends as cream

volume and viscosity, the few data did not reveal any definite interrelationships of these properties.

*Variability in cream volume and in rate of flow.* One of the marked characteristics of colostrum was its variability in cream volume and in rate of flow. The range in values for these respective properties was from 3 to 100 per cent and from 2.6 to 212 ml. per minute in first-milking samples produced under the variety of conditions usually prevailing in a dairy herd; even colostrum from comparable cows under similar experimental conditions exhibited diversity. Variability decreased during the transition to normal milk; after the seventh day, the ranges were only from 9 to 24 per cent for cream volume and 176 to 221 ml. per minute for rate of flow. Scatter diagrams employed in showing relationships of these two properties to one another and to other properties reveal further the wide variations in colostrum secretions.

*Interrelationship of cream volume and rate of flow.* In all the interrelationship studies that follow, observations on only the first postpartum samples are included.

As shown in fig. 3, there was a tendency toward an inverse relationship be-

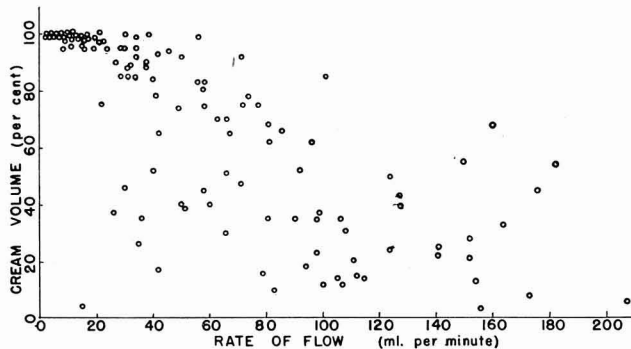


FIG. 3. Relation of cream volume to rate of flow in first postpartum milkings.

tween the rate of flow and the cream volume. The correlation coefficient ( $r$ ) was statistically significant. In the most viscous colostrum, in which the rate of flow was from 3 to 15 ml. per minute, the cream volume was least variable, ranging within the limits of 95 to 100 per cent, but when rate of flow ranged from 15 to 75 ml. per minute, the cream volume was most variable, ranging from 4 to 100 per cent. Furthermore, when the rate of flow ranged from 75 to 210 ml. per minute, the cream volume exceeded 70 per cent in only two instances.

*Relationship of creaming and of rate of flow to other properties of first postpartum colostrum.* A. *Specific gravity.* As specific gravity increased, cream volume tended to increase and rate of flow to decrease (fig. 4). The relationship of specific gravity to rate of flow was more pronounced than it was to creaming, inasmuch as separation into two distinguishable layers did not occur in about one-fourth of the samples having a specific gravity greater than 1.060.

B. *Milk fat.* Changes in fat concentrations in colostrum were reflected in

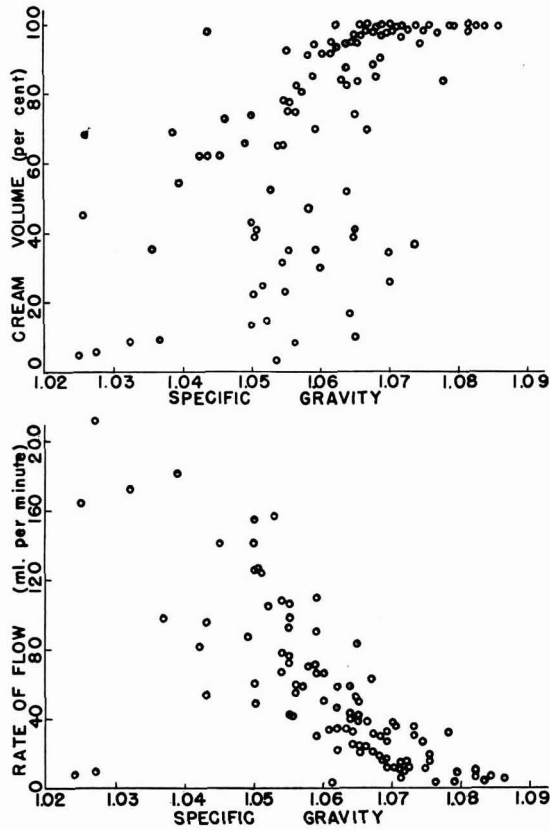


FIG. 4. Relation of cream volume and of rate of flow to specific gravity in first postpartum milkings.

cream volume and in rate of flow only in a general way (fig. 5). As the fat content increased from approximately 1 to 7 per cent, the cream volume, though varying over a wide range, also tended to expand. The relationship of fat content to rate of flow showed no consistent trend.

C. *Solids-not-fat*. The relationships of cream volume and rate of flow of colostrum to solids-not-fat are shown in fig. 6. When solids-not-fat of colostrum exceeded 22 per cent, the cream volume was 97 to 100 per cent, but at lower levels of solids-not-fat, the cream volume was highly variable. There was a tendency, however, for the solids-not-fat to vary directly with cream volume and inversely with rate of flow. (a) *Proteins*. Quantitatively, the most important constituent of the colostrum solids-not-fat is protein of which albumin and globulin generally constitute the major portion (10, 11). Relationships of cream volume and rate of flow to total protein and albumin-globulin (fig. 7) were similar to those observed for solids-not-fat. The protein concentration above which colostrum usually failed to separate into two layers, resulting in nearly

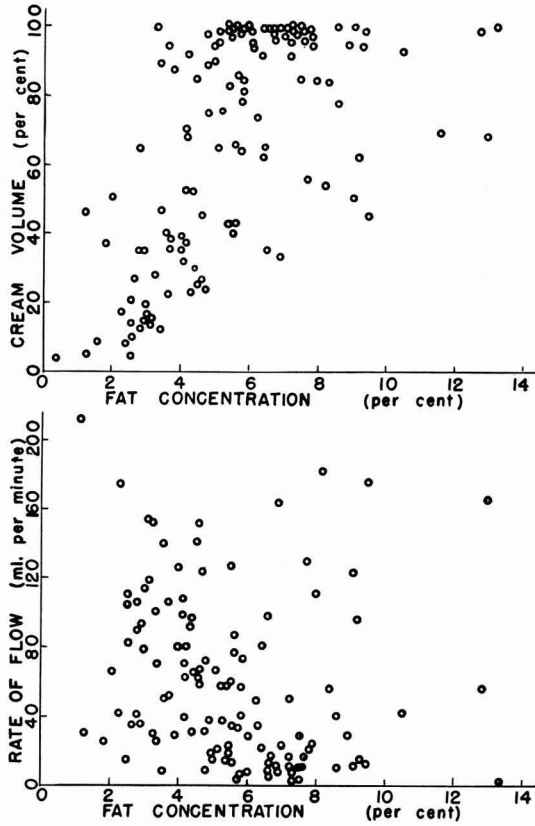


FIG. 5. Relation of cream volume and of rate of flow to fat concentration in first post-partum milkings.

100 per cent cream volume, seemed to be 16 per cent total protein and 8 per cent albumin-globulin. The casein content bore no apparent relationship to either cream volume or rate of flow (8). (b) *Lactose*. The data, few and variable, showed no specific relationship of lactose to either cream volume or rate of flow (8). (c) *Ash*. The ash content showed no definite relationship to either creaming or rate of flow (8); consequently, neither of these measurements would be indicative of total mineral content.

DISCUSSION

Colostrum having a high total solids content presumably is superior nutritionally to that having fewer solids (2, 11). In spite of wide variations in the values for cream volume and rate of flow, the foregoing results tended to indicate a relationship of these properties to concentrations of: solids-not-fat, total protein, and albumin and globulin. Of these constituents, high concentrations of protein, particularly the albumin-globulin fraction, seemed to be associated with

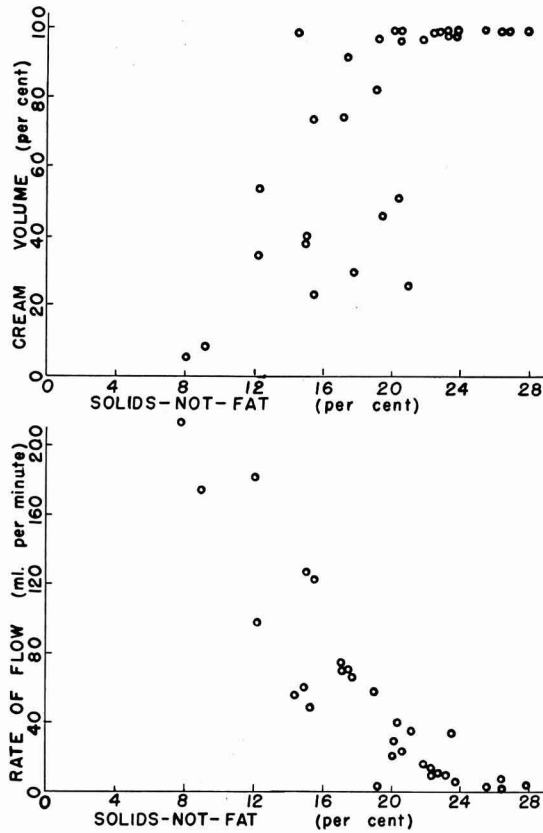


Fig. 6. Relation of cream volume and rate of flow to solids-not-fat in first postpartum milkings.

large cream volumes and small rates of flow. Hansson (4) reported that the addition of globulins to milk improved the creaming, and Jacobson and Wallis (7) ascribed the thick, syrupy character of colostrum to its high protein content.

It should be emphasized that the number of samples involved in most of the studies reported herein was inadequate to establish the significance of the observations. It is possible that physical measurements made under conditions other than those employed in this study might have shown different relationships.

The wide variations noted in cream volume and in rate of flow prompted a study of the possible involvement of production conditions; whereupon the following were surveyed as possible contributing factors: breed, extent of parturient edema, mastitic condition, length of gestation, length of dry period (1), number of lactation, type of ration, season of the year, and yield of colostrum. Only the last four of these factors gave any indication of relationships to creaming and viscosity (8); colostrum from cows in the first lactation tended to have a greater cream volume and a lower rate of flow than colostrum from cows in

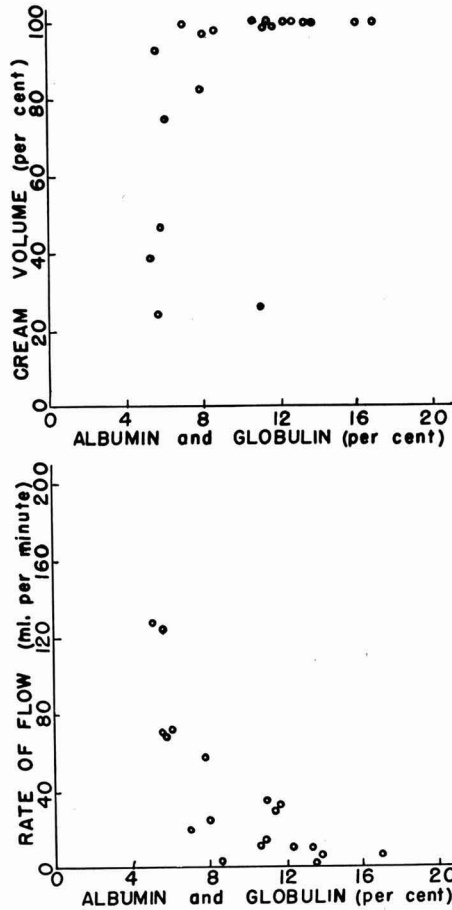


FIG. 7. Relation of cream volume and of rate of flow to the combined albumin-globulin fraction in first postpartum milkings.

later lactations; colostrum from cows receiving only dry feed with no pasture in the terminal stages of lactation, colostrum produced in cooler seasons and colostrum produced in smaller amounts tended to show an increased cream volume and decreased rate of flow (increased viscosity). Further experimentation under conditions designed to study the foregoing factors in detail is necessary to establish their etiological role.

Even though observation of dry periods ranging from 29 to 224 days revealed no effect on cream volume and on rate of flow, periods of less than 29 days might have yielded different results. The mammary secretions from one cow (126A), which was milked throughout gestation, resembled normal milk in general appearance during the pre- and post-partal stages, but several days before and after calving there was marked lipolysis, absence of a cream line and a

slight decrease in rate of flow. These observations would indicate that extreme abbreviation of the non-lactating period might affect the physical properties of the post-partal mammary secretion.

Another possible contributor to the variability of colostrum properties is the udder leakage that occasionally occurred prior to the first milking. Since this factor was not measurable, its effect on cream volume and rate of flow of the collected colostrum remains unknown.

It seems that the peculiarities of the individual cow are the major causes for colostrum variability. Although minor alterations in certain physical characteristics and related chemical components of colostrum may be possible through extremes in feeding and managemental practices, measures to obtain a specific type of colostrum seem to be beyond the practicable realm at present.

#### SUMMARY

A study conducted over a period of 3 yr. involved colostrum from as many as 120 lactations. Observations on these samples showed that:

(a) During the transition period, cream volume decreased and rate of flow increased, the changes being most rapid during the first three milkings.

(b) In samples of colostrum from the first postpartum milking, the cream volume and the rate of flow (inverse expression of viscosity) were extremely variable, even when the mammary secretions were produced under apparently similar conditions. However, in any given sample there was a tendency for an inverse relationship between cream volume and rate of flow.

(c) There appeared to be the following relationships of cream volume and rate of flow, respectively, to other properties and to various components of the first postpartum mammary secretions: when high values for specific gravity, solids-not-fat, total protein and albumin-globulin fraction were noted, the percentage of cream volume tended to be large and the rate of flow small. Though there was a general tendency for large cream volumes to be associated with high fat percentages, variability was pronounced; rate of flow apparently was independent of fat content. Neither differences in concentrations of casein, lactose and ash, nor presence of visible blood, nor variations of leucocyte numbers was related to cream volume and to rate of flow.

(d) The data suggest that the variability in cream volume and in rate of flow are attributable primarily to the individual characteristics of the cow; environmental factors normally encountered in a dairy herd apparently played only a secondary role.

#### REFERENCES

- (1) ECKLES, C. H., AND PALMER, L. S. The Influence of Parturition on the Composition and Properties of the Milk and Fat of the Cow. *J. Biol. Chem.*, 27: 313-326. 1916.
- (2) ENGEL, H., AND SCHLAG, H. Beitrage zur Kenntnis des Colostrums der Kuh. *Milchw. Forsch.*, 2: 1-15. 1924.
- (3) GRADWOHL, R. B. H. *Clinical Laboratory Methods and Diagnoses*, 2nd ed. C. V. Mosby, St. Louis, Mo. p. 784. 1938.
- (4) HANSSON, E. The Creaming of Milk. *Proc. 12 Intern. Dairy Congr.*, 2: 30-35. 1949.



- (5) HOUDINIÈRE, A. Le colostrum de vache. Etude physico-chimique du colostrum. *Lait*, **24**: 226-253. 1944.
- (6) INTERNATIONAL ASSOCIATION OF MILK DEALERS. *Methods of Analyses of Milk and its Products*. Chicago, Ill. pp. 84-89. 1933.
- (7) JACOBSON, D. H., AND WALLIS, G. C. Factors Affecting the Composition of Milk. S. Dak. Agr. Expt. Sta. Bull. 331. 1939.
- (8) MOODY, E. G. Relation of Certain Factors to Cream Volume and Viscosity of Colostrum from Dairy Cows. Unpublished Masters Thesis. Kansas State College. 1947.
- (9) NAIR, J. H., AND MOOK, D. E. Viscosity Studies of Fluid Cream. *J. Dairy Sci.*, **16**: 1-9. 1933.
- (10) PARRISH, D. B., WISE, G. H., HUGHES, J. S., AND ATKESON, F. W. Properties of the Colostrum of the Dairy Cow. II. Effect of Prepartal Rations upon the Nitrogenous Constituents. *J. Dairy Sci.*, **31**: 889-895. 1948.
- (11) PARRISH, D. B., WISE, G. H., HUGHES, J. S., AND ATKESON, F. W. Properties of the Colostrum of the Dairy Cow. V. Yield, Specific Gravity and Concentrations of Total Solids and its Various Components of Colostrum and Early Milk. *J. Dairy Sci.*, **33**: 457-465. 1950.
- (12) ROGERS, L. A., ASSOCIATES OF *Fundamentals of Dairy Science*, 2nd. ed. Reinhold Publishing Corp., New York. pp. 466-474. 1935.
- (13) SMITH, E. L. The Isolation and Properties of the Immune Proteins of Bovine Milk and Colostrum and their Role in Immunity: A Review. *J. Dairy Sci.*, **31**: 127-138. 1948.

# EFFECTS OF ADDED HYALURONIDASE AND HYALURONIC ACID ON THE MOTILITY OF BULL SPERMATOZOA<sup>1</sup>

JAMES E. JOHNSTON<sup>2</sup> AND JOHN P. MIXNER

*New Jersey Agricultural Experiment Station, Sussex*

The enzyme hyaluronidase is a constituent of mammalian semen and a role has been proposed for it in the process of mammalian fertilization (1). It is of biological interest, therefore, to determine the effect of added hyaluronidase and its substrate, hyaluronic acid, on the motility characteristics of mammalian semen.

## MATERIALS AND METHODS

Six semen samples were obtained from four highly fertile Holstein-Friesian dairy bulls. These semen samples were diluted 1:20 with a diluter composed of one part by volume of fresh egg yolk and three parts by volume of a 3.0 per cent solution of sodium citrate dihydrate. Diluted semen was used in these experiments because studies in this laboratory have shown that a negative correlation exists between the hours duration of spermatozoan motility of stored undiluted semen and spermatozoan concentration and that use of diluted semen eliminates this relationship. The hyaluronidase used in these studies was a dry, partly purified preparation of bull testes hyaluronidase. Turbidimetric assay of this material indicated that 0.7 mg. was equivalent in potency to 1.0 mg. of a similar preparation which had been assayed by Schering Corporation<sup>3</sup> at 30 turbidity-reducing units (TRU) per milligram (July, 1947). On this basis, the hyaluronidase used in this study would have a potency of 43 TRU per milligram Schering equivalent. Dosages of hyaluronidase used were 0, 5, 10, 20 and 40 mg. per milliliter of diluted semen. Hyaluronic acid (Schering 55-129 VI) was added at the rate of 0, 2, 4, 8, 16 and 32 mg. per milliliter of diluted semen. To estimate the relative concentration of hyaluronidase contained in the undiluted semen as compared to the amount added, the semen samples were assayed for hyaluronidase initially within 1 hr. of ejaculation and again following 24-hr. incubation at 37° C. under toluene as described by Johnston and Mixner (2). The 24-hr. assay gives an estimation of the initial seminal plasma hyaluronidase plus the hyaluronidase released by the spermatozoa as they die.

The estimated percentage of spermatozoa exhibiting motility after 5 and 10 days storage at 5° C. was used as the criterion of the effects on motility of added hyaluronidase and hyaluronic acid. Five-hundredths ml. of semen was added to 1.0 ml. of diluter to which hyaluronidase or hyaluronic acid previously had

Received for publication July 24, 1950.

<sup>1</sup> Paper of the Journal series, New Jersey Agricultural Experiment Station, Rutgers University—the State University of New Jersey, Department of Dairy Industry.

<sup>2</sup> Present address: Dairy Department, Louisiana State University, Baton Rouge.

<sup>3</sup> Hyaluronidase and hyaluronic acid preparations used in this study were generously supplied by Schering Corp., Bloomfield, N. J.

been added. The semen and diluter were mixed well and then cooled to 5° C. Motility estimates were made by examining a thin film of the diluted semen under a cover slip with a microscope (440X) using a stage incubator set at 37° C.

## RESULTS

The estimated mean percentage motilities at 5 and 10 days for the six semen samples and for the various dosage levels of hyaluronidase are presented in table 1. An analysis of variance of the ungrouped data indicated that there

TABLE 1

*Effect of several dosage levels of hyaluronidase on mean percentages of motile spermatozoa on storage for 5 and 10 days at 5° C.*

Hyaluronidase dosage (mg./ml.)	Hyaluronidase potencies equivalent to Schering TRU	Mean % motility	
		5 d.	10 d.
0	0	47.5	20.0
5	215	45.8	20.0
10	430	45.0	21.7
20	860	46.7	20.8
40	1720	45.0	18.3

were no significant differences among the mean percentages of motile spermatozoa of the various dosage levels of hyaluronidase.

The mean initial and 24-hr. hyaluronidase potencies for all undiluted semen samples (Schering TRU equivalents) were 1,494 and 2,715 TRU per milliliter, respectively. These quantities compare favorably to the quantities of added hyaluronidase. The mean spermatozoa concentration of the six semen samples was 1,688,000,000 per milliliter.

TABLE 2

*Effect of several dosage levels of hyaluronic acid on mean percentages of motile spermatozoa on storage for 5 and 10 days at 5° C.*

Dosage of hyaluronic acid (mg./ml.)	Mean % motility	
	5 d.	10 d.
0	47.5	20.0
2	46.7	20.0
4	45.8	20.8
8	43.3	18.3
16	26.7	7.0
32	.....	.....

Table 2 presents similar motility data for the hyaluronic acid. Analysis of variance of the ungrouped data indicated that there were highly significant differences ( $P < 0.01$ ) among the mean percentages of motile spermatozoa of the various dosage levels of hyaluronic acid. The data indicate that toxicity is shown at a dosage level between 8 and 16 mg. per milliliter.

## DISCUSSION

The results of this experiment indicate that hyaluronidase is relatively non-toxic to bull semen. The dosage levels used were chosen so that the maximum concentration of hyaluronidase (in terms of TRU) would be greater in the diluted semen than the concentration which might be expected in a fresh sample of undiluted bull semen. Though Johnston and Mixner (3) have found no significant correlation between hyaluronidase titer and fertility of semen, Sallman and Birkeland (4) have reported a negative correlation between these two factors and have even suggested that the removal of hyaluronidase from bull semen might increase the fertility of the semen. In any case, results reported here indicate that hyaluronidase could not be reducing fertility through interference with the motility characteristics of spermatozoa. In this connection it may be noted that Johnston *et al.* (5) have shown a negative correlation to exist between hyaluronidase titer and the percentage of live spermatozoa in bull semen, whereas Stone *et al.* (6) have shown a positive correlation between percentage of live spermatozoa and fertility of bull semen.

Although hyaluronic acid in excess of 8 mg. per milliliter exerted a detrimental effect on spermatozoa motility, the cause of this was not ascertained. Gross pH of the medium seemingly was not a factor, since it was decreased very little by the added hyaluronic acid. However, the high viscosity exhibited in the samples to which 16 and 32 mg. hyaluronic acid per ml. had been added may have exerted an indirect effect on motility characteristics (*viz.* prevention of dispersion of metabolic products of spermatozoa).

## SUMMARY

Amounts of hyaluronidase up to 40 mg. per milliliter of diluted bull semen did not affect significantly the motility characteristics of the spermatozoa after storage at 5° C. for 5- and 10-day periods. Under similar storage conditions, amounts of hyaluronic acid greater than 8 mg. per milliliter of diluted bull semen caused a significant reduction in the percentage of motile spermatozoa.

## REFERENCES

- (1) MEYER, K. The Biological Significance of Hyaluronic Acid and Hyaluronidase. *Physiol. Rev.*, **27**: 335-359. 1947.
- (2) JOHNSTON, J. E., AND MIXNER, J. P. Development of Hyaluronidase in Bull Semen. *J. Animal Sci.*, **7**: 440-446. 1948.
- (3) JOHNSTON, J. E., AND MIXNER, J. P. Relationship of Hyaluronidase Concentration to Fertility of Dairy Bull Semen. *J. Dairy Sci.*, **33**: 847-850. 1950.
- (4) SALLMAN, B., AND BIRKELAND, J. M. Interrelationships of Spermatozoa Count, Hyaluronidase Titer and Fertilization. *J. Physiol.*, **152**: 271-279. 1948.
- (5) JOHNSTON, J. E., STONE, E. J., AND MIXNER, J. P. Hyaluronidase Relationships in Dairy Bull Semen. *J. Dairy Sci.*, **32**: 574-579. 1949.
- (6) STONE, E. J., JOHNSTON, J. E., AND MIXNER, J. P. Live Spermatozoa Relationships and Fertility of Dairy Bull Semen. *J. Dairy Sci.*, **33**: 442-448. 1950.

## IMPROVED METHODS IN THE RECOVERY OF LACTOSE USING ION EXCHANGE RESINS<sup>1</sup>

E. D. McGLASSON AND J. C. BOYD<sup>2</sup>

*Department of Dairy Husbandry, College of Agriculture, University of Idaho, Moscow*

Some lactose is being manufactured from skimmilk but whey is the source of nearly all commercial lactose. Present-day methods of lactose recovery, which are essentially the same as a decade ago, require expensive equipment and yield only about 50 per cent of the original lactose in whey. As a result production costs are high and utilization of lactose is restricted to those industries which can profitably use a high cost ingredient.

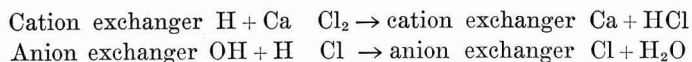
Any method of increasing present-day yields or reducing production costs should materially benefit the dairy industry, as an increased utilization of whey will provide more products to sell and aid in eliminating an increasingly serious whey disposal problem.

Through the use of ion exchange resins, Almy and Garrett (1) have been able to produce a high grade crude lactose in which substantially all of the lactose present in the original whey was recovered, thus reducing the cost of lactose manufacture through greater recovery, use of less power, labor and equipment than that required in present-day lactose manufacture.

Associates of Rogers (3) state that refined lactose of commerce should be a white, odorless, crystalline powder, pulverized sufficiently to pass through a no. 100 screen. In addition, it should be 99.7 per cent pure as determined by the polariscope, comply with USP heavy metal test and make a solution which is clear, colorless and neutral to litmus. For use in modified milks and as a substrate for fermentation processes, a comparatively large percentage of other components of whey is not objectionable and may be desirable in some instances according to Whittier (9).

Almy and Garrett (1), Meade and Clary (7) and Miller and Arrigoni (8) have used ion exchange materials for the removal of certain constituents from whey. The use of ion exchange resins in the purification of beet sugar is described, however, by Haagensen (5).

The general reaction to these materials is as follows:



Through such reactions any positively or negatively charged ion theoretically can be removed. As the exchange materials become exhausted, they are regenerated with either an acid or alkaline solution.

Received for publication July 29, 1950.

<sup>1</sup> Published with the approval of the director of the Idaho Agricultural Experiment Station as Research paper no. 318.

<sup>2</sup> Associate Dairy Husbandman, Idaho Agricultural Experiment Station, now on military leave.

## EXPERIMENTAL PROCEDURE

Through preliminary experiments with different ion exchange resins, Amberlite exchange materials proved to be the more satisfactory. Amberlite IR-120, a high density, nuclear sulfonic-acid type ion exchanger, was used for the cation cycle. Amberlite IR-120 is an extremely stable product with a very high exchange capacity. Amberlite IRA-400, a strongly basic, synthetic resin absorbent, was used in the anion cycle of operation. Amberlite IRA-400 behaves as solid caustic with only its hydroxyl ions in solution and will absorb negatively charged ions from acid, neutral and even mildly alkaline solutions. The high basicity of this material is comparable to the acidity in the hydrogen cycle of the sulfonic-type cation exchangers.

Two methods were used in the recovery of lactose from cheddar cheese whey. One method was based on the assumption that the principal material to be recovered was lactose, while the other method assumed that the recovery of a whey protein which is fairly soluble also was desirable. The effect of ion exchange resins on the recovery of the protein fraction of cheese whey is reported in another paper.

Where the recovery of a soluble whey protein was a consideration, the cheese whey was passed through the ion exchange resins prior to the removal of the protein material. As the resulting whey was neutral to slightly basic, HCl was used to standardize the acidity to 0.10 to 0.13 per cent. Where the lactose only was considered, the whey was not passed through ion exchange resins and the acidity was standardized to 0.10 to 0.13 per cent with NaOH.

After standardizing the acidity of the whey, the procedure was the same for both methods. The standardized whey was heated to near the boiling point and  $\text{CaCl}_2$  added to precipitate the whey protein. The  $\text{CaCl}_2$  solution was prepared by dissolving 1.3 lb. of  $\text{CaCl}_2$  in water and making the solution up to 1000 ml. One ml. of this solution was added per pound of hot whey. In preliminary experiments, using  $\text{CaCl}_2$ ,  $\text{H}_2\text{SO}_4$ , HCl and NaOH, it was found that  $\text{CaCl}_2$  gave a more complete protein precipitation than did the acids and NaOH. Use of  $\text{CaCl}_2$  not only gave a more complete protein precipitation but also eliminated the deteriorating fumes that attack equipment and buildings when acids are added to the near boiling whey.  $\text{CaCl}_2$  also is more easily handled by employees under normal cheese factory operations.

After the whey protein was precipitated, the remaining liquid containing the lactose was siphoned off, cooled to 70° F. and clarified. The lactose solution then was concentrated in a single-effect vacuum pan to a Baume reading of 10°, cooled to 40° F. and filtered. After filtering, the temperature was raised to 68 to 70° F. and the liquid passed through cation (Amberlite IR-120) and anion (Amberlite IRA-400) resinous exchange materials. It then was heated to 100° F. and spray dried, using an air inflow temperature of 210° F., a chamber temperature of 140° F. and an exhaust air temperature of 120° F.

Samples were taken at various points in the processing and analyzed for nitrogen and ash content by the methods outlined by the Association of Official Agricultural Chemists (2). The purity of several samples of the finished lac-

tose was determined by melting point determinations as outlined by the Association of Official Agricultural Chemists (2) and saccharimeter readings converted to polariscope readings, as outlined by Brown and Zerban (4). Reagent grade lactose was used as the basis for the saccharimeter readings.

#### RESULTS AND DISCUSSION

Table 1 shows the effect of the various processes on the ash, nitrogen and lactose content of the total solids in the lactose solution. The results were arrived at by taking arithmetical averages of 12 determinations. Whereas the average lactose content of the total solids in the dried lactose in method I was 95.6 per cent and of method II, 97.6 per cent, several lots of lactose produced by the second method were better than 99 per cent pure lactose.

The comparative purity of five of the best runs as determined by saccharimeter readings converted to polariscope readings and by melting point determinations, using reagent grade lactose as 100 per cent pure, is as follows: for method I, 93.7 and 94.1 per cent purity, and for method II, 98.3 and 98.7 per cent purity for polariscope and melting point determinations, respectively. It should be pointed out here that in some cases lactose of better than 99 per cent purity was obtained but in no instance was lactose of USP grade obtained.

It has not been determined why the passage of the original whey through the exchange resins results in a less pure lactose (method I) than when the original whey is not so treated (method II).

It is possible, however, that passing the original whey through the exchange resins removes a considerable amount of divalent ions such as calcium and that the addition of  $\text{CaCl}_2$  as a precipitating agent does not replace these ions sufficiently to allow complete coagulation of the protein material. Whether or not the addition of a greater amount of  $\text{CaCl}_2$  would overcome this difficulty has not been determined.

As the lactose solution passes through the cation resin (Amberlite IR-120) and anion resins (Amberlite IRA-400) there occurs a wide pH range. Before the lactose solution enters the first cation cell it is at an approximate pH of 5.8, and as the lactose solution leaves the cation cell the pH has been reduced to approximately 3.5. When the lactose solution is recovered from the anion cell, the lactose solution has a pH of approximately 10.0, thus completing the first cycle of the three battery operations. As the lactose solution was recovered from the cation cell of the second battery of operation, the pH was reduced to approximately 5.4, and from the anion cell the pH was approximately 8.4. When the lactose solution was subjected to the third battery of cation and anion cells, the pH of the recovered lactose solution was approximately 7.0, thus completing the operation using ion exchange materials and leaving the lactose solution ready for spray drying.

Since there is a wide range in the pH of the lactose solution and since there is some inversion of sucrose when cation and anion exchange materials are used (5), four samples of the powdered lactose were analyzed by the Barfoed's copper acetate test (6) for monosaccharides. In every case negative results

TABLE 1

*Effects of various processes on the ash, nitrogen and lactose content of the total solids in the lactose solution<sup>a</sup>*

Steps in processing	Method I			Method II		
	Original whey passed thru cation and anion exchange resins			Original whey not passed thru cation and anion exchange resins		
	Ash in T. S.	Nitrogen in T. S.	Lactose in T. S. (appt. purity)	Ash in T. S.	Nitrogen in T. S.	Lactose in T. S. (appt. purity)
	(%)	(%)	(%)	(%)	(%)	(%)
Separated whey	5.96	2.18	80.13	5.43	2.04	81.55
Passed thru ion exchange	4.82	2.15	81.36	Omitted		
Removal of protein	4.43	1.02	89.06	4.97	0.99	88.71
Clarified solution	4.43	1.02	89.06	4.92	0.94	89.07
Condensed to 10° Baume'	4.43	0.99	89.25	4.93	0.89	89.38
Filtered	4.40	0.93	89.67	4.87	0.88	89.51
1st passage thru exchange resins	2.67	0.76	92.21	1.90	0.86	92.62
2nd passage thru exchange resins	0.66	0.71	95.81	0.48	0.63	95.50
3rd passage thru exchange resins	0.41	0.70	95.12	0.29	0.46	96.88
Spray-dried lactose	0.39	0.68	95.66	0.18	0.34	97.68

<sup>a</sup> Each step in process arrived at by taking arithmetical average of 12 determinations.

were obtained. These results would indicate that there is little likelihood of hydrolysis occurring in the purification of lactose using the average exchange resin.

As the partially deproteinized whey enters the first cycle of the cation and anion exchange resins, it is a brilliant fluorescent green. As the lactose solution comes from the third cycle of ion exchange columns, it is colorless, or, in some cases, very slightly turbid. The dry lactose is a fine white powder, slightly sweet and readily soluble in water. In solution it is colorless, odorless and neutral to litmus.

#### SUMMARY

Two methods of recovering lactose from cheddar cheese whey were studied. One method involved passing the original whey through ion exchange resins, removing the whey protein and further purifying the removed lactose solution with ion exchange resins. The purified lactose solution then was spray dried. The second method differed from the first only in that the original whey was not treated with ion exchange resins prior to the recovery of the protein fraction.

Lactose of a higher degree of purity was recovered when the original whey was not treated with ion exchange resins. An average purity of 97.0 per cent on 12 runs was obtained. Five selected runs yielded lactose of an average purity of better than 98 per cent. Three runs yielded lactose of better than 99 per cent purity although none quite met specifications of USP grade.



The recovered lactose was a fine, white powder, slightly sweet and readily soluble. In solution the recovered lactose was colorless, odorless, and neutral to litmus.

Analysis of the recovered lactose for monosaccharides gave negative results.

## REFERENCES

- (1) ALMY, E. F., AND GARRETT, O. F. Preparation of High-grade Crude Lactose. United States Patent no. 2,477,558. Aug. 2, 1949.
- (2) ASSOCIATION OF OFFICIAL AGRICULTURAL CHEMISTS. *Official and Tentative Methods of Analysis*. pp. 292-293. College Offset Press, Philadelphia, Pa.
- (3) ASSOCIATES OF ROGERS. *Fundamentals of Dairy Science*. pp. 130. Reinhold Publishing Corp.
- (4) BROWNE, C. A., AND ZERBAN, F. W. *Physical and Chemical Methods of Sugar Analysis*. pp. 180-181. John Wiley and Sons. New York.
- (5) HAAGENSEN, E. A. Ion Exchange Applied to Sugar Juice Purification. Presented Before American Society of Sugar Beet Technologists, Denver, Colo. Feb., 1946.
- (6) KLINER, I. S., AND DOTTIE, L. B. *Laboratory Instructions in Biochemistry*. The C. V. Mosby Publishing Co., St. Louis, Missouri.
- (7) MEADE, R. E., AND CLARY, P. D., JR. Method of Making Novel Products from Whey. United States Patent no. 2,465,906. March 29, 1949.
- (8) MILLER, O. H., AND ARRIGONI, L. Removal of Nitrogen from Deproteinized Whey by Ion Exchange Treatment. Presented before American Chemical Society, 116th Meeting. Atlantic City, N. J. Sept. 1949.
- (9) WHITTIER, E. O. Lactose and its Utilization: A Review. *J. Dairy Sci.*, **27** (12): 505-528. 1944.

A COMPARISON OF U.S. NO. 2 FIELD-CURED FIELD-BALED ALFALFA  
HAY WITH ARTIFICIALLY DRIED AND GROUND AND PELLETED  
ALFALFA HAYS AS A SOURCE OF CAROTENE AND  
ROUGHAGE FOR HOLSTEIN CALVES<sup>1</sup>

H. D. EATON, C. A. CARPENTER, R. J. CAVERNO, R. E. JOHNSON AND F. I. ELLIOTT  
*Animal Industries Department, Storrs Agricultural Experiment Station  
University of Connecticut, Storrs*

AND

L. A. MOORE

*Bureau of Dairy Industry, Agricultural Research Administration, U. S. D. A., Washington, D. C.*

The increased use of dry calf starters and other milk replacements in the rations of dairy calves in fluid milk areas has changed the early nutrition of the calf from that of milk to hay and grain primarily. Until the calf consumes appreciable amounts of hay under these limited milk feeding systems, there appears to be a period between birth and 4 mo. of age when there is not sufficient carotene intake to maintain adequate levels of blood plasma vitamin A (6). More recently it has been demonstrated (4, 17, 18) that an early intake of hay by young calves is desirable because it is accompanied by a development of rumen microorganisms as well as increases in blood plasma ascorbic acid and carotene.

Although the use of excellent quality roughage in limited whole milk calf feeding systems has been considered axiomatic (16, 23), the actual production of such hay by field-curing in the northeastern states, and particularly in Connecticut, is not realized by many dairymen because of unfavorable weather conditions (20). Barn-curing of hay in the northeastern states has not resulted in material decreases in carotene losses, especially those occurring during storage. In general, the carotene content of barn-cured hay when fed is only slightly higher than that of field-cured hay (2, 24).

The value of artificial drying in the preservation of nutrients in hay has been recognized for some time (5, 13). Pelleting of artificially-dried hay appears to afford a possible method of conserving carotene and decreasing shipping costs. Therefore, a study was undertaken to determine the relative values of field-cured, field-baled alfalfa hay with artificially-dried either ground or pelleted alfalfa hays as a source of carotene and roughage for dairy calves. The criteria used were the hay consumed, the weight changes, the blood plasma carotene and vitamin A levels, the time to deplete the calves of their vitamin A stores after 105 days of age and daily health observations.

Received for publication August 15, 1950.

<sup>1</sup> This study was made in part with funds provided by the Research and Marketing Act of 1946, through a contract between the Storrs Agricultural Experiment Station and the Bureau of Dairy Industry.

## EXPERIMENTAL

*Animals.* Fifteen Holstein calves, twelve males and three females, were obtained during October, November and December, 1949, from the Mansfield State Training School and Hospital, Mansfield, Conn. Shortly after birth, they were separated from their dams and transported to the University research barn. There they were placed in individual tie stalls in a separate portion of the barn in which radiant heat was applied from a ceiling panel when the temperature went below 10° C. A randomized block design was used with the three different types of hay and the 15 calves. The first three calves obtained were assigned at random to the three hay groupings, and successive trios of calves were treated in the same manner. The only exception was that the three female calves were restricted to one replication of the hay groups.

The three hays were taken from a field of alfalfa estimated to be in one-quarter bloom when cut in Dawson County, Nebr., in the Platte River Valley, which produces large quantities of dehydrated alfalfa meal. The field of alfalfa was laid out into plots by three members of the Bureau of Dairy Industry staff. The alfalfa for artificial drying and processing into meal and pellets was cut with a field harvester and immediately hauled by trailer truck to dehydrators. The alfalfa for field-curing and field-baling was cut the same day, raked into windrows and, in some instances, turned before being baled over a 5-day period. Some of the hay was rained on during this period. The average chemical composition of the hays as fed is given in table 1. The field-cured, field-baled hay as

TABLE 1  
*The mean chemical composition of the feeds fed<sup>a</sup>*

Feed	Composition						Carotene (mg./lb.)
	Dry matter (%)	Per cent of dry matter					
		Protein	Fat	Fiber	N.F.E.	Ash	
Field-cured field-baled alfalfa hay .....	88.52	21.13	1.64	29.85	37.01	10.37	3.59
Artificially-dried and ground alfalfa hay .....	91.96	19.57	2.47	28.81	38.83	10.32	39.09
Artificially-dried and pelleted alfalfa hay .....	89.60	20.74	3.23	25.96	39.89	10.18	53.80
Calf starter .....	87.50	22.25	4.00	5.81	61.34	6.60	0.89
Depletion ration .....	86.56	17.17	2.85	10.84	62.50	6.64	< 0.25

<sup>a</sup> The herd milk contained 170  $\gamma$  of carotene/lb. and 141  $\gamma$  of vitamin A/lb.

fed graded U. S. no. 2 alfalfa hay and contained a trace of grass, 2 per cent foreign material and 33 per cent leafiness of legumes. The per cent color of the hays was 50, 69 and 69, respectively, for the field-baled field-cured, the artificially-dried and ground and the artificially-dried and pelleted. Passing a 100-g. sample of the artificially-dried and ground hay through a series of U. S. standard

sieves resulted in 19 per cent of the sample being retained by a no. 10 sieve, 30 per cent by no. 40, 16 per cent by no. 60, 11 per cent by no. 80 and 6 per cent by no. 100, with 18 per cent passing through all sieves. The pellets were cylindrical in shape, 1.75 cm. in diameter and, on the average,  $2.2 \pm 0.6$  cm. in length.

Each calf received 8 lb. of its dam's colostrum the first day and 8 lb. of Holstein herd milk the second through the seventh days. Thereafter each received 7, 5, 4 and 2 lb. per day of the Holstein herd milk for the second, third, fourth and fifth weeks, respectively. From the seventh day all calves were allowed free access to water and hay and a maximum of 3 lb. of starter<sup>2</sup> per calf per day. All hays were fed so as to allow at least a 10 per cent weighback, since hay allowance affects consumption (25).

When each calf reached 106 days of age, it was placed on a low carotene ration so that the vitamin A storage could be determined. This ration, the "depletion" ration,<sup>3</sup> was fed at the rate of 2.67 lb. per 100 lb. of body weight. The amounts fed were adjusted to the weight of individual calves at successive 7-day intervals. When the blood plasma level of vitamin A reached less than  $4.0 \gamma$  per cent for two consecutive 7-day intervals, each calf was considered depleted of its vitamin A stores as defined by Jacobson *et al.* (6).

Scours, more fully discussed later, were treated with an initial intravenous injection of a 25 per cent solution of sulfamethazine at the rate of 26.3 ml. per 100 lb. of body weight and followed by oral administration of sulfamethazine at the rate of 6.5 g. per 100 lb. live weight daily for 2 additional days. In a few instances sulfaguanidine was used orally in place of the sulfamethazine.

*Samples and analyses.* Venous blood samples were obtained from each calf upon its arrival at the research barn and at 7-day intervals until the calf was depleted of its vitamin A stores as defined above. The blood was immediately chilled, centrifuged and a measured amount of plasma stored at  $-18^{\circ}$  C. and analyzed for carotene and vitamin A within 7 days after collection.

Each calf was weighed upon arrival at the research barn and at 7-day intervals thereafter. Feed intakes and refusals were weighed to the nearest 0.1 lb. and representative samples of all feeds fed were obtained at monthly intervals.

Analytical and statistical procedures used were essentially those employed previously (3).

#### RESULTS AND DISCUSSION

*Feed consumed.* The total amounts of milk and starter consumed by individual calves (table 2) were essentially the same. Also, average daily intakes of starter (fig. 1) were quite similar between groups.

<sup>2</sup> The starter contained per ton 519.5 lb. of cracked corn, 400 lb. crimped oats, 300 lb. wheat bran, 140 lb. linseed oil meal (expellar process), 280 lb. soybean oil meal (expellar process), 140 lb. dried skimmilk, 40 lb. B-Y 500-potency dried fermentation solubles, 0.5 lb. irradiated yeast (Standard Brands type 9-F), 20 lb. bone meal, 10 lb. iodized salt and 150 lb. cane molasses.

<sup>3</sup> A mixture by weight of one-third of dried beet pulp and two-thirds of a grain mixture composed of the following: 419.5 lb. ground barley, 500 lb. crimped oats, 500 lb. wheat bran, 150 lb. linseed oil meal (expellar process), 150 lb. soybean oil meal (expellar process), 200 lb. cane molasses, 40 lb. B-Y 500-potency dried fermentation solubles, 20 lb. steamed bone meal, 20 lb. iodized salt, and 0.5 lb. irradiated yeast (Standard Brands type 9-F) per ton of mixture.

TABLE 2  
Values for individual calves of criteria used

Repli- cate	Calf no. <sup>a</sup>	Feed consumed <sup>b</sup>		Total <sup>b</sup> carotene in hay consumed	Plasma carotene		Plasma vitamin A		Live weight		Depletion time
		Milk	Starter		Hay	Value at 7 d.	Mean value 14-105 d.	Value at 7 d.	Mean value 14-105 d.	Value at 7 d.	
	( <i>lb.</i> )	( <i>lb.</i> )	( <i>lb.</i> )	( <i>mg.</i> )	( <i>γ per cent</i> ) Field-cured field-baled alfalfa hay	( <i>γ per cent</i> ) Artificially-dried alfalfa hay	( <i>γ per cent</i> ) Artificially-dried and ground alfalfa hay	( <i>γ per cent</i> ) Artificially-dried and pelleted alfalfa hay	( <i>lb.</i> )	( <i>lb.</i> )	( <i>wt.</i> )
I	3	127	252	751	26	27	12.8	11.2	108	235	0.323
II	4	123	252	640	40	43	15.3	16.5	71	216	0.432
III	7	123	239	740	21	36	12.9	13.2	90	235	0.370
IV	10	137	249	1,073	19	35	16.2	14.4	107	267	0.358
V	15	130	231	818	20	45	16.8	12.5	105	233	0.336
Mean		128.0	244.6	804.4	25.2	37.2	14.80	13.56	96.2	237.2	0.3638
I	1	123	252	8,889	26	91	9.6	13.5	116	256	0.330
II	5	123	249	6,512	29	113	11.4	17.0	91	228	0.364
III	9	123	258	10,668	11	98	9.0	14.6	114	274	0.350
IV	12	123	246	8,576	15	81	11.0	13.5	98	242	0.336
V	13	126	214	3,768	22	72	8.0 <sup>d</sup>	12.4	93	191	0.254
Mean		123.6	243.8	7,682.6	20.6	91.0	9.80	14.20	102.4	238.2	0.3268
I	2	123	251	15,946	33	183	22.7	20.5	93	267	0.416
II	6	123	242	17,797	22	113	17.6	18.0	87	248	0.422
III	8	123	247	19,126	19	141	15.5	19.3	103	284	0.406
IV	11	123	250	17,587	21	136	11.1	20.6	106	287	0.400
V	14	128	239	16,646	22	182	19.2	21.7	98	283	0.425
Mean		124.0	245.8	17,420.4	23.4	151.0	17.22	20.02	97.4	273.8	0.4138

<sup>a</sup> Replicates I, III, IV, and V males; replicate II female.

<sup>b</sup> Total for 7 through 105 d. of age.

<sup>c</sup> Rate of gain using weekly liveweight from 7 days through 105 d. of age.

<sup>d</sup> Calculated missing value.

The pelleted hay was eaten at an earlier age (fig. 1) and in greater amounts during the experimental period, 7 through 105 days of age (table 2), than field-cured, field-baled hay or artificially-dried and ground hay. Since, within a particular hay group, calves which were large at birth tended to consume greater amounts of hay than small calves, an analysis of covariance was used in which the total amount of hay consumed was adjusted to the initial weight at 7 days of age. Both the total amount of hay and dry matter as hay were consumed in sig-

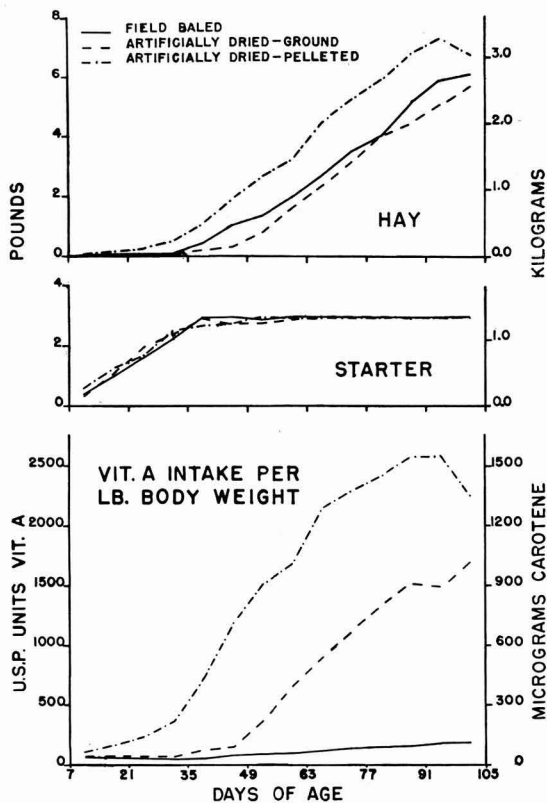


FIG. 1. The effect of type of hay on the weekly mean intake of vitamin A per lb. of body weight and on the weekly mean consumption of starter and hays by young dairy calves.

nificantly greater amounts ( $P < 0.01$ ) by calves fed the pelleted hay than by calves fed either the field-baled or ground hays. There was no appreciable difference in hay consumption between the latter two groups of calves.

There were marked differences in the total intake of carotene from the various hays (table 2). When both carotene and vitamin A content in the feeds fed were calculated to U.S.P. units of vitamin A and the daily intake of U.S.P. units of vitamin A was expressed as per pound of live weight (fig. 1), the differences between hay groups still were very apparent.

These data indicate the desirability of further investigation into factors in-

fluencing the palatability of roughages to the very young dairy calf. Although they were working with older animals than those used in this experiment, other workers (8, 25) have found a relationship between quality of field-cured alfalfa hays and consumption. The ground hay used in this experiment was clearly not as palatable to the calves as the pelleted hay. Whether pelleting improves palatability above that for long hay, such as field-baled, was not determined in this experiment because there were differences in hay quality. Besides possibly influencing palatability, pelleting may increase the quantity of roughage the young calf can consume by decreasing bulk. This appears to be important in the very young calf because of the relatively undeveloped capacity of the rumen (10). However, in experiments with dry calf starters (15), pelleting decreased consumption of the starter.

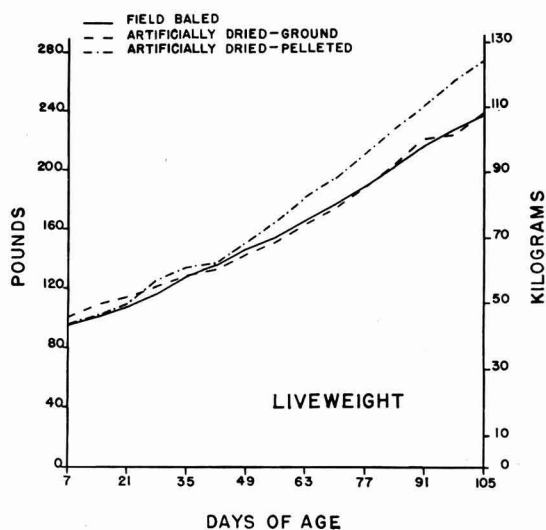


FIG. 2. The effect of type of hay on the mean live weight changes in young dairy calves.

The use of high quality hay accompanied by an early intake of the hay appears to be essential to maintain an adequate consumption of carotene in the young calf raised under limited whole milk systems. On the basis of physiological requirements for carotene, as recently proposed by Moore *et al.* (14), the U. S. no. 2 field-cured, field-baled alfalfa hay used in this experiment allowed only a borderline intake of carotene until the calf reached 6 wk. of age. In contrast, the pelleted hay allowed more than sufficient carotene and the ground hay an intermediate amount.

*Live weight.* An analysis of the live weight data (table 2 and fig. 2), which included adjustment for differences between individual calves in weight at 7 days of age, showed that those calves fed the pelleted hay made greater total gains ( $P < 0.01$ ) and more rapid rates of gain ( $P < 0.01$ ) than those calves fed the other two types of hay. No significant differences were found between calves receiving the field-cured, field-baled and artificially-dried and ground hays.

Adjustment of the live weight data by multiple covariance for differences in dry matter consumed as hay as well as for differences in weight at 7 days of age revealed no significant differences between hay groups. Therefore, the differences in live weight, as observed in this experiment, were largely attributable to differences in amounts of dry matter consumed as hay and not to differences in nutritive value of the hays.

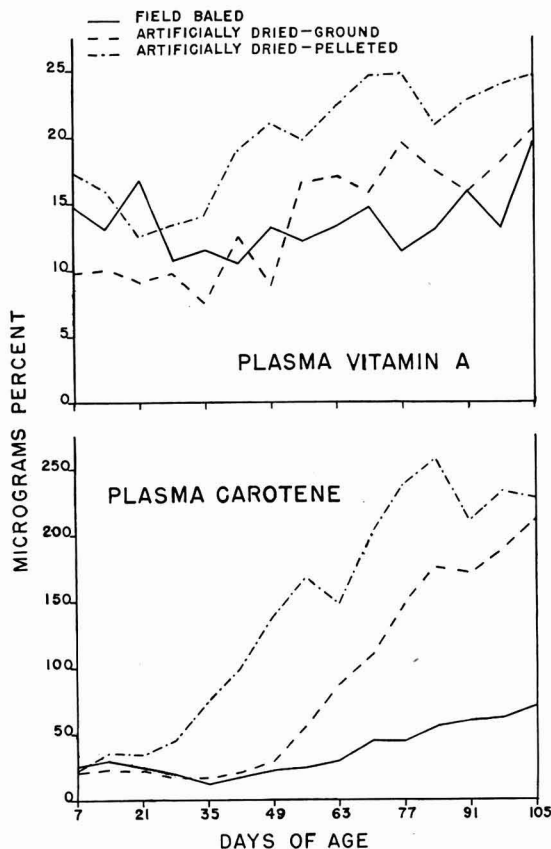


FIG. 3. The effect of type of hay on the mean carotene and vitamin A plasma levels in young dairy calves.

These results indicate the importance of palatability of roughage fed to young dairy calves to insure maximum consumption accompanied by optimum performance such as increase in live weight. Although the hays did not differ markedly in their proximate composition (table 1) and live weight changes did not indicate nutritive differences between the hays, possible differences as might be revealed by more precise measurements than those used should not be overlooked. There were no demonstrable responses in live weight changes due to widely different carotene intakes between groups of calves. Live weight does



content of a ration for dairy calves except at very low intakes of vitamin A accompanied by blood plasma levels of less than 5.0  $\gamma$  per cent (6, 7, 11). Various workers (9, 19, 22) have reported greater gains in live weight due to supplements of carotene or vitamin A; however, adjustments of the data for feed intakes were not undertaken.

*Carotene and vitamin A metabolism.* Blood plasma carotene and vitamin A levels (table 2 and fig. 3) differed among hay groups. The calves fed the pelleted hay had the highest plasma carotene values, those on ground hay intermediate values and those on field-baled hay the lowest values. These differences, when adjusted to the initial plasma carotene value at 7 days of age, were statistically significant ( $P < 0.01$ ). These plasma carotene levels were related directly to the carotene intake, because an additional adjustment of these values for the mean carotene intake per pound of mean body weight accounted for the differences between hay groups.

The differences in blood plasma vitamin A, although not of the same magnitude as those for carotene, followed somewhat the same trends between hay groups. Upon statistical analysis, the only difference found was between those calves fed the pelleted hay and those fed the other two hays ( $P < 0.01$ ). The carotene intake per pound of body weight accounted for a part of the difference in vitamin A levels in the plasma, for when these values were corrected for both the plasma vitamin A level at 7 days of age and the mean carotene intake, the differences between hay groups were reduced ( $P < 0.10$ ).

As determined by feeding a depletion ration after the calf reached 105 days of age and by weekly analyses of the plasma vitamin A until the level reached less than 4.0  $\gamma$  per cent for 2 consecutive wk., the amount of vitamin A storage differed between hay groups ( $P < 0.01$ ). The average depletion time (table 2) was 4.8 wk. for calves fed the field-cured, field-baled hay, 8.2 wk. for the artificially-dried and ground hay and 11.2 wk. for the artificially-dried and pelleted hay. Adjustment of the depletion time for the mean carotene intake as hay per pound of body weight for the period 7 through 105 days reduced the differences between hay groups ( $P < 0.05$ ). Graphic illustration (fig. 4) of the relationship between depletion time and mean carotene intake from hay per pound of the mean live weight indicated a linear relationship between these variables. When variations within hay groups were accounted for, statistical analysis showed this relationship not to be significant. When all data were grouped, which assumes that the carotene from the three hays was of equal value to the calves, this trend proved to be significant ( $P < 0.01$ ).

The criteria used for measuring the differences in the vitamin A metabolism were sensitive to differences in carotene intakes from the various hays. As expected, plasma carotene levels followed carotene intake. Similarly, the depletion time was dependent largely upon carotene intake and the data agree with those of Jacobson *et al.* (6). Graphic inspection of the data reported herein combined with that of Jacobson *et al.* (6) indicated a more efficient use of carotene when the intake was less than 50  $\gamma$  per pound of body weight than at higher levels. Differences between sex in depletion time might be considered

not appear to be a critical measurement of the adequacy of the vitamin A in future investigations, since the female replicate averaged 10.0 wk. and the male replicates 7.6 wk. This difference may be due largely to differences in rate of gain in live weight, but other factors inherent to sex might well be considered.

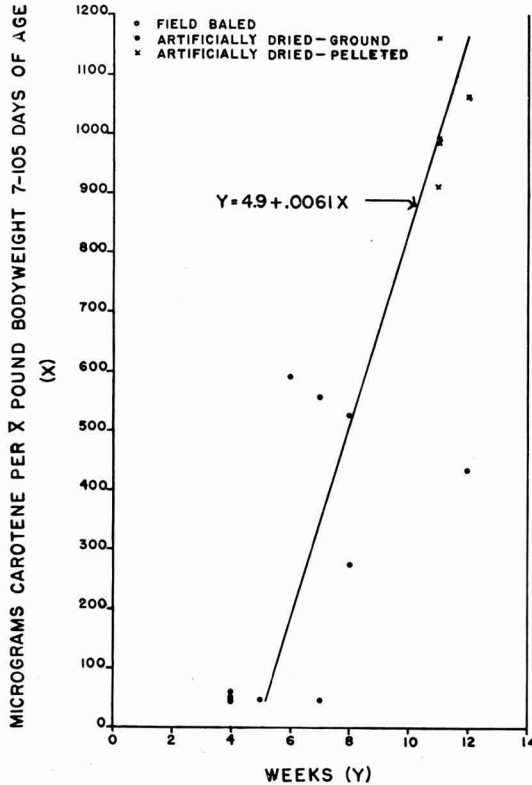


FIG. 4. The effect of mean carotene intake from hay per lb. of average body weight of calves from 7 through 105 days of age on the depletion time.

The differences in blood plasma vitamin A levels were not as apparent between hay groups as were those of the blood plasma carotene levels and depletion time. The reasons why there were not more marked differences in blood plasma vitamin A levels between those calves receiving the field-baled hay and the ground hay is not very clear. Possible differences in the physiological make-up of the calves (12) might be an important factor. This might be eliminated by grouping calves of similar blood plasma levels of vitamin A after a standardization period into the same replicate at the start of the experiment. Other factors, such as prenatal influence (21, 26) and method of determination of blood plasma vitamin A (1), need further investigation to allow more precise measurement.

*Health observations.* Three calves fed the field-baled hay had scours for a

total of 12 days, three calves fed the ground hay scoured for a total of 8 days and three calves fed the pelleted hay scoured for a total of 9 days. Analyses of the data revealed no difference due to treatment.

The calves fed the ground hay salivated excessively during the hay-feeding period and rolled their tongues frequently. Calf no. 1 had three attacks of bloat over a 9-day period immediately prior to being placed on the depletion ration. Calf no. 13 developed the habit of filling excessively on water. Upon being placed on the depletion ration, this habit disappeared. Although this calf's performance was poor, especially in regard to live weight, histological examination of all major tissues upon slaughter at the termination of the depletion period revealed no abnormalities which might have contributed to the poor performance. Therefore, the observations obtained from this calf were included in the data. Statistical analyses with this calf removed from the data did not change the interpretation.

Since the data obtained were with Holstein calves, the applicability of these results to other breeds, especially Guernseys and Jerseys, cannot be assumed. Experiments are now being planned to include these breeds of calves.

#### SUMMARY

The relative values of field-cured, field-baled hay, artificially-dried and ground hay and artificially-dried and pelleted hay as sources of carotene and roughage for the young calf were studied in 15 Holstein calves. Measurements of daily feed intakes and live weight changes and blood plasma carotene and vitamin A levels at 7-day intervals were made for each calf from 7 through 105 days of age and, in addition, the time to deplete individual calves of their vitamin A stores after 105 days of age was determined.

The artificially-dried and pelleted hay was consumed in greater amounts accompanied by higher blood plasma levels of carotene and vitamin A as well as greater increases in live weight than the field-cured, field-baled hay or the artificially-dried and ground hay. The only demonstrable difference in these measurements between the field-baled and ground hays was in the higher blood plasma carotene levels of those calves fed the ground hay.

The time required to deplete the calves of their vitamin A stores after each calf reached 105 days of age was 4.8 wk. for those calves fed the field-cured, field-baled hay, 8.2 wk. for those fed the artificially-dried and ground hay and 11.2 wk. for those fed the artificially-dried and pelleted hay.

With the exception of occurrence of bloat in one calf fed the ground hay and of excessive salivation in all calves fed the ground hay, the remaining observations as to the health of the calves were not associated with hay groups.

#### ACKNOWLEDGMENTS

The authors are grateful to M. W. Dicks and B. A. Donohue for the technical assistance during the course of this experiment, to C. I. Bliss for aid in the statistical analysis and interpretation of the data and to H. J. Fisher for the proximate analyses of the feeds.

## REFERENCES

- (1) ALLEN, R. S., WISE, G. H., AND JACOBSON, N. L. Occurrence in Blood Plasma from Certain Dairy Calves of Factors which Interfere with the Color Reactions of Activated Glycerol Dichlorohydrin with Vitamin A and Carotenoids. *J. Dairy Sci.*, **32**: 688-694. 1949.
- (2) ARCHIBALD, J. G., BLAISDELL, M. L., AND STAPLETON, H. N. A Study of Quality in Roughages; Composition, Palatability, and Nutritive Value of Hays as Affected by Curing, Harvesting, and Storing Procedures. *Mass. Agr. Expt. Sta. Ann. Rpt.*, 1948-49.
- (3) EATON, H. D., SPIELMAN, A. A., JOHNSON, R. E., AND MATTERSON, L. D. The Utilization of Fetal Stores of Vitamin A by the Newborn Calf. *J. Dairy Sci.*, **32**: 764-768. 1949.
- (4) HIBBS, J. W., AND POUNDEN, W. D. The Influence of the Ration and Early Rumen Development on the Changes in the Plasma Carotenoids, Vitamin A and Ascorbic Acid of Young Dairy Calves. *J. Dairy Sci.*, **31**: 1055-1061. 1948.
- (5) HUFFMAN, C. F. Roughage Quality and Quantity in the Dairy Ration. A Review. *J. Dairy Sci.*, **22**: 889-966. 1939.
- (6) JACOBSON, W. C., CONVERSE, H. T., AND MOORE, L. A. Effects of Vitamin A and Carotene Intake on Depletion Time of Young Dairy Calves. *J. Dairy Sci.*, **32**: 418-428. 1949.
- (7) JUNGHER, E. L., HELMBOLDT, C. F., AND EATON, H. D. Unpublished data. Storrs (Conn.) Agr. Expt. Sta., Storrs, Conn.
- (8) KELKAR, C. N., AND GULLICKSON, T. W. Importance of Hay Quality as Indicated by Feeding Trials with Identical Twin Dairy Heifers. *J. Dairy Sci.*, **33**: 288-292. 1950.
- (9) KEYES, E. A., BECHTEL, S. I., AND THORP, W. T. S. The Effect of Additional Vitamins A and D in the Standard Calf Starter (Abs.). *J. Dairy Sci.*, **26**: 741-742. 1943.
- (10) LAGERLOF, N. *Skand. Vet. Tidskr.*, **19**: 253-365. 1929. (As cited by Savage, E. S. and McCay, C. M. The Nutrition of Calves; a Review. *J. Dairy Sci.* **25**: 595-650. 1942.)
- (11) LEWIS, J. M., AND WILSON, L. T. Vitamin A Requirements in Calves. *J. Nutrition*, **30**: 467-475. 1945.
- (12) MATHER, R. E. Genetic Variation in the Levels of Blood Plasma Carotene and Vitamin A in Dairy Cattle (Abs.). *J. Dairy Sci.*, **31**: 658. 1948.
- (13) MONROE, C. F., HILTON, J. H., HODGSON, R. E., KING, W. A., AND KRAUSS, W. E. The Loss of Nutrients in Hay and Meadow Crop Silage during Storage. *J. Dairy Sci.*, **29**: 239-256. 1946.
- (14) MOORE, L. A., SYKES, J. F., JACOBSON, W. C., AND WISEMAN, H. G. Carotene Requirements for Guernsey and Jersey Calves as Measured by Spinal Fluid Pressures. *J. Dairy Sci.*, **31**: 533-538. 1948.
- (15) NEWMAN, P. E., AND SAVAGE, E. S. The Use of Yeast in Calf Meals and Pellets. *J. Dairy Sci.*, **21**: 161-167. 1938.
- (16) NORTON, C. L., AND EATON, H. D. Dry Calf Starters for Dairy Calves. *Cornell Agr. Expt. Sta. Bull.* 835. 1946.
- (17) POUNDEN, W. D., AND HIBBS, J. W. The Influence of the Ration and Rumen Inoculation on the Establishment of Certain Microorganisms in the Rumen of Young Calves. *J. Dairy Sci.*, **31**: 1041-1050. 1948.
- (18) POUNDEN, W. D., AND HIBBS, J. W. The Influence of the Ratio of Grain to Hay in the Ration of Dairy Calves on Certain Microorganisms. *J. Dairy Sci.*, **31**: 1051-1054. 1948.
- (19) ROSS, R. H., AND KNOTT, C. B. The Effect of Supplemental Vitamin A upon Growth, Blood Plasma, Carotene, Vitamin A, Inorganic Calcium, and Phosphorous of the Holstein Heifers. *J. Dairy Sci.*, **31**: 1062-1067. 1948.
- (20) ROSS, V. E. An Economic Evaluation of the Barn Finishing Method of Harvesting Hay. Unpublished M.S. thesis, University of Connecticut, Storrs, Conn. 1950.

- (21) SPIELMAN, A. A., THOMAS, J. W., LOOSLI, J. K., NORTON, C. L., AND TURK, K. L. The Placental and Mammary Transmission and Fetal Storage of Vitamin A and Carotene in the Bovine. *J. Dairy Sci.*, **29**: 707-715. 1946.
- (22) SPIELMAN, A. A., EATON, H. D., LOOSLI, J. K., AND TURK, K. L. The Effect of Prepartum Vitamin A Supplementation on the Health and Performance of the Young Calf. *J. Dairy Sci.*, **32**: 367-374. 1949.
- (23) TURK, K. L., AND BURKE, J. D. Raising Dairy Calves and Heifers. Cornell Extension Bull. 761. 1949.
- (24) TURK, K. L., MORRISON, S. H., TRIMBERGER, G. W., REID, J. T., NORTON, C. L., SPIELMAN, A. A., BLASER, R. E., AND HARTWIG, H. B. The Effects of Curing Methods upon the Feeding Value of Hay. Mimeographed. Animal Husbandry and Agronomy Depts., Cornell University. 1950.
- (25) WILLARD, H. S. Standardizing Hay Palatability Trials with Calves. *J. Dairy Sci.*, **32**: 10-16. 1949.
- (26) WISE, G. H., CALDWELL, M. J., HUGHES, J. S. The Effect of Prepartum Diet of the Cow on the Vitamin A Reserves of Her Newborn Offspring. *Science*, **103**: 616-618. 1946.

## A COMPARISON OF DRY SWEET CREAM BUTTERMILK AND NON-FAT DRY MILK SOLIDS IN BREADMAKING<sup>1, 2, 3</sup>

J. V. REGER, W. B. COMBS, S. T. COULTER AND R. B. KOCH

*Divisions of Dairy Husbandry and Agricultural Biochemistry,  
University of Minnesota, St. Paul*

The use of dry sweet cream buttermilk as a concentrated source of defatted milk solids recently has received increased attention from many food processors. This interest in sweet cream buttermilk is of significant economic value, particularly, in large butter producing areas where an increasing proportion of this milk by-product is being processed for use as human food. From the standpoint of general composition, sweet cream buttermilk powder closely resembles that of non-fat dry milk solids except that it contains a greater percentage of fat and fat-like substances.

The authors are not aware of any published experimental investigations on the baking quality of sweet cream buttermilk powder or the effect which variations in processing procedures and storage conditions may have on its baking properties. Hunziker (6) has discussed its potentialities and use in bread and cracker making; however, no experimental work was cited. The use of non-fat dry milk solids in bakery products has been studied extensively, but only certain of the pertinent investigations will be cited in this article.

Grewe and Holm (3) were among the early investigators to note that when skimmilk was preheated at normal pasteurization temperatures (62° C. for 30 min.), dried and incorporated into bread dough, a definite dough-softening action occurred. They found that this defect did not occur when the milk powders utilized were preheated at temperatures above 70° C. Larsen *et al.* (8) established the minimum preheating treatment necessary to insure optimum baking quality of non-fat dry milk solids at 74° C. for 30 min., or about 90° C. when the flash method of preheating was employed.

The agent or agents responsible for the dough-softening action were shown by Stamberg and Bailey (9) and confirmed by Harland *et al.* (5) to reside in the whey proteins of the milk.

Harland *et al.* (4) developed a rapid turbidimetric method for determining the serum protein content of nonfat dry milk solids and utilized this test as a means of predicting the baking quality of this milk product. The test is based on the fact that the baking quality of defatted milk powders is dependent upon, or at least coincidental with, the extent of heat denaturation of the whey proteins. By employing this test, Harland *et al.* (4) were able to classify properly

Received for publication, August 18, 1950.

<sup>1</sup> Paper 2526, Scientific Journal Series, Minnesota Agricultural Experiment Station.

<sup>2</sup> Data presented in this paper are taken from a thesis submitted by J. V. Reger in partial fulfillment of requirements for a Master of Science degree, University of Minnesota, St. Paul, 1949.

<sup>3</sup> This investigation was aided by a grant from the Kraft Foods, Inc., Chicago, Illinois.

29 out of 35 samples of good and poor baking quality non-fat dry milk solids using 2.2 mg. of serum protein nitrogen per gram of milk powder as the maximum amount of undenatured whey protein associated with good baking quality.

Van Dam *et al.* (10), as well as Hutchinson *et al.* (7), obtained loaves bigger than water control bread by the addition to the bread dough of non-fat dry milk solids plus fat in the form of an emulsion.

The objective of this experiment was to secure some information on the baking quality of sweet cream buttermilk powder as affected by (a) variations in storage conditions and processing procedures and (b) presence of neutralizer.

#### MATERIALS

Commercially prepared dried sweet cream buttermilk and non-fat dry milk solids were obtained from Kraft Foods, Inc. Separate portions of these milk powders were adjusted by exposure to humid air to secure samples varying in moisture content from 3.0 to 6.0 per cent, as determined by the toluol distillation method (2). Samples at each moisture level were stored at 6.6 to 8.3° C. and at 37.5° C., with controls maintained at below -10° C.

Other lots of dried milk were manufactured with the experimental equipment of the University of Minnesota. In all cases, both the buttermilk powders and non-fat dry milk solids originated from the same lot of mixed whole milk.

Preparation of neutralized buttermilks for drying was accomplished by inoculation of sweet cream buttermilk with a lactic acid culture at a 3 per cent level and allowing it to incubate at 21° C. When 0.02, 0.05 and 0.10 per cent developed acidities were obtained, separate portions were withdrawn, neutralized to the original acidity with a 10 per cent solution of the U.S.P. grade NaOH or with a 10 per cent lime solution and pasteurized at 74° C. for 30 min.

Butter oil was prepared by decanting and filtering the fat layer from melted butter.

Lecithin used was a commercial product (Arlec) prepared from soybean (67 per cent phospholipid).

*Baking quality of fresh and stored sweet cream buttermilk powders as compared with similarly treated non-fat dry milk solids.* Bread doughs were prepared using 100 g. of bakers patent hard red spring wheat four, (14.5 per cent protein on a 14 per cent moisture basis), 3 g. yeast, 2 g. salt, 5 g. sugar and 6 g. of the dry milk under test. To these ingredients sufficient water was added to give the dough the proper consistency. The baking procedure used was essentially the same as that recommended by the American Association of Cereal Chemists (1) for the preparation of pup loaves. The breads were scored using an arbitrary system which allows 10 points for a perfect score for grain, texture, crust color and crumb color. Type, which is a measure of symmetry and break was allowed 10 points for a perfect score and was determined by comparing experimental pup loaves with a standard set of photographs.

Loaf volumes were determined on the day the bread was baked, as follows: The pup loaf was placed in a constant volume container and rape seed was allowed to fill the container to overflowing. The excess seed was removed by a

scraper. The pup loaf then was removed and the rape seed poured into a volumetric cylinder. The reading obtained was converted to loaf volume in cubic centimeters by means of a suitable conversion table.

Bake tests were made on all of the fresh milk powders involved in this experiment and the results obtained are summarized in tables 1, 3, 4 and 5. Examination of these data showed that spray-dried sweet cream buttermilk consistently produced larger loaf volumes than when like samples of non-fat dry milk solids were included in the bread formula. Roller-dried buttermilk powders also produced an increase in loaf volume as compared with either spray or roller-dried non-fat milk solids; however, this increase was not as consistent nor as pronounced as when the spray-dried buttermilk powders were utilized.

The results obtained from the bake tests of the stored milk powders are recorded in tables 1 and 2. These data show that storage at 37.5° C. for 1 yr. was

TABLE 1

*Comparison of loaf volumes secured using dry sweet cream buttermilk and non-fat dry milk solids when fresh and after storage at 5 to 7.22° C. at three moisture levels for periods of up to 1 yr.  
(6 g. of the dry milk were used with each 100 g. of flour)*

Drying method	Moisture level <sup>b</sup>	Loaf volume <sup>b</sup> (c.c.) after storage time of:				
		0 mo.	1 mo.	2 mo.	6 mo.	12 mo.
Sweet cream buttermilk powder <sup>c</sup>						
Spray	Low	735	748	730	735	742
	Medium	733	730	734	740	747
	High	734	738	743	731	727
Roller	Low	700	721	693	700	705
	Medium	718	713	700	701	715
	High	709	707	701	690	.....
Non-fat dry milk solids <sup>c</sup>						
Spray	Low	685	680	680	689	690
	Medium	694	687	686	700	699
	High	684	692	684	689	695
Roller	Low	699	704	678	700	696
	Medium	697	710	687	698	702
	High	696	699	697	697	702

<sup>a</sup> Average of 4 loaves.

<sup>b</sup> Low (3-4%), medium (4-5%), high (5-6%).

<sup>c</sup> Commercially prepared.

detrimental to the baking quality of the milk powders, particularly those samples having an initial moisture content of over 4 per cent. Utilization of the latter powders in bread making resulted in a definite decrease in loaf volume, grain and texture. It also was observed that any discoloration present in the milk powders carried over into the finished loaf. The extent to which these deteriorative changes occurred in the baking quality was approximately the same for corresponding samples of both the sweet cream buttermilk powders and non-fat dry milk solids, regardless of whether they were spray or roller-dried. Storage at 6.6 to 8.3° C. was found not to materially affect the baking quality of any of the samples.



TABLE 2  
*Effect of storage for 1 yr. at 37.5° C. on the baking quality of dry sweet cream buttermilk as compared with similarly treated nonfat dry milk solids*  
 (av. of 4 loaves)  
 (6 g. of the dry milk were used with each 100 g. of flour)

Drying method	Initial moisture (%)	Lipide content (%)	Change in loaf volume (Control minus 1 yr. sample)		Grain		Texture		Crust color		Type		Crumb color
			Control	1 yr.	Control	1 yr.	Control	1 yr.	Control	1 yr.	Control	1 yr.	
Spray	3.15	5.98											
	4.89		-19	8.5	9	8.5	8.5	8.5	Dark	Dark	8.5	8.5	8.5 C <sup>c</sup>
	5.97		-17	8.5	8.5	8.5	8.5	8.5	Dark	Dark	8.5	8.5	8.5 C
Roller	3.91	5.86	-82	8.75	8.5	8.5	8	8	Dark	bV. Dark	9	7.5	8.5
	4.92		-20	8.5	8.5	8.5	8	8	Dark	Dark	8	8	8.5
	6.53		-73	8.5	8.5	8.5	8	8	Dark	V. Dark	8	7.5	8.5
Spray	3.05	0.90	-90	8.5	8.5	8.5	8	8	Dark	V. Dark	8	7.25	8.5
	4.85												4 B
	5.94												3 RB
Roller	2.56	0.75											
	4.79		-7	8.5	8.5	8.5	8.5	8.5	Dark	Dark	7.75	7.75	8.5 C
	5.94		-26	8.5	8.5	8.5	8.0	8.0	Dark	Dark	8	7.75	8.5 C
Total possible score			-57	8.0	8.0	8.0	8.0	8.0	Dark	Dark	8	7.5	5 B
			-18	8.5	8.0	8.5	8.0	8	Dark	Dark	7.5	7.5	8.5 C
			-25	8.5	8.0	8.5	8.0	8	Dark	Dark	7.5	7.5	8.5 C
		-66	8.5	8.5	8.5	8.5	8	Dark	V. Dark	7.5	7	8.5	
													4 C

<sup>b</sup> V—Very  
<sup>c</sup> C—Creamy  
<sup>a</sup> Commercially prepared

YB—Yellowish brown  
 RB—Reddish brown  
 B—Brown

*Effect of added heat treatment on the baking quality of resultant dried milks.* In the commercial production of butter, sweet cream usually is pasteurized at approximately 74° C. for 30 min. prior to churning. Re-pasteurization of this buttermilk often is necessary when it is to be utilized in the production of sweet cream buttermilk powder. The results obtained from the bake tests of the resultant buttermilk powders as compared with non-fat dry milk solids are summarized in table 3. These data show that the additional heating of the fluid

TABLE 3

*Influence of preheating treatment used in the preparation of spray- and roller-dried sweet cream buttermilk<sup>a</sup> and non-fat dry milk solids on the loaf volume of breads made utilizing these powders (av. of 4 loaves)*  
(6 g. of the dry milk were used with each 100 g. of flour)

Milk powder utilized	Sweet cream buttermilk <sup>b</sup>				Non-fat dry milk solids <sup>b</sup>			
	Spray		Roller		Spray		Roller	
Method of drying								
Method of preheating	HTST <sup>c</sup>	30 min. hold	HTST	30 min. hold	HTST	30 min. hold	HTST	30 min. hold
Preheating temp. (°C.)	Loaf volume, (c.c.)							
74.0	725	745	709	715	618	682	614	673
79.4	716	715	691	710	669	675	663	658
85.0	704	725	701	692	668	690	674	665
90.5	704	711	702	710	685	703	662	664
93.3	692	708	678	703	681	677	665	672

<sup>a</sup> Sweet cream buttermilk obtained from sweet cream which was pasteurized at 74.0° C. for 30 min. prior to churning.

<sup>b</sup> (m)—Samples manufactured at Univ. of Minn.

<sup>c</sup> HTST—High temp.—short time method.

buttermilk, whether following the holding method for a 30-min. period or the continuous heating method (holding time approximately 14.5 sec.), at temperatures of 74, 79.4, 85.9, 90.5 or 93.3° F., did not materially affect the baking

TABLE 4

*Comparison of the baking qualities of dry sweet cream buttermilk, non-fat dry milk solids and dry skimmilk containing added butter-oil*  
(6 g. of the dry milk were used with each 100 g. of flour)

Milk powder utilized <sup>a</sup>	Lipide content	Loaf volume	Type	Crust color	Grain score	Texture score	Crumb color
	(%)	(c.c.)					
Sweet cream		705	7.5	Dark	8	7.5	8.5 Creamy
Buttermilk	5.86	695	7.5	Dark	8.5	7.5	8.5 Creamy
Non-fat dry milk solids	.66	650	7	Dark	8	7	8.5 Creamy
		635	7	Dark	8	7	8.5 Creamy
Dried skimmilk plus butter-oil	5.62	650	7	Dark	8	7	8.5 Creamy
		653	7	Dark	8	8	8.5 Creamy
Total possible score			10		10	10	10

<sup>a</sup> Samples manufactured at the Univ. of Minn.

TABLE 5  
*Comparison of the baking quality of dry sweet cream buttermilk, nonfat dry milk solids and dry skim milk containing added phospholipide material (Arlec) emulsified in buttersols*  
*(6 g. of the dry milk were used with each 100 g. of flour)*

Milk powder utilized <sup>a</sup>	Method dried	Lipide content (%)	Loaf volume <sup>b</sup>	Type <sup>b</sup>	Crust color <sup>b</sup>	Grain score <sup>b</sup>	Texture score <sup>b</sup>	Crumb color <sup>b</sup>
Sweet cream buttermilk	Spray	7.83	Flour 1	8	Dark	8.5	8.5	8.5 Creamy
			Flour 2	8	Dark	8.5	8.5	8.5 Creamy
Nonfat dry milk solids	Spray	.66	Flour 1	8.5	Dark	8.5	8.5	8.5 Creamy
			Flour 2	7.75	Dark	8.0	7.5	8.5 Creamy
Dried skim milk plus butteroil and phospholipide (Arlec)	Spray	7.77	Flour 1	8	Dark	8.5	7.25	8.5 Creamy
			Flour 2	8	Dark	8.0	7.5	8.5 Creamy

<sup>a</sup> Samples manufactured at Univ. of Minn.

<sup>b</sup> Average of duplicate loaves.

quality of the dried product. This does not suggest, therefore, that the optimum heat treatment necessary to insure maximum loaf volume had already been obtained from the pasteurization of the cream.

*Effect of butter oil and phospholipid material on the baking quality.* Since, from a general composition standpoint, buttermilk differs from skimmilk mainly in the content of fat and fat-like substances, it was felt that perhaps these constituents were responsible for the observed differences in loaf volume. The results recorded in table 4 were obtained from bake tests utilizing (a) non-fat dry milk solids, (b) sweet cream buttermilk powder and (c) non-fat dry milk solids containing butter oil in an amount equivalent to that present in the buttermilk. These data show that the addition of butter oil had little or no influence on the loaf volume. In another trial, 0.17<sup>4</sup> per cent additional phospholipid material (Arlec) was incorporated with butter oil and emulsified in skimmilk before drying. The product containing the added phospholipid material when used in breadmaking gave loaves having essentially the same volume as those containing dry sweet cream buttermilk. These data are shown in table 5. This is evidence that the greater loaf volume secured with dry sweet cream buttermilk as compared to non-fat dry milk solids is due to the additional phospholipid material contained in the buttermilk.

*Influence of small amounts of neutralizer on the baking quality of buttermilk powders.* The results summarized in table 6 were obtained from bake tests

TABLE 6  
Effect of neutralization of buttermilk on the loaf volume of breads made utilizing spray and roller-dried products<sup>a</sup>  
(6 g. of the dry milk were used with each 100 g. of flour)

Method dried	Amount of developed acidity neutralized	Type neutralizer	Loaf volume <sup>a</sup>	
			Fresh powder	(c.c.) Powder after storage at 37° C. for 60 d.
Spray	Unneutralized		740	715
		0.02	733	700
	0.05	NaOH	715	715
		Ca(OH) <sub>2</sub>	725	730
		NaOH	723	700
		Ca(OH) <sub>2</sub>	718	723
	0.10	NaOH	715	705
		Ca(OH) <sub>2</sub>	683	678
Roller	Unneutralized		703	715
		0.02	703	715
	0.05	NaOH	703	710
		Ca(OH) <sub>2</sub>	703	688
	0.10	NaOH	690	708
		Ca(OH) <sub>2</sub>	695	700

<sup>a</sup> Average of 4 loaves.

of doughs supplemented with buttermilk powders prepared from ripened buttermilk neutralized with either caustic soda or lime. These data indicate that the

<sup>4</sup> It was assumed that buttermilk obtained from the churning of 30 per cent sweet cream contained 0.17 per cent more phospholipid material than did the corresponding skimmilk.

baking quality is not influenced by the presence of small amounts of neutralizer, even in those samples stored at 37.5° C. for 2 mo.

*Use of the serum protein turbidity method as a means of predicting the baking quality of badly deteriorated milk powders.* Application of the serum protein turbidity test was made to control samples and like samples which showed a marked decrease in baking quality as a result of adverse storage conditions. The results obtained are recorded in table 7. These data show no significant

TABLE 7

*Effect of storage at 37.5° C. on the serum protein content of spray and roller-dried sweet cream buttermilk and non-fat dry milk solids*

Drying method	Lot no.	Serum protein nitrogen	
		Initial	After 1 yr.
			(mg./g.)
			Sweet cream buttermilk powder <sup>b</sup>
Spray	1	0.96	0.95
	1H <sup>a</sup>	0.96	0.96
	2	0.48	0.36
	2H	0.48	0.60
Roller	2	0.48	0.48
			Non-fat dry milk solids <sup>b</sup>
Spray	1	1.02	1.02
	1H	1.02	0.96
	2	0.84	0.84
	2H	0.84	1.02
Roller	2	0.84	0.95

<sup>a</sup> H—High moisture (5–6%).

differences in the whey protein content of any control sample and the corresponding sample after storage; consequently, it is concluded that a decrease in baking quality as a result of storage cannot be detected by the use of this test. Thus, this test, when applied to stored powders, may indicate acceptable baking quality when actually the baking quality is inferior due to deteriorative changes.

## SUMMARY

Roller or spray-dried sweet cream buttermilk powder, when used in breadmaking at the rate of 6 g. per 100 g. of flour, gave consistently greater loaf volume than like samples of non-fat dry milk solids. Non-fat dry milk solids containing 0.17 per cent added soybean lecithin (Arlec) produced loaves having the same volume as those made with sweet cream buttermilk.

The baking quality of milk powders may deteriorate in storage, the rate of deterioration increasing with increase in moisture content of the powder and in storage temperature. The serum turbidity test, when applied to dry sweet cream buttermilk and non-fat dry milk solids, does not detect loss of baking quality due to storage deterioration.

Additional preheating treatment of buttermilk secured from cream pas-

teurized at 74° C. for 30 min. did not affect the quality of the powder for use in breadmaking.

Neutralization of ripened buttermilk to an amount equivalent to 0.10 per cent acidity with either caustic soda or lime did not significantly change the quality of the powder for use in breadmaking.

## REFERENCES

- (1) AMERICAN ASSOCIATION OF CEREAL CHEMISTS.  
*Cereal Laboratory Methods*, 5th ed. pp. 137-143. 1947.
- (2) AMERICAN DRY MILK INSTITUTE, INC.  
The Grading of Non-fat Dry Milk Solids and Sanitary and Quality Standards including Standard Methods of Analysis. American Dry Milk Institute, Inc., B. 911, 1948.
- (3) GREWE, E., AND HOLM, G. E.  
Effect of Variation in the Method of Manufacture on the Baking Quality of Dry Skimmilk. *Cereal Chem.*, 5: 461-469. 1928.
- (4) HARLAND, H. A., AND ASHWORTH, U. S.  
A Rapid Method for Estimation of Whey Proteins as an Indication of Baking Quality of Non-fat Dry Milk Solids. *Food Research*, 12 (3): 247-251.
- (5) HARLAND, H. A., ASHWORTH, U. S., AND GOLDING, N. S.  
Chemical Factors Affecting the Baking Quality of Dry Milk Solids. III. The Effect of Several Milk Fractions on Loaf Volume. *Cereal Chem.*, 20: 535-541. 1943.
- (6) HUNZIKER, O. F.  
*Condensed Milk and Milk Powder*. 7th ed., pp. 512-513. The author, La Grange, Ill. 1949.
- (7) HUTCHINSON, R. C., WEST, H. E., AND JEFFREYS, J. T.  
The Addition of Skimmilk Powder to Australian Bread. *Australian J. Council Science, Ind. Research*, 20 (4): 483-499. 1947.
- (8) LARSEN, R. A., JENNESS, R., AND GEDDES, W. F.  
Effect of Heat Treatment of Separated Milk on the Physical and Baking Properties of Doughs Enriched with Dry Milk Solids. *Cereal Chem.*, 26: 189-201. 1949.
- (9) STAMBERG, O. E., AND BAILEY, C. H.  
The Effect of Heat Treatment of Milk in Relation to Baking Quality as Shown by Polarograph and Farinograph Studies. *Cereal Chem.*, 19: 507-517. 1942.
- (10) VAN DAM, B., ABMA, R. R., AND REVALTIER-WARFFENIUS, J. D.  
The Use of Dried Skim in Bread Making. *Netherlands Milk Dairy J.*, 2: 148-161. 1948. (Original not seen) Abstract in *Chem. Abs.*, 43: 1871. 1949.

# B-VITAMIN STUDIES IN CALVES. I. THE RELATION BETWEEN AGE OF CALF AND LEVELS OF THIAMINE, RIBOFLAVIN AND NICOTINIC ACID FOUND IN THE DIGESTIVE TRACT<sup>1, 2</sup>

E. M. KESLER AND C. B. KNODT

*The Pennsylvania Agricultural Experiment Station, State College*

## INTRODUCTION

It has been well established that synthesis of the members of the B-complex occurs in the digestive tract of the adult ruminant. The literature relative to rumen and intestinal synthesis has been reviewed by Najjar and Barrett (7), by Fairbanks and Krider (2) and by Kon and Porter (5). No one has reported whether this phenomenon occurs in the young calf or the age at which the synthesis of the vitamins begins to take place. Recently, Illinois workers (3, 4, 11) demonstrated deficiency syndromes for thiamine and riboflavin, but not for nicotinic acid, in the calf when purified diets were fed. The need for riboflavin was confirmed by Warner and Sutton (9, 10).

In view of these facts it seemed desirable to make a study of the levels of certain of the B-vitamins in the digestive tract of calves at various ages; and to compare these levels with those in the feed supplied to the animal. It was hoped that a determination of the age of beginning synthesis would serve as a criterion of the initiation of rumen activity. Furthermore, it appeared desirable to follow the concentration of these vitamins from the feed as ingested through to the material in the lower regions of the intestines.

## EXPERIMENTAL PROCEDURE

Male Holstein calves were procured from various Pennsylvania state institutional herds. They were quartered in individual pens in an artificially heated and ventilated barn and all were fed and managed alike. Each calf received medium grade alfalfa hay *ad lib.* and a calf starter free choice up to a maximum of 5 lb. per calf per day. Each was placed on a milk feeding schedule which permitted him to receive colostrum plus a total of 300 lb. of Holstein herd milk during the first 36-day period after which time milk was removed from the ration.

Twenty-one calves were divided at random into age groups for slaughter, three each being sacrificed at the ages of 2, 4, 6, 8, 10, 12 and 14 wk. Records were kept of birth weights, weight at the time of slaughter, the amounts of feed consumed during the 48 hr. prior to slaughter, the weight, consistency and appearance of the rumen contents, the weight of the rumen tissue and any miscellaneous observations that seemed appropriate.

Received for publication August 21, 1950.

<sup>1</sup> Taken from data presented in a thesis to the graduate faculty of The Pennsylvania State College by E. M. Kesler in partial fulfillment of the requirements for the degree of Doctor of Philosophy.

<sup>2</sup> Authorized for publication as paper no. 1614 in the Journal series of the Pennsylvania Agricultural Experiment Station, on August 9, 1950.

Upon slaughter, representative samples were collected from the contents of the rumen, omasum, abomasum, small intestine and the combined contents of the caecum and large intestine, this latter sample being called simply "large intestine." The samples were acidified, taken to the laboratory and blended into a slurry with a Waring blender. If possible, analysis of the wet material was begun immediately. However, part of the samples could not be assayed at once and these were frozen until time was available for assay.

Thiamine was determined by the thiochrome method (1), using a Farrand photofluorometer, riboflavin by the fluorometric procedure (6) and nicotinic acid by the colorimetric cyanogen bromide-metol determination (8). Dry matter was determined on all samples assayed and the concentration of the three vitamins was calculated on a dry matter basis.

The milk, hay and grain fed were assayed several times during the trial and levels of the three vitamins similarly calculated.

#### RESULTS

The results of thiamine analyses are presented in table 1. The average level of intake for each calf was calculated according to the relative amounts of hay, grain and milk consumed and the assay values of each. Mean levels of intake

TABLE 1

*Concentrations of thiamine in the digestive tract of dairy calves compared to the feed eaten<sup>a</sup>*

Age at slaughter	Av. levels of intake	Thiamin concentrations ( $\mu\text{g/g.}$ of dry matter)				
		Rumen	Omasum	Abomasum	Small intestine	Large intestine
(wk.)						
2	2.7	9.4	b	3.2	4.4	7.4
4	3.9	11.2	b	2.1	4.7	8.6
6	4.8	8.5	13.2	5.6	6.9	5.9
8	5.1	6.4	7.8	5.9	3.7	3.5
10	5.9	7.9	9.8	3.7	6.6	5.8
12	7.3	8.4	8.7	5.1	5.3	3.1
14	6.6	9.1	11.0	6.7	6.7	4.7

<sup>a</sup> Mean of three calves slaughtered in each age group.

<sup>b</sup> Insufficient contents present for analysis.

were compared with the concentrations found in each region of the tract. It was possible to compute this level since the weights of grain, hay and milk which each calf ate during the 48-hr. period prior to slaughter were known. The per cent dry matter for each feedstuff was known so that the dry matter intake of each could be computed, and from this the micrograms of each vitamin consumed could be estimated. Dividing the total dry matter intake in grams into the total vitamin intake in micrograms gave the average intake for each calf. The data presented in table 1 represent the mean of three calves in each age group. No data are presented on omasum samples for the 2- and 4-wk. age groups because in these animals there were insufficient amounts of material in the omasum for adequate analysis. Levels of thiamine were higher generally on a dry matter



basis in all parts of the digestive tract than in the feed which the animals had received. This was true of all ages studied. Concentrations of the vitamin were higher in the rumen and omasum than in the lower regions of the tract.

TABLE 2

*Concentrations of riboflavin in the digestive tract of dairy calves compared to the feed eaten<sup>a</sup>*

Age at slaughter	Av. levels of intake	Riboflavin concentrations ( $\mu\text{g/g.}$ of dry matter)				
		Rumen	Omasum	Abomasum	Small intestine	Large intestine
(wk.)						
2	18.8	34.0	b	20.8	43.9	108.4
4	13.0	22.2	b	13.1	35.7	60.9
6	10.8	19.5	26.2	18.1	46.6	35.3
8	10.7	21.3	33.1	17.6	41.4	21.7
10	8.9	16.3	34.9	15.5	43.1	40.7
12	10.3	17.9	37.1	14.6	44.3	35.9
14	10.6	25.4	34.2	16.7	53.4	24.6

<sup>a</sup> Mean of three calves slaughtered in each age group.

<sup>b</sup> Insufficient contents present for analysis.

Table 2 presents the values for riboflavin. Levels of this vitamin were higher in all parts of the digestive tract than in the feed. This was true of all ages studied. Concentrations in the small intestine were significantly higher than in the other parts studied, except in the 2- and 4-wk. age groups. In these an increase over abomasum values is apparent, with a continued increase in the large intestinal samples.

TABLE 3

*Concentrations of nicotinic acid in the digestive tract of dairy calves compared to the feed eaten<sup>a</sup>*

Age at slaughter	Av. levels of intake	Nicotinic acid concentrations $\mu\text{g/g.}$ of dry matter)				
		Rumen	Omasum	Abomasum	Small intestine	Large intestine
(wk.)						
2	54.5	72.7	b	68.8	86.3	113.8
4	39.0	51.8	b	22.2	104.1	76.4
6	30.5	32.5	49.5	36.9	87.1	62.2
8	29.6	14.2	37.4	24.0	36.6	30.0
10	37.2	45.4	59.7	74.4	119.4	59.3
12	22.9	22.4	32.7	31.6	129.3	41.3
14	23.2	56.5	63.3	56.7	133.4	53.0

<sup>a</sup> Mean of three calves slaughtered in each age group.

<sup>b</sup> Insufficient contents present for analysis.

As seen in table 3 niacin values were, as a rule, higher in the digestive contents than in the feed. Levels in the small intestine again were found to be higher than those in the other regions. This table does show several instances where feed values are higher, pointing out the large variations that occur. Great differences also were noted between individuals of the same age.

As can be seen from these data, no relationship could be established between age of the calf and levels of any of the three vitamins studied. Neither did

there appear to be any relation between levels of B-vitamins and body size, nor between levels and the amounts of material found in the rumen. It is possible that the increase in values which was obtained for the contents of the gastrointestinal tract, particularly the small intestines, might have been due in part to selective absorption of nutrients other than the vitamins studied, with resultant concentrating effect of the vitamins in the unabsorbed material in the tract. The data are not conclusive in this respect.

#### SUMMARY

Chemical assays of thiamine, riboflavin and nicotinic acid were performed on the contents of the digestive tracts of calves slaughtered at various ages from 2 to 14 wk. Levels of all three vitamins generally were higher in all parts of the tract than in the feed the animals had received. Riboflavin and niacin were found in greater concentrations in the small intestine than in other regions of the tract. Thiamine was found in highest amounts in the rumen and omasum. Under the conditions of this experiment, no relationship could be noted between the age of the calf and levels of B-vitamins in the digestive tract.

#### REFERENCES

- (1) ASSOCIATION OF VITAMIN CHEMISTS. *Methods of Vitamin Assay*. Interscience Publisher's, Inc., N. Y. pp. 69-85. 1947.
- (2) FAIRBANKS, B. W., AND KRIDER, J. L. Factors Involved in the Synthesis of Vitamins in the Ruminant. *North Am. Vet.*, 25: 22-25; 97-100. 1944.
- (3) JOHNSON, B. C., WIESE, A. C., MITCHELL, H. H., AND NEVENS, W. B. The Metabolism of Nicotinic Acid and its Role in the Nutrition of the Calf. *J. Biol. Chem.*, 167: 729-736. 1947.
- (4) JOHNSON, B. C., HAMILTON, T. S., NEVENS, W. B., AND BOLEY, L. E. Thiamine Deficiency in the Calf. *J. Nutrition*, 35: 137-145. 1948.
- (5) KON, S. K., AND PORTER, J. W. G. The Role of the Microflora of the Alimentary Tract of Herbivora with Special Reference to Ruminants. 5. Synthesis of Vitamins in Relation to Requirements. *Nutrition Abstracts and Revs.*, 17: 31-37. 1947.
- (6) LOY, H. W., JR. Report on Chemical Methods for Riboflavin. *J. Assn. Official Agr. Chemists*, 30: 392-398. 1947.
- (7) NAJJAR, V. A., AND BARRETT, RACHEL. The Synthesis of B-vitamins by Intestinal Bacteria. *Vitamins and Hormones*, 3: 23-48. 1945.
- (8) STEELE, H. K. Report of the 1944-45 Committee on Niacin (Nicotinic Acid) Assay. *Cereal Chem.*, 22: 448-454. 1945.
- (9) WARNER, R. G., AND SUTTON, T. S. Performance of Calves on a Photolyzed Milk Diet. *J. Dairy Sci.*, 31: 675-676. 1948.
- (10) WARNER, R. G., AND SUTTON, T. S. The Nutrition of the Newborn Dairy Calf. III. The Response to a Photolyzed Milk Diet. *J. Dairy Sci.*, 31: 976-985. 1948.
- (11) WIESE, A. C., JOHNSON, B. C., MITCHELL, H. H., AND NEVENS, W. B. Riboflavin Deficiency in the Dairy Calf. *J. Nutrition*, 33: 263-270. 1947.

## A COMPARISON OF COMFORT AND TIE-CHAIN STALLS<sup>1, 2</sup>

I. D. PORTERFIELD, GEORGE HYATT, JR., D. P. BROWN AND A. D. LONGHOUSE

*West Virginia University, Morgantown*

In northern United States dairy cows spend more time in the barn than on pasture during the year. Therefore, the type of stall that will give the best results is of special importance to the dairyman.

Monroe *et al.* (3) found that milk and butterfat production of Holstein cows, changed at 50-day intervals, were not influenced by the stall.

Ellington and Knott (2) found that the box stall required 21.25 lb. of bedding per day as compared to 4.7 lb. for the stanchion stall and 3.8 lb. for the Hoard's Model Stall and the Modified Hoard Stall.

The object of this study was to determine if there were significant differences in milk production, cleanliness, incidences of mastitis, injuries and in time spent lying down between Holstein cows kept in comfort stalls and those kept in tie chain stalls. The amount of bedding used and time required for cleaning also were studied for each type of stall.

### PROCEDURE

Data for this experiment were collected between October and May in 1947, 1948, 1949 and 1950, in the Holstein herd of the West Virginia Agricultural Experiment Station. All cows in this herd were above average in size and have averaged more than 400 lb. of butterfat per year since 1943.

The so-called comfort stall was a modified Hoard Stall. It was 84 in. long and 49 in. wide, with a crossbar at the rear of the stall, adjustable to the size of the cow as shown in figure 1. When the cow was standing, she was forced by the pipes in the front of the stall to stand with her rear legs over the crossbar so that the droppings and urine were back of the crossbar. When lying down the cow's head would go under the pipes enabling her to lie in front of the crossbar.

The tie-chain stalls were 66 in. long and 42 in. wide as shown in figure 2.

In the fall of 1947, two Holstein cows were placed in comfort stalls and production data were collected. Upon the addition of six more comfort stalls in the fall of 1948, the Holstein herd was divided, and eight cows, representative of the various ages, were placed in the comfort stalls and seven in the tie-chain stalls. In the fall of 1949, the cows that were in comfort stalls the previous year were placed in the tie-chain stalls and those that were in the tie-chain stalls the previous year were placed in comfort stalls. During the experiment, all cows were fed the same amount of roughage similar in quality. Each cow was

Received for publication August 28, 1950.

<sup>1</sup> The data in this paper are from a thesis submitted by the senior author to the Graduate School of West Virginia University in partial fulfillment of the degree of Master of Science, 1950.

<sup>2</sup> Published with the approval of the Director of the West Virginia Agricultural Experiment Station as Scientific paper no. 430.



times per week for injuries for which the stall was considered to be the contributing cause.

Data were obtained on the amount of bedding used in each type of stall for a 7-day period in 1948 and a 15-day period in 1949. Soft wood shavings were used for bedding material. Nine time and motion studies were made on the amount of time necessary to clean the different types of stalls.

Three trials on the cleanliness of the cows in the comfort stalls and those in tie-chain stalls were made. During the trials each cow was scored daily on the following basis:

0—No visible signs of manure or stain; 1—Stained but no manure; 2—Stained and/or manure on hocks or tail; 3—Stained and/or manure on hocks and tail; 4—Stained and/or manure on hocks, tail and one flank; 5—Stained and manure on hocks, tail and both flanks.

The amount of time cows spent lying down in each type of stall for a 24-hr. period was determined by using thermocouples. Each time a cow got up or lay down the device recorded the number of the cow and the time to the nearest five minutes. Data were obtained over a period of 19 days. Recordings that were incomplete due to broken thermocouples or that were abnormal due to the heat period of the cows were omitted.

#### RESULTS

The average weekly milk ( $2 \times$  M.E.) production for eight cows is shown in figure 3. Each cow has one production record made in each type of stall, with the exception of 539. She has two production records in each stall. Her first two records were made in the tie-chain stall and the last two in the comfort stall. In her case, the first record made in the tie-chain stall was compared with the first record in the comfort stall and the second record in the tie-chain stall was compared with the second record in the comfort stall. Seven of the cows, representing eight comparisons, produced between 12.0 and 97.2 lb. more milk per week while in the comfort stall than while in the tie-chain stall. One cow produced 19.9 lb. more milk per week in the tie-chain stall than in the comfort stall.

During the experiment eight injuries were sustained in the comfort stall, as compared to 26 in the tie-chain stall. Five flank injuries were sustained in the tie-chain stalls. All other injuries were either bruised hocks or skinned knees.

The difference in the amount of bedding required for the tie-chain and comfort stalls for 7- and 14-day periods was analyzed by the analysis of variance and found to be not significant.

Time and motion studies were made on the amount of time required to clean the tie-chain and comfort stalls. The comfort stall required 6.48 sec. longer; however, this difference was not statistically significant.

The total score for cleanliness in each trial is shown in table 1. In each trial the cows in comfort stalls remained cleaner than the cows in the tie-chain stalls. The first trial was conducted shortly after the cows were moved into their

TABLE 1  
*Total score for cleanliness of cows in comfort and tie-chain stalls*

Trial	Length of trial (d.)	Comfort stall		Tie-chain stall		Score difference
		Total score	Cows used	Total score	Cows used	
1	8	65.5	7	130.5	7	65.0
2	8	37.5	7	66.5	7	29.0
3	12	37.5	8	109.5	7	72.0

stalls from pasture and they had not become properly adjusted to the stall; this probably accounts for the poorer cleanliness scores during this trial.

During a 19-day period, cows in comfort stalls spent an average of 10.2 hr. per day lying down, while the cows in tie-chain stalls spent an average of 8.8 hr. per day lying down. The difference was found to be highly significant.

#### DISCUSSION

The fact that the cows in the comfort stalls lay down 1.4 hr. per day more than those in tie stalls indicates that they were more comfortable. This more nearly approaches the 13 hr. per day spent lying down while on pasture, as reported by Atkeson *et al.* (1). This may have been one of the important reasons why the cows in comfort stalls produced more milk.

The comfort stalls were wider than the tie stalls, which eliminated flank injuries caused by the pipe partitions of the stalls. The comfort stalls were longer than the tie stalls and had a crossbar in the rear which helped retain

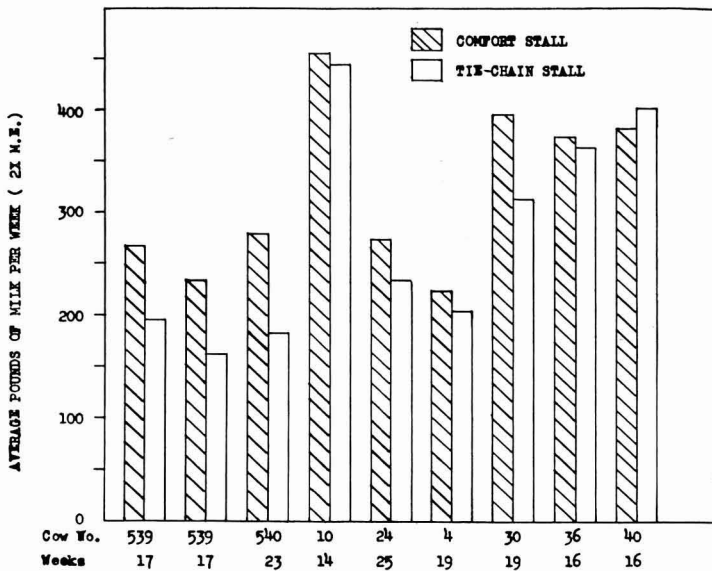


FIG. 3. A comparison of the average weekly milk production of cows kept in comfort and tie-chain stalls.

the bedding beneath the cow, thus keeping them cleaner. A combination of added length and more bedding beneath the rear and front legs prevented hock and knee injuries.

Six comfort stalls occupied the same space in width as seven tie-chains. This means that fewer cows can be kept in a given space. The comfort stall is 18 in. longer than the tie-chain. When two rows of comfort stalls are installed the additional 18 in. requirement for length per row will require a barn 3 ft. wider than the conventional barn. If additional comparisons on production continue to show an increase, the designers of dairy barns may need to consider a change in barn designs.

Most manufacturers of barn equipment sell more stanchion-type stalls than either the tie-chain or comfort. A criticism of this experiment is that the comparison should have been made between the stanchion and the comfort stall. Tie-chain stalls were being used at the farm several years prior to the beginning of this experiment. Since the tie-chain offers the cow more freedom of movement than does the stanchion, it would be expected that the difference in the performance of cows would be even greater if stanchion stalls had been used.

#### SUMMARY

A comparison of Holstein cows kept in comfort and tie-chain stalls was made. Based on nine comparisons, eight produced more milk while in comfort stalls. This increase amounted to 12 to 97.2 lb. of milk per week.

Those kept in comfort stalls sustained fewer injuries, remained cleaner and spent significantly more time lying down.

The difference in the amount of bedding used and cleaning time for each type stall was not statistically significant.

#### REFERENCES

- (1) ATKESON, F. W., SHAW, A. O., AND CAVE, H. W. Grazing Habits of Dairy Cattle. *J. Dairy Sci.*, 25, 9: 779-784. 1942.
- (2) ELLINGTON, E. V., AND KNOTT, J. C. Methods of Stabling Dairy Cows. *Washington Agr. Exp. Sta. Bull.*, 208: 19-20. 1926.
- (3) MONROE, C. F., KRAUSS, W. E., AND HAYDEN, C. C. Box Stalls Versus Tie and Stanchion Stalls for Milk-producing Cows. *Ohio Agr. Exp. Sta. Bimonthly Bull.* 20, 177: 196-200. Nov.-Dec., 1935.

# TYPE DIFFERENCES AND BLOOD ANTIGENS IN A GUERNSEY HERD<sup>1</sup>

A. A. DUNLOP<sup>2</sup>

## INTRODUCTION

This study deals primarily with type differences scored in a Guernsey herd in which a moderate amount of close breeding had been practiced. The questions asked were: (a) How far did the inbreeding make distinctly separate inbred lines? (b) What was the effect of inbreeding on body form as measured by various type scores? (c) Was type correlated with the presence or absence of any of the blood antigens?

## MATERIAL

*The herd.* The Guernsey herd at Blakeford Farms, Queenstown, Md., was founded in the middle thirties. The present data pertain to foundation animals born from 1929 to 1934 and to animals born from 1937 to the early part of 1945. Two foundation bulls designated as sires 1 and 3 were 12.5 per cent related.<sup>3</sup> Both these sires were used extensively, while sire 4, a son of sire 3 out of a daughter of sire 1, was used to a moderate degree. Several other bulls, sons of sires 1 and 2, were used to a slight extent in the later years.

While the foundation bulls were slightly related to some of the foundation cows, no intensive inbreeding took place until these bulls were mated to their own daughters. At the same time, a considerable number of matings were made of one sire to the daughters of another, so that animals with much and with little or no inbreeding existed contemporarily in the herd.

*Type scores.* Scores for type were given jointly by an official of the Guernsey Cattle Club and the manager of the property, who also was a widely experienced judge of dairy cattle. Thirteen anatomical features were scored. These were neck, shoulders, back, loin, rump, tailhead, barrel, ribs, skin, legs, fore udder, rear udder and teats. For convenience, fore udder, rear udder and teats are termed udder characters, while the remainder are referred to as body characters. Each character was given a numerical score with possible values from one to six, one being the highest score and six the lowest. The scoring system thus had the same number of grades as the official type classification of the breed. Scoring was done at any convenient time coinciding with the visit of the breed official,

Received for publication Sept. 25, 1950.

<sup>1</sup> Journal paper J-1818 from the Iowa Agricultural Experiment Station, Ames. Project 1053.

<sup>2</sup> Now Research Officer, Commonwealth Scientific and Industrial Research Organization, Sydney, N.S.W., Australia.

<sup>3</sup> All relationship and inbreeding coefficients were calculated from pedigrees traced to ancestors born in 1927 or earlier. With the generation interval being about 5 yr. in dairy cattle, this makes the coefficients relative to the Guernsey breed, as of about 1925, so far as this herd's ancestors which were born in 1927 or just before can be considered typical of the breed at that time.



so that scores were not all made at the same age, animals being scored from ages of a few months upwards. Some animals were scored on several occasions, but in this study only one set of scores was used for any one animal. The set used was chosen as far as possible to have the animal in the first half of one of its earlier lactations when scored. This was done with a view to minimizing the effects which variations in age and in nearness to calving might have had on udder scores. Some over-all type scores, presumably similar to official type classifications, as well as some production data, were recorded but the numbers were too small to promise definite answers to the questions which seemed interesting.

*Antigen determinations.* The antigenic types of the Blakeford animals were determined at the University of Wisconsin prior to the present study.<sup>4</sup> Blood samples were sent for testing on five occasions from 1942 to 1945. The data studied here were taken from the Blakeford records.

#### METHODS OF ANALYSIS

*Family structure.* Attempts were made to discover how much separation into distinct families or inbred lines had resulted from the mating system followed. These were made in the three following ways: (a) A complete pedigree map of the herd of over 200 animals was drawn and twice redrawn in trying to sort the network of relationships into fairly separate groups of animals with little relationship between the groups. Such a pedigree situation may be visualized as being like several parallel strips of wire netting with only a few wires connecting any two strips. (b) The pedigrees of the descendants of sires 1, 2 and 3 were mapped, each sire's descendants being placed on a separate map. If a sire's descendants constituted a rather homogeneous group genetically, each group should have had few recent ancestors in common with any other group. (c) Ten pedigree maps were drawn, each for a foundation cow and her descendants, to see whether any distinct pedigree groups could be recognized on that basis.

Groups of the latter type frequently are called "cow families" (Bartlett and Margolin, 1944) in a general sense. The term has several disadvantages. First, it emphasizes the female side of the pedigree to the exclusion of the male side. Second, the reproductive rate of cows is so low that cow families are either very small or the members are only distantly related to the foundation cow. Unless a cow's descendants are bred to each other, the sampling and halving nature of inheritance makes the genes from the foundation cow, which are common to all the animals in the group, only a small fraction of the total genes present in each animal. The number of living descendants a cow has after several generations usually is more dependent on chance in determining sex of the calves than on

<sup>4</sup> For information about these antigens see: Ferguson (1941), Ferguson *et al.* (1942), Irwin (1947 and 1949), Stormont (1950), Stormont and Cumley (1943) and Stormont *et al.* (1951).

the genes which she possesses or on the breeder's attempts to form a family around her.

Following these procedures, inbreeding coefficients were calculated for all the animals in the herd.

*Outline of methods.* The first step in the statistical analysis of the scores was to estimate the effects of date of scoring, of age at classification and of inbreeding, so that the scores could be corrected for differences in these factors. Date and age were treated as discrete classifications and inbreeding as a continuous variable. Because of unequal sub-class numbers, the method of least squares was used to obtain these corrections, as well as to estimate the differences between sires. An analysis of variance was carried out for each characteristic scored, and components of variance for the sire classification and for error were computed. These components were used to estimate heritability for each characteristic. Those characteristics with apparently high heritability then were chosen for further study as most likely to reveal any existing correlations with antigens. The original scores for these selected characters were adjusted for differences in date of scoring, in age at scoring and in inbreeding. With these corrected scores, heritability then was estimated by the intra-sire regression of daughter on dam, as a nearly independent and more reliable check on the earlier estimates of heritability. Association between the most highly heritable characters and specific antigens was tested by analysing the variance of adjusted scores within and between the two classes: antigen present and antigen absent.

*Estimation of environmental and genetic effects.* (a) Non-orthogonality: Animal data, particularly where they do not come from designed experiments, tend to be unbalanced or non-orthogonal, *i.e.*, the numbers in the sub-classes in any multiple classification are unequal. As a result, the border means alone do not provide unbiased estimates of the effects on which the classification has been made. For instance, it might be that the mean score of the offspring of a certain sire as compared with another was made higher than it should be, merely because one sire had a higher fraction of his offspring scored at an age when scores tend to be above average and so on. This disproportionate frequency can cause partial confounding of two or more variables the effects of which are to be estimated. Under these conditions the usual method of computing the analysis of variance is not wholly valid. A least squares procedure suitable for obtaining unbiased estimates of effects and tests of significance has been described by Yates (1933, 1934) for the case of a two-way classification, while Hazel (1946) has extended the procedure. This method, with various aids, was outlined in considerable detail by Henderson (1948). Henderson's procedures for obtaining estimates of variance components from sums of squares computed in such analyses were followed in this study, as the data were distinctly non-orthogonal with some sub-classes empty. This was true even after discarding a small amount of data where the extra computation required to handle additional classes seemed likely to be excessive in proportion to the expected increase in information. Thus, only nine dates of classification were considered, although

a few animals were classified on other occasions. Similarly, too few male animals were scored to make a classification for sex profitable, while the only females considered were the daughters of sires 1, 2, 3 and 4 mentioned previously, although several additional bulls each had a few daughters. This method of partially balancing the data by discarding some of the least numerous classes could not be carried far. When it was completed, only 76 animals with all the body scores and 73 of these 76 with all the udder scores finally were available for analysis.

(b) Mathematical model: A preliminary analysis of the fore udder scores had shown the effect of age on these scores to be curvilinear and that the nearness to calving did not influence the scores appreciably. It also had shown that including linear and higher order regression coefficients would increase considerably the already heavy labor of computation. Consequently, nearness to calving was not considered further and the effect of age was split into an additional classification. The model selected as being most suitable to the data was:

$$Y_{ijkl} = \mu + a_i + s_j + c_k + b(X_{ijkl} - X) + e_{ijkl}$$

Here  $Y_{ijkl}$  is the score of the  $l$ th individual in the  $k$ th  $c$  class, in the  $j$ th  $s$  class and in the  $i$ th  $a$  class.

The constant  $\mu$  is the average of the parent population from which these data are considered to be a sample. The breadth of the conclusions which may be drawn from the study are dependent on what one knows or assumes about the parent population.

The  $a_i$  are the effects of the dates when the animals were scored. The  $a_i$  might have real values because of changes in the classifiers' standards, or environmental conditions may have made the animals appear better at some dates than at others, or the genetic mean of the herd may have changed with time.

The  $c_k$  are the effects of the differences in age when the animals were scored. The age groupings were:

Constant	Age
$c_1$	0 to 1 yr.
$c_2$	1 to 2 yr.
$c_3$	2 to 3 yr.
$c_4$	3 to 4 yr.
$c_5$	4 yr. and over

The constant  $b$  is the average intra-subclass regression of type score on one per cent of inbreeding.  $X_{ijkl}$  is the per cent inbreeding of the individual which received the score,  $Y_{ijkl}$ . Each subclass contained animals by the same sire, in the same age group, and classified at the same time.

The  $s_j$  are the effects of the different sires on the scores of their daughters. These are expressed as deviations from  $\mu$ . The variance,  $\sigma_s^2$ , of the  $s_j$  would be

<sup>5</sup> This is the variance due to the average effects of genes in that population. "Genic" as used here means additively genetic. It does not include the genetic variance due to dominance deviations or to epistatic deviations.

equal to  $\frac{G(1-F)}{4}$ , where  $G$  is the genic variance<sup>5</sup> for the character under study and  $F$  is the average inbreeding coefficient of the population, if mating within this herd was substantially random. If the inbreeding in the herd had been so conducted as to tend to form separate inbred lines around each sire, then  $\sigma_s^2$  would be more than  $\frac{G(1-F)}{4}$ , since  $\sigma_s^2$  would then contain some variation from the mates of each sire being more closely related to him than they were to the other sires. This was slight in the present case and in any case would not have changed the *order* of heritabilities of the different characters.

Each  $e_{ijkl}$  is the discrepancy between the actual observation,  $Y_{ijkl}$ , with which it is associated and what the observation is expected to be because of its  $a, s, c$  and  $X$  characteristics. The  $e_{ijkl}$  may be due to Mendelian sampling variations, to dominance, to epistasis, to inexactness of the model in accounting for the various factors considered in it, or to any environmental factors not included in the model. Their variance is  $\sigma_e^2$ . The remaining  $\frac{3(1-F)}{4}$  of  $G$ , where  $G$  and  $F$  are as defined earlier, is contained in  $\sigma_e^2$ .

(c) The analysis of variance: Let  $R(\dots)$  be defined as the reduction in sum of squares due to fitting the parameters listed within the brackets. Let  $\Sigma Y^2$  be the sum of the squares of the uncorrected scores. In the present situation, where 76 scores were used, the analyses of variance took the form of table 1.

TABLE 1  
*The analysis of variance*

Source of variation	d.f.	Sum of squares
Total	75	$\Sigma Y^2 - \bar{Y}(\Sigma Y)$
$a, s, c,$ and $b$ jointly	16	$R(\mu, a, s, c, b) - \bar{Y}(\Sigma Y)$
$a$	8	$R(\mu, a, s, c, b) - R(\mu, s, c, b)$
$s$	3	$R(\mu, a, s, c, b) - R(\mu, a, c, b)$
$c$	4	$R(\mu, a, s, c, b) - R(\mu, a, s, b)$
$b$	1	$R(\mu, a, s, c, b) - R(\mu, a, s, c)$
Error	59	$\Sigma Y^2 - R(\mu, a, s, c, b)$

*Estimates of heritability.* (a) Least squares estimates of components of variance: Henderson has set out general rules which enable one to write down most of the expectations of the pertinent reductions of table 1 in terms of variance components and their coefficients. Here  $a, c$  and  $b$  may be considered as correction factors for variables which are not pertinent to the main problem, but have some effects. The interesting components to be estimated here are  $\sigma_s^2$  and  $\sigma_e^2$ . The latter is obtained directly as the mean square for error.

(b) The correlation between paternal half sibs: Since  $\sigma_s^2 = \frac{G(1-F)}{4}$  while  $\sigma_e^2 = E + \frac{3G(1-F)}{4}$ , where  $E$  is most of the non-additive genetic variance plus

the environmental variance not accounted for by the model, the estimate of heritability is:

$$\frac{G}{G+E} = \frac{\frac{4}{1-F} \sigma_s^2}{\frac{4}{1-F} \sigma_s^2 + \sigma_e^2 - 3\sigma_s^2}$$

Estimates of heritability for each of the 13 characters scored were made in this way.

(c) The intra-sire regression of daughter on dam: At this stage three characters which appeared to be most highly heritable on the basis of the half-sib estimates were selected for further study as being least likely to have their correlations, if any, with antigens obscured by the effects of non-additive genetic variations and of environmental variations on the scores. Before the scores in these three characters were studied further, they were corrected for the effects of age and date and of variations in inbreeding as estimated in the least squares analyses.

As the data contained many daughter-dam pairs, the corrected scores offered the opportunity to make additional estimates of heritability by doubling the intra-sire regression of daughter on dam. This gives an estimate of the genic fraction of the variance within the group of dams which are mated to a sire (Lush, 1948).

The number of daughter-dam pairs was, of course, smaller than the number of offspring used in making the half-sib estimates, since not every animal had a dam with a corrected score. Yet the estimates of heritability obtained by the daughter-dam method are distinctly more reliable, since the half-sib resemblances reflect genetic differences between only four sires. Of the three selected characters for which estimates were made, two had 24 daughter-dam pairs while the third had 28 pairs scattered among the four sires. There were three sets of full sibs for each character. These bias the estimates slightly upward by reducing the apparent variance between dams within sires in the denominator of the regression coefficient.

*Tests for relationship between antigens and type scores:* Whether an antigen was associated with a score was tested by analyzing the variance of the adjusted scores within and between the two groups,—the group which had the antigen in question and the one which lacked it. Tests were made only for the 11 antigens for which there were at least 23 animals in the least frequent of the two classes. These are listed in table 4. The other antigens were omitted because the information to be gained about an all-or-none character decreases as the frequency of the character approaches zero or one.

If the antigens had any joint or epistatic effects in certain combinations but not in others, that would have been indicated in part in the present analysis unless the opposite combination had similar effects; *i.e.* unless the *BC* and *bc* effects were like each other and opposite to the *Bc* and *bC* effects in sign, and the frequencies of the four types were such that these effects cancelled each other com-

pletely in the separate comparisons of  $B$  and  $b$  and of  $C$  and  $c$ . That contingency seems highly unlikely, although not wholly impossible. In any event, the small numbers made it seem unprofitable to test for such interactions of two or more antigens.

#### RESULTS AND DISCUSSION

*Family formation.* The three complete pedigree maps of the herd appeared as extremely tangled networks without clear separation into distinctly different lines. These maps were, nevertheless, useful in calculating inbreeding coefficients.

When the pedigrees of the descendants of the three most widely used bulls were drawn separately, each such map contained a large proportion of the animals in the complete map. Each map included many descendants of the other important bulls, many animals being in all three such maps. The bulls had not been used mainly as centers around which to form distinctly separate lines.

The pedigrees of the descendants of any single foundation cow usually contained two, and often all three, of the main bulls, any incipient family formation having been checked by mating to comparatively unrelated stock. In two cases where family formation had not been halted by mating to unrelated stock, the families were almost without representatives in the final year covered by the data.

This lack of differentiation into distinctly separate lines made it appear fruitless to study further any differences between inbred lines or any methods of evaluating and using such lines. Inbreeding was present, but not according to a plan which would have made inbred lines so separate from each other that marked differences between the lines could be expected. As soon as this was seen clearly, major attention was focussed on other aspects of the data. The herd was regarded thereafter as a single moderately inbred population, while intensity of inbreeding was considered as a factor which might have influenced the type scores of individuals.

This part of the analysis emphasized that if inbred but distinctly unrelated lines are wanted, the mating system should be planned primarily and especially to produce them. The foundation stock should be grouped into as many different lines as are wanted in such a way as to minimize the relationship between groups. Then each line should be mated within itself and kept isolated genetically thereafter, except when outcrosses are wanted for testing purposes and subsequent sale. The reproductive rate of cattle is such that the smallest practicable group of animals on which to found an inbred line would be a group of female half-sibs mated to their sire or brother.

(b) *Sire differences and inbreeding effects.* The solution of the groups of equations concerning each of the thirteen scores gave rise in each case to estimates of the effects of sire, dates, ages and of inbreeding; that is, estimates of the  $s_j$ , the  $a_i$ , the  $c_k$  and of  $b$ . The  $a_i$  and the  $c_k$  are only correction factors to adjust the primary data so as to remove the differences between observation dates and between ages, thus making more sensitive the tests for the other factors. In

consequence, they are not presented here. The sizes of the  $s_j$  gave some preliminary idea of the relative heritabilities of the different characters, as well as of the general differences among the four bulls in merit in all the characters studied. While the various values of  $b$  were obtained mainly for correction purposes, they do show the size of the inbreeding effect on these characters. Together with the  $s_j$ , they are presented in table 2.

TABLE 2  
*The parameter estimates*

Character \ Parameter	$b$ †	$s_1$	$s_2$	$s_3$	$s_4$
Shoulders .....	0	0.22	-0.32	0.19	-0.08
Neck .....	-0	0.04	0.15	-0.31	0.13
Legs .....	0.02	0.16	-0.12	0.04	-0.08
Barrel .....	0.03**	-0.15	-0.25	0.35	0.05
Tailhead .....	-0.01	-0.03	-0.10	0.38	-0.25
Rump .....	0.01	0.24	-0.07	-0.30	0.12
Loin .....	0.02	-0.15	0.14	-0.03	0.03
Back .....	0.01	0.31	-0.32	0.22	-0.21
Skin .....	-0.01	-0.09	0.18	-0.25	0.16
Ribs .....	0.01	0.07	-0.39	0.54	-0.22
Fore udder .....	0	0.17	-0.60	0.40	0.03
Rear udder .....	0.03*	0.17	0.11	-0.40	0.13
Teats .....	-0	0.37	-0.45	-0.26	0.34

† Where the original estimates of  $b$  were negative, but so small that reduction in decimal places has made them zero, this is indicated by a minus sign preceding the cipher.

The regression coefficients of score per 1 per cent of inbreeding range from -0.01 to 0.03. Nine are positive, indicating a decline in merit with inbreeding, while four are negative. Only the two largest positive coefficients, for barrel and rear udder, are statistically significant. Even this significance may be questioned, as it would not be surprising if two out of thirteen happened by chance to appear significant. The excess of positive over negative signs conforms to the usual decline in merit following inbreeding, but certainly the results of inbreeding are not catastrophically bad or else the regressions would have had larger positive values. Yet the average inbreeding levels reached in this material were only 11.2 per cent for the three udder characteristics and 11.4 per cent for the body characteristics; hence there is no evidence here of what would happen under really intense inbreeding.

The sire constants are the average scores of the daughters expressed as deviations from  $\mu$ . The relatively large differences between sires in ribs, fore udder and teats indicate that these characters were more highly heritable than the others studied. The preponderance of large negative values in the estimates of  $s_2$  indicate that the daughters of the widely used sire 2 were superior in most of the characters examined.

The various reductions in sums of squares in the analyses similar to table 1 are too lengthy to reproduce. The main features of interest were the differences between sires. In the case of the rib scores the differences were significant at

the 0.01 level, for teat scores significance was well above the 0.05 level, while in the fore udder scores significance approached the 0.05 level.

(c) *The estimates of heritability.* The estimates of heritability by the half-sib correlation method are based on  $\sigma_s^2$  and  $\sigma_e^2$  and are shown in table 3.

TABLE 3  
*The variance components and heritabilities*

Character	$\sigma_s^2$	$\sigma_e^2$	$\frac{G}{G+E}$
Shoulders .....	0.86	-0.01	-0.04
Neck .....	0.31	0	0.02
Legs .....	0.84	-0.05	-0.28
Barrel .....	0.67	0.02	0.11
Tailhead .....	0.91	-0	-0.01
Rump .....	0.79	-0.01	-0.03
Loin .....	0.61	-0.03	-0.26
Back .....	0.98	0.02	0.08
Skin .....	0.56	-0	-0.02
Ribs .....	0.43	0.11	0.85
Fore udder .....	0.74	0.09	0.45
Rear udder .....	0.72	0.02	0.10
Teats .....	0.53	0.12	0.79

These estimates have very large sampling errors, as is to be expected when so small a number of sires is involved. Presumably, the negative figures are due to such sampling errors. The high estimates for the fore udder and teat scores seem to be a reasonable result. The writer, at least, has a clearer mental picture of what would be considered high scoring phenotypes in these characters than for most of the body features scored. Observation leads him to believe that the shape of the teats is a relatively permanent feature of an animal and that the shape of the fore udder is less affected by the passage of time than is that of the rear udder. Although it is not known precisely what features are considered in assigning rib scores, a heritability of 0.85 is unexpectedly high.

By doubling the intra-sire regression of daughter on dam, using the corrected scores of the daughter-dam pairs, the following estimates of heritability in these three characters were obtained:

rib score:	0.50
fore udder score:	0.41
teat score:	0.34

Each of these estimates, although large, is smaller than the corresponding estimates in table 3. This was to be expected as the three characters were chosen for further study because the heritabilities in table 3 seemed high. These additional and independent estimates, based on daughter dam resemblances, reinforce the belief that the characters chosen for further study are in fact relatively highly heritable and therefore suited to the tests which follow.

(d) *Tests for association between scores and the presence of Antigens.* The  $F$  values from the analyses of variance are shown in table 4, along with the mean



TABLE 4  
*Mean adjusted scores for animals with and without each antigen*

Antigen	Ribs			Fore udder			Teats		
	With	Without	F	With	Without	F	With	Without	F
B	2.36	2.46	0.37	2.75	2.82	0.09	3.00	2.84	0.78
C	2.35	2.45	0.38	2.80	2.75	0.05	3.09	2.78	3.25
H	2.21	2.49	3.14	2.82	2.73	0.17	3.11	2.83	2.23
I	2.52	2.33	1.28	3.05	2.64	3.99*	3.06	2.88	0.98
K	2.36	2.44	0.26	2.76	2.81	0.06	2.98	2.88	0.34
O <sub>2</sub>	2.46	2.28	1.26	2.91	2.54	3.25	3.05	2.74	2.98
Y <sup>a</sup>	2.46	2.34	0.62	2.99	2.60	3.97	3.08	2.82	2.23
Z	2.35	2.45	0.40	2.63	2.94	2.62	2.83	3.06	1.73
A'	2.52	2.31	1.88	3.00	2.61	3.79	3.13	2.79	3.93
C' <sup>b</sup>	2.46	2.34	0.62	2.99	2.60	3.97	3.08	2.82	2.23
I'	2.46	2.31	0.89	2.97	2.60	3.23	3.09	2.81	2.52

<sup>a</sup> Designation recently changed to Y<sub>2</sub>.

<sup>b</sup> Designation recently changed to Y<sub>1</sub>, as it was found allelic to the original Y.

corrected scores. The degrees of freedom within groups varied between 66 and 74.

In 24 of the 33 analyses the mean score of the animals having the antigen was larger than that of the group not having the antigen. If this is not a chance deviation, no rational explanation is apparent.

In only one case, that of fore udder and antigen *I*, was the 0.05 level of significance reached. At least one significant case in 20 is to be expected just as a sampling deviation. As 33 tests were made, the significance of this one is questionable. However, seven other *F* values, namely those from the tests of:

rib score × antigen *H*,  
 fore udder score × antigens *O*<sub>2</sub>, *Y*, *A'* and *C'*, and teat  
 score × antigens *C* and *A'*, were moderately close to  
 the 0.05 level.

This seems a little more than is expected from the chance alone, especially for the fore udder and teat scores. Under the null hypothesis, the expected value of *F* in each case is unity. The means of the *F* values for the three characters were:

<i>Character</i>	<i>F</i>
ribs	1.01
fore udder	2.29
teats	2.11
all	1.80

While further confirmation of these results is desirable, they suggest that fore udder and teat scores, at least, are affected by the presence or absence of some of the antigens. Yet the observed relationships might have arisen incidentally because all the animals were daughters of the four principal herd sires. These three characters were chosen explicitly because the progeny groups by these sires had already been found to differ widely for them. Any antigens in which these four sires happened also to differ would thus seem to be associated with those

characters, the association being due to coincidence in four sires, rather than to a general tendency for association in all the individual cows.

If we accept as real these relationships between the presence or absence of certain antigens and certain type scores, the question arises as to what genetic or other circumstances could have brought about such a correlation. Linkage can give rise to a correlation only when the coupling and the repulsion double heterozygotes are not equally frequent. This is unlikely except when the population has only recently emerged from a cross between two groups differing widely in the frequency of the two genes. That was not the case here. All the published reports indicate that the control of an antigen by genes is unaffected by environmental variations; hence, there could be no environmental contribution to the correlation between the antigen and the score. A correlation might reasonably be due to pleiotropic action of the genes controlling the antigen. While such pleiotropy can be a basis for genetic correlations between continuous characters in domestic animals, specific instances of such pleiotropy between the discrete effects of a gene and its effects on a continuous character have been confined largely to more closely studied organisms (Castle, 1941). Although the immunologists have been on the alert for such relations, none seem yet to have been found. The most complete published report of a search for multiple effects of genes controlling blood antigens appears to have been that made incidentally by Kloepper (1946) in a linkage study in man. His results were negative.

While no non-allelic interactions of the genes controlling blood antigens in cattle are known, the antigen is dominant over its complete absence in most cases. Among the 11 antigens studied in table 4, *Z* is the only one with dominance incomplete enough for the heterozygote to be identified (Stormont, 1948). On the other hand, each score is influenced by a considerable amount of non-genic variation. These influences may well include dominance and epistasis as well as environmental effects. For these several reasons, a high correlation between an antigen and a body character is rarely to be expected.

The work reported here seems unlikely to have any direct application, as few of the type scores considered would normally be of enough economic importance to receive much attention in a selection program. The additional progress to be expected in improving a continuous character, by using information about the presence or absence of a correlated antigen, could be estimated if the heritability of both these characters and the genetic and environmental correlations between them were known. This situation could not be examined here. As most economic characters are less heritable than the three characters studied, the usefulness of any single antigen would almost certainly be small. Conversely, however, it is for just such characters that additional aids to selection are most useful.

Utilizing antigenic information would increase progress only as it increased the correlation between real transmitting ability and the index or other composite of information used in selection. Some of any such information which the antigens might furnish might be utilized indirectly by decreasing the necessary

amount of other information, such as production records, and thereby shortening the generation interval. If such information permitted more of the selection to be done at younger ages, this would allow more adult animals to be carried and thus might increase economic returns, although the rate of genetic progress would not necessarily be increased. If the antigen-production relationship were close, antigens could become a valuable aid in selecting between young bulls, since it is the young bulls who offer the most opportunity and freedom to select and about whose transmitting ability the least is known. This, of course, must wait till the antigen-production relationships, if any, are clearly understood.

If the effects of antigens on economic characters of dairy cattle are of the same order as those found here, the additional genetic progress accruing from considering antigens in selection, will almost certainly be small.

#### SUMMARY

The original data of this study were the records of a Guernsey herd over a period of 9 yr., during which an inbreeding program had been followed. The pedigrees were examined as complete herd pedigree maps, as pedigree maps of the descendants of the main sires and as pedigree maps of cow families. Only slight and irregular differentiation into separate lines had taken place.

Many of the animals had been scored for the following thirteen characters: shoulders, neck, legs, barrel, tailhead, rump, loin, back, skin, ribs, fore udder, rear udder and teats. The presence or absence of some forty blood antigens had been recorded. The main object of this investigation was to determine whether any of the scores for type were influenced by the presence of the various antigens.

The regression coefficients of score on inbreeding hint at slightly decreasing desirability of the character with increasing inbreeding. The half-sib correlations indicated heritability to be high for ribs, fore udder and teats. The scores in these three characters were adjusted by least squares analyses to remove the effects of variations in time of classification, in age and in inbreeding. Using these corrected scores, the intrasire regressions of daughters on dam indicated the heritability of these three characters to be between 0.34 and 0.50.

The eleven antigens which had zygotic frequencies nearest to 0.5 were selected as being most likely to demonstrate their effects, if any, on the three selected type scores. In only one of 33 analyses did the difference between the mean of the scores of those animals possessing the antigen and the mean of those not possessing it reach the 0.05 level of significance, although seven other differences approached this level. The average  $F$  value in these analyses was 1.80. This mean  $F$  value may indicate that some of the antigens do have real, though small, effects on some of the type scores, although other interpretations are not excluded. The only tenable genetic explanation of such joint effects seems to be pleiotropy of the genes controlling the antigens. Where genes at many loci control the expression of a continuous character, the correlation between the presence of an antigen, controlled by genes at only one of those loci, and the continuous character is unlikely to be large.

## ACKNOWLEDGMENTS

The writer is indebted to G. M. Moffett, the owner, and to G. Harmon, the manager of Blakeford Farms at Queenstown, Md., for making available the data on which this study was based. Thanks also are due to M. R. Irwin for advice as to changes in antigen nomenclature. The writer also is indebted to L. N. Hazel and J. L. Lush for helpful counsel and criticism in all aspects of this investigation.

## REFERENCES

- (1) BARTLETT, J. W., AND MARGOLIN, S. A Comparison of Inbreeding and Outbreeding in Holstein-Friesian Cattle. New Jersey Agr. Expt. Sta. Bull. 712. 1944.
- (2) CASTLE, W. E. Size Inheritance. Am. Nat., 75: 488-498. 1941.
- (3) FERGUSON, L. C. Heritable Antigens in the Erythrocytes of Cattle. J. Immunol., 40: 213-242. 1941.
- (4) FERGUSON, L. C., STORMONT, C., AND IRWIN, M. R. On Additional Antigens in the Erythrocytes of Cattle. J. Immunol., 44: 147-164. 1942.
- (5) HAZEL, L. N. The Covariance Analysis of Multiple Classification Tables with Unequal Sub-class Numbers. Biometrics Bull., 2: 21-25. 1946.
- (6) HENDERSON, C. R. Estimation of General, Specific and Maternal Combining Abilities in Crosses among Inbred Lines of Swine. Unpublished Ph.D. thesis, Iowa State College Library, Ames. 1948.
- (7) IRWIN, M. R. Immunogenetics. Advances in Genetics, 1: 133-159. 1947.
- (8) IRWIN, M. R. Immunological Studies in Embryology and Genetics. Quarterly Review of Biology, 24: 109-123. 1949.
- (9) KLOEPPER, H. W. An Investigation of 171 Possible Linkage Relationships in Man. Ann. Eug., 13: 35-71. 1946.
- (10) LUSH, J. L. *The Genetics of Populations*. Ames, Iowa. 1948.
- (11) STORMONT, C. Differentiation of Heterozygotes from Homozygotes in Two Cellular Characters in Cattle. (Abs.) Genetics, 33: 125. 1948.
- (12) STORMONT, G. Additional Gene-controlled Factors in the Bovine Erythrocyte. Genetics, 35: 76-94. 1950.
- (13) STORMONT, C., AND CUMLEY, R. W. Cellular Antigens in Cattle Blood. J. Heredity, 34: 34-41. 1943.
- (14) STORMONT, C., OWEN, R. D., AND IRWIN, M. R. The B and C Systems of Bovine Blood Groups. Genetics (In Press). 1951.
- (15) YATES, F. The Principles of Orthogonality and Confounding in Replicated Experiments. J. Agr. Sci., 23: 108-145. 1933.
- (16) YATES, F. The Analysis of Multiple Classifications With Unequal Numbers in the Different Classes. J. Am. Stat. Assn., 29: 51-66. 1934.

# JOURNAL OF DAIRY SCIENCE

## ABSTRACTS OF LITERATURE

Prepared in cooperation with the  
International Association of Ice Cream Manufacturers  
and the Milk Industry Foundation

### BOOK REVIEWS

**59. Byproducts from milk.** EARLE O. WHITTIER and BYRON H. WEBB. Reinhold Publishing Corp., New York, N. Y. 317 pp. \$6.00. 1950.

This textbook is entirely new and collects in one volume various methods of dairy byproduct utilization from federal and state bulletins, patents and technical journals.

The physical and chemical properties of milk, skimmilk, whey and buttermilk and their components are described in detail, with methods given for converting these dairy products and by-products into both food and non-food products.

In the 11 chapters, detailed processing directions are given for condensed and dried milks, ice cream mix, fermented milks, whey cheeses, condensed and dried whey, bread, candy, milk sugar, casein, etc.

Theoretical discussions necessary for understanding the principles on which these manufacturing procedures are based have been included. Also, basic information is included on the chemistry and nutritional value of the components of milk and on behavior of milk byproducts during heating and other processing treatments.

An excellent bibliography of references to original articles is included, which will be useful to research workers and others who wish to refer to the original articles.

The chapter on Bakery Products will be of special interest to the baker and the dairy products manufacturer. It included detailed discussions on milk products used in baking, effect of the heat treatment of milk on its baking quality, skimmilk, whey products and buttermilk in bakery goods and formulas and procedures for bakery goods.

This book is a valuable source of information and will be extremely useful to dairy products manufacturers, dairy research workers, research workers in foods and nutrition and students of dairy science and technology.

W. S. Rosenberger

**60. Public health engineering. A text book of the principles of environmental sanitation. Vol. II,**

**part 3. The food contact.** E. B. PHELPS and W. D. TIEDEMAN. John Wiley & Sons, Inc. N. Y. 213 pp. \$4.00. 1950.

This is a description of relations of food to disease followed by a discussion of sanitation aspects of milk production and pasteurization and of shellfish production and control. Milk and shellfish were selected because they involve special conditions and problems of public health importance and because the general principles that apply to each of these specific foods also can be applied to other foods. There is a chapter on public eating and drinking places and also one on garbage disposal and rodent control.

The chapters on relation of food to disease and those on production and processing of milk are of particular interest to milk sanitarians and milk processors. The chapter on public eating and drinking places contains information of interest and value to sanitarians.

The point of view of the sanitarian is stressed throughout and the book contains many practical suggestions of help to sanitarians in milk or restaurant work.

M. P. Baker

**61. The condensed chemical dictionary, 4th ed.** FRANCIS M. TURNER, ARTHUR ROSE and ELIZABETH ROSE. Reinhold Publishing Corp., New York, N. Y. 726 pp. \$10.00. 1950.

This edition is reputed to contain more than 23,000 entries covering a variety of chemical and related terms, as well as information on a considerable number of "trade mark or brand name products". It constitutes a very useful reference book in the areas covered.

F. E. Nelson

### ANIMAL DISEASES

W. D. POUNDEN, SECTION EDITOR

**62. A comparison of the immunity produced in cattle by the inoculation of Br. abortus strain 19 intradermally, intracaudally and subcutaneously.** A. McDIARMID, Agr. Research Council Field Sta., Compton, Berks. Vet. Record, 62, 25: 361-364. 1950.

This experiment was designed to test in cattle the results of a preliminary guinea pig experiment where an intradermal dose of S-19 vaccine

produced as good results as a 25 times larger dose given subcutaneously. Experimental animals consisted of 42 Ayrshire heifers free of *Br. abortus* infection. Three groups of 10 heifers each were given, respectively, 5.0 ml. subcutaneously under the skin of the chest wall; 0.2 ml. intradermally in the same area; 1.0 ml. intracaudally in fibrous tissue under skin about 0.5 in. from tip of tail. The remaining 12 heifers were the non-vaccinated control group. After majority of heifers were 5 mo. in calf, an infective dose of *Br. abortus* was given via the conjunctival sac. Adequacy of infective dose was demonstrated in the control group when all became infected and 10 calves were born dead. A high degree of immunity was demonstrated in the 3 vaccinated groups, with the 0.2 ml.-intradermal and 1.0-ml. intracaudal being equally as effective as the 5.0-ml. subcutaneous inoculation. R. P. Niedermeier

**63. Preliminary studies on the effect of streptomycin and other agents on *Mycobacterium paratuberculosis*.** A. B. LARSEN, T. H. VARDAMAN and A. H. GROTH, Reg. Animal Disease Research Lab., Auburn, Ala. *Am. J. Vet. Research*, **11**, 41: 374-377. Oct., 1950.

*In vitro* studies showed that concentrations of streptomycin greater than 1 µg./ml. inhibited the growth of *M. paratuberculosis*. Promin, 4-aminosalicylic acid, stilbamidine and pentamidine isethionate were effective only at very high concentrations and were not considered practical in the treatment of Johne's disease. Blood from a cow given 2.5 g. streptomycin/100 lb. in a 24-hr. period inhibited the growth of *M. paratuberculosis* cultures. 4 cows were treated with 8-36 g. of streptomycin over 24-60-hr. periods. Temporary clinical improvement resulted, but the animals became worse after 3-4 wk. and died or were destroyed as incurable. E. W. Swanson

**64. Research on foot-and-mouth disease. II. The cultivation of the virus in explantations of tongue epithelium of bovine animals.** H. S. FRENKEL, State Vet. Research Inst. Amsterdam, Holland. *Am. J. Vet. Research*, **11**, 41: 371-373. Oct., 1950.

A method is presented, with photographs of equipment and procedure, for culturing the virus of foot-and-mouth disease. Fresh bovine tongues from susceptible cattle are used. The deep epithelium is removed in a unique manner and the virus grown on it in the presence of high concentrations of penicillin and streptomycin. 250 ml. of virus of  $10^{-5}$  titer are secured from each tongue. This method has many advantages over cultivating the virus in living animals. E. W. Swanson

**65. Mastitis in the dairy cow.** W. H. HASKELL, Klenzade Prod., Inc. Beloit, Wis. *Milk Plant Monthly*, **39**, 9: 74-76. Sept., 1950.

A discussion of the importance of sanitary milk production and its relation to mastitis is given.

J. A. Meiser, Jr.

**66. Applicator for treatment of diseases of the udder.** H. S. DARLINGTON (assignor to A. H. Wirz, Inc.). U. S. Patent 2,529,836. 3 claims.

Nov. 14, 1950. Official Gaz. U. S. Pat. Office, **640**, 2: 525. 1950.

A collapsible tube having a long smooth tapered outlet, protected by a close fitting cap and designed for introducing medication into cows' teats for the treatment of mastitis and other udder diseases is described.

R. Whitaker

Also see abs. no. 82.

**67. Toxicologic studies of aldrin (compound 118) on large animals.** C. H. KITSELMAN, P. A. DAHM and A. R. BORGMANN, Kans. Agr. Expt. Sta., Manhattan, Am. J. Vet. Research, **11**, 41: 378-381. Oct., 1950.

Aldrin, a chlorinated hydrocarbon insecticide, was fed to cattle and sheep in measured amounts of the pure chemical and from alfalfa hay which had been treated with 0.5 lb./acre 8 d. before cutting. Both biological and chemical assays were made of milk, fat and various other tissues from the animals. The hay contained 8 ppm. of aldrin. No toxic effect followed feeding this hay alone for periods up to 213 d. to sheep or cattle. The aldrin concentration in body fat from the hay-fed animals generally was below 1 ppm. A milking cow fed the aldrin-treated hay secreted none into the milk. Three heifers fed aldrin at the rate of 0.1-0.5 mg./kg. body weight tolerated it for a 64-d. period. After a 100-d. pasture period their fat contained less than 1 ppm. Animals fed from 7.5-2 mg. aldrin/kg. body weight died after 10-41 d. Death was preceded by hyperirritability, incoordination, leg weakness and, in the extended periods, emaciation and stoppage of milk secretion. Meat from animals that died was fed to a dog and produced death from aldrin toxicity. E. W. Swanson

#### CONDENSED AND DRIED MILKS; BY-PRODUCTS

F. J. DOAN, SECTION EDITOR

**68. Device for processing yogurt.** A. STEINBERG. U. S. Patent 2,531,237. 2 claims. Nov. 21, 1950. Official Gaz. U. S. Pat Office, **640**, 3: 994. 1950.

This device, of sanitary construction, is designed for producing yogurt and other fermented dairy products in the home. A bell-shaped vessel, provided with adjustable ventilating ports, is placed over a heated round box on the top of which containers of inoculated milk are placed. Heat, supplied by electric lamps, rises through the vessel and maintains the desired incubating temperature.

R. Whitaker

**69. Forsøg vedrørende Fremstilling af Løbe-og Syrekasein (Experiments concerning the manufacture of rennet and acid casein).** State Expt. Sta. Creamery, Hillerød, Denmark. Report no. 56. 1949.

Only milk of good bacteriological quality and of low fat content should be used for making casein. It should be heated only enough to reach the required skimming temperature. If pasteurized,  $\text{CaCl}_2$  should be added before renneting. The effect on casein quality of added water before renneting is presented. Experiments to determine

the most suitable quantity of rennet, the most suitable temperature for coagulation and the right time for cutting the curd are discussed.

Washing the curd with mineralized water showed that Fe causes a light brownish tint in the casein and a Cu container for the milk produces a bluish tint. Cutting the curd into very small pieces before drying is advantageous. Pressing the curd promptly after rinsing was found desirable. The finer and lighter in weight the particles, the firmer the casein and the lighter the color.

Low-temperature pasteurization is preferable to high-temperature for increased solubility and higher dry matter content. Low fat skimmilk is essential for quality casein. Fresh skimmilk starter (2% for slow acid development and 25% for quick development) give best results. For slow development, the temperature is 20–25° C., depending upon season; for rapid acid production, 30–33° C. Slow acid development yields a lower ash and sugar content product. Quick acid development gives greater solubility and higher viscosity of solutions. In quick souring, heating (50–60° C.) started before coagulation.

Alkaline wash waters can be corrected with HCl. Pressing the curd facilitates the drying process and prevents bacterial action. Drying is speeded by grinding the curd finely. Drying temperatures should not be too high or solubility will suffer. Properly prepared casein can be stored for several years under appropriate conditions. Various practical methods of producing acid casein are suggested from these experiments. The report included 109 pages, several large charts and 12 actual casein samples, mounted in cellophane on the inside of the bulletin.

G. H. Wilster

**70. Process for making casein fibers for felting.** R. F. PETERSON and R. W. JACKSON (assignors to U. S. A. as represented by Sec'y of Agr.). U. S. Patent 2,525,825. 5 claims. Oct. 17, 1950. Official Gaz. U. S. Pat. Office, 639, 3: 731. 1950.

Alkaline casein is extruded into an acetic acid bath to form a tow. After stretching, it is hardened under tension with formaldehyde and an aluminum salt and then treated in a bath of Na acetate and formate at pH 6–8. Following another formaldehyde treatment at pH 6, the fiber is dried to less than 5% moisture. R. Whitaker

Also see abs. no. 59, 80.

## DAIRY BACTERIOLOGY

P. R. ELLIKER, SECTION EDITOR

**71. Metode til påvisning af smørsyrebakterier i vegetativ form. (Methods for determining butyric acid bacteria in vegetative form.)** State Exptl. Creamery, Hillerød, Denmark. Report no. 59. 1949.

The number of spores was determined by the van Beynum and Pette method. Material to be examined was plated on a brain or liver medium, which permitted growth of butyric acid-producing bacteria as well as other types. The butyric acid was determined by the continuous fraction

method. The experiments showed that there may be several times as many butyric acid bacteria in the form of vegetative cells than in the form of spores in milk and cheese. Some butyric acid bacteria did not form spores under the conditions that existed. G. H. Wilster

**72. Untersuchungen über den Purinstickstoffgehalt von Milchzuckerhefen. (Investigation concerning the nitrogen content of purines in lactose-fermenting yeasts).** J. CHRISTOPHERSEN. Die Milchwissenschaft, 5, 2: 55–56. Feb., 1950.

When grown on deproteinized whey for from 2–4 d. at 26° C., the 7 *Torulopsis* sp. cultures were highest both in purine N and in total N, followed closely by the 2 *Saccharomyces* sp. cultures, with the *Zygosaccharomyces* sp. cultures being lowest in both nitrogen sources. I. Peters

**73. Note on the estimation of bacterial populations.** P. B. HUTCHINSON, Dept. of Sci., Ind. Research Auckland, N. Z. N. Z. J. Sci. Technol., 30, 2: 81A–82A. Aug., 1948.

A photo-electric exposure meter is used to estimate light transmitted through bacterial suspensions. W. C. Frazier

## DAIRY CHEMISTRY

H. H. SOMMER, SECTION EDITOR

**74. Eine Schnellmethode zur Ermittlung des Eiweißgehaltes der Kuhmilch. (A rapid method for the determination of milk protein).** E. KOFRÁNYI. Die Milchwissenschaft, 5, 2: 51–54. Feb., 1950.

This method is as sensitive as the Kjeldahl method but requires only 15 min./sample. The method is as follows: In a Kjeldahl distillation apparatus of Parnas and Wagner, 10 ml. of milk are brought to a boil in 10N NaOH. The boiling results in the splitting off of ammonia which in turn is carried with the steam into a condenser. The condensate is collected in 25 ml. N/40 H<sub>2</sub>SO<sub>4</sub> for 10 min. The residual H<sub>2</sub>SO<sub>4</sub> is titrated against N/40 NaOH to the endpoint of methyl blue/methyl red indicator. The ml. of H<sub>2</sub>SO<sub>4</sub> neutralized by the condensate when multiplied by the factor 0.191 gives % protein in the sample examined.

A diagram of the apparatus, description of reagents required and the detailed procedure of the method are given. I. Peters

**75. Immune lactoglobulin.** A. HOLM (assignor to E. R. Squibb & Sons, Corp.). U. S. Patent Application 628,987. Dec. 5, 1950. Official Gaz. U.S. Pat. Office. 641, 1: 318. 1950.

Cows are immunized with antigens of such diseases as whooping cough, diphtheria, smallpox, etc. The milk from such cows is separated and the skimmilk coagulated with HCl at pH 4.6. The precipitated casein is discarded and the whey neutralized with NaOH to pH 7.1. Crude immune-lactoglobulin is obtained by salting out with ammonium sulphate at pH 6.9 or by ethanol precipitation at –5° C. This is purified by reprecipitation, dialyzed until salt-free and finally freeze-dried. The product is suitable for oral



administration for conferring upon human beings passive immunity to the indicated disease.

R. Whitaker

Also see abs. no. 95.

### DAIRY ENGINEERING

A. W. FARRALL, SECTION EDITOR

**76. Lift plug valve.** W. EBERT (assignor to Tri-Clover Machine Co.). U. S. Patent 2,529,544. 9 claims. Nov. 14, 1950. Official Gaz. U. S. Pat. Office, **640**, 2: 447. 1950.

A tapered plug valve of the type used for sanitary fittings has the handle pivoted to the valve plug so that the plug may be lifted slightly out of the tapered valve body to permit easy rotatory movement of the plug. A cover extending over the top of the valve body provides a circular fulcrum upon which the handle may be pressed at any time as a lever for raising the plug. The plug is held in the body by a spring on the bottom.

R. Whitaker

**77. Churn.** A. W. HOLSTEIN and D. F. McCARRON (assignors to Knapp-Monarch Co.). U. S. Patent 2,529,691. 8 claims. Nov. 14, 1950. Official Gaz. U. S. Pat. Office, **640**, 2: 486. 1950.

A motor mounted vertically on the top of a metal or glass jar, drives an agitator of the impeller type which is attached to the lower end of the motor shaft.

R. Whitaker

**78. Aprobvning af Piskeriskaernen, Type G. J. (Experiments with the Whipper-Churn, Type G. J.).** State Expt. Sta. Creamery, Hillerød, Denmark. Report no. 58. 1948.

Of 180 churnings made, 85 were parallel churnings with 3 other types of churns. The whipper churn is of metal and is cylindrical in shape. The body and lid are jacketed, so that either hot or cold water can circulate inside the jacket. The lid is equipped with a safety valve and a small observation window. Inside the churn is a wire whipper which is removed when butter is ready to be worked.

The influence of the volume of cream in the churn on the churning efficiency factor was of little significance when cream with a high fat content was used. When cream with low fat content was used, the churning efficiency was best when the churn was 50 and 75% filled. A whipping speed of 100, 130 or 170 r.p.m. made little difference in churning efficiency. However, the slower the speed, the longer was churning required. Churning time was slightly shorter in the whipper-churn than in the control churn. The churning efficiency of the whipper-churn was slightly better than that of the control churn on most types of cream.

After 3 mo. in storage at 10° F. no difference in quality of the butter made with the different churns was observed. The whipper-churn was slightly less satisfactory for working butter after churning and moisture distribution was not as satisfactory as in the control churn; there was as much as 0.2-0.3% difference in moisture from various parts in a churning. For a churning of 300 kg. cream of 32% fat content, 1.49 kwh

were consumed in 39 min. For working 115 kg. of butter, 0.90 kwh was consumed. The whipper-churn was found to be easy to clean but the bearings of the whipper mechanism easily became loose, resulting in a leakage of cream and a possibility of contamination of the butter.

G. H. Wilster

**79. Eine neue milchtrocknungsanlage in Schweden.** (A new milk drying plant in Sweden.) English summary H. HERZ. Die Milchwissenschaft, **5**, 1: 22-23. Jan., 1950.

Patent has been applied for a new type spray milk drier by Ingenieurfirma Jan Campbell, Rosenlundsgaten, 4c, Jönköping, Sweden. Advantages listed over present type spray driers are: (a) Lower initial cost, (b) more economical operation (requires 30% less heat), (c) higher quality powder (better taste, greater solubility, better keeping quality). The spray drier will be manufactured for use in Sweden and for export. A diagram and pictures are shown. I. Peters

**80. Apparatus for sterilizing.** H. L. SMITH, JR. (assignor to Chain Belt Co.). U. S. Patent 2,531,478. 13 claims. Nov. 28, 1950. Official Gaz. U. S. Pat. Office, **640**, 4: 1165. 1950.

Cans of food, such as evaporated milk, are placed end to end in a long horizontal tubular vessel having a diameter slightly larger than the cans. The vessel is placed between 2 rotating jaws which close the ends and cause the tubular vessel to revolve and the cans within to roll about their longitudinal axes and thus agitate the contents. First, steam and then cooling water is admitted through 1 jaw and discharged through the other to effect rapid heating to the sterilizing temperature and then rapid cooling. By having a multiplicity of rotating jaws fixed on the circumference of a revolving structure, the equipment is made to operate continuously.

R. Whitaker

**81. Apparatus for cooling milk and for filling milk containers.** J. A. KINGSTON (assignor to Gascoignes, Ltd.) U. S. Patent 2,532,557. 2 claims. Dec. 5, 1950. Official Gaz. U. S. Pat. Office, **641**, 1: 150. 1950.

Milk from a vacuum-type milker is discharged into a closed vessel containing a milk cooler. From this cooler the cold milk flows by gravity through a tube into a milk can. The vessels containing the cooler and the can being filled are connected by tubes and maintained under the same reduced pressure as the milker. An automatic cutoff is provided when the can is full of milk.

R. Whitaker

### FEEDS AND FEEDING

W. A. KING, SECTION EDITOR

**82. The relation of feed to an outbreak of bovine hyperkeratosis.** C. OLSON, R. H. COOK and E. M. BROUSE, Nebr. Agr. Expt. Sta., Lincoln. Am. J. Vet. Research, **11**, 41: 355-365. Oct., 1950.

Bovine hyperkeratosis or X disease appeared in a group of Hereford calves about 2 mo. after they had been divided into 16 groups of 10 each



for feeding various test rations. The feed lots were in 2 parallel rows of 8 each with common drinking troughs between adjacent lots. The calves were maintained in their lots regardless of the severity of the disease. The rations consisted of prairie hay alone and with various combinations of alfalfa, di-calcium phosphate, corn, vitamin A concentrate, soybean meal, cottonseed meal or urea. The only lot showing no sickness received only prairie hay. All of the calves in the next lot died from X disease; their ration had been supplemented with alfalfa hay and dicalcium phosphate. Other lots receiving alfalfa hay and dicalcium phosphate supplements also had severe cases of the disease, with exception of the lot which also received corn. Soybean meal and phosphate supplement was fed to 2 lots which developed mild attacks of X disease, but when a vitamin A concentrate was added the symptoms were less severe. Mineral analyses of the water and of each of the rations are presented, with no exceptional departures from "normal". Blood analyses showed a progressive anemia in the sick calves with other values essentially normal. 10 calves were treated with usual therapeutic doses of penicillin, sulfamerazine, KI and Na cacodylate, with no effect upon the course of the disease or the secondary infections. Other cattle at the same experiment station did not show signs of the disease and the herd from which the calves were secured remained free of the disease. E. W. Swanson

**83. A quantitative investigation of some mineral components of wheat plants.** M. STUART, F. G. HARBAUGH and J. DENNIS, Tex. Tech. College, Lubbock. *Am. J. Vet. Research*, **11**, 41: 400-404. Oct., 1950.

Wheat plants were gathered during the period from Oct. to Apr. in parts of 2 seasons. Analyses were made of the whole material, of a water extract and of the residue from the extraction. Considerable variation in composition was noted with varying stages of growth. Average values for the whole material on a fresh basis were 0.713% SiO<sub>2</sub>, 0.099% Ca, 0.0654% Mg, 0.0975% P, 0.223% Na, 0.95% K and 77 ppm. Mn (dry basis). It was calculated that a daily intake of 140 lb. wheat would more than furnish the mineral requirements of cattle. The high Mg content might interfere with the optimum Ca absorption. Wheat has a narrower K:Na ration than that reported for other grasses, hence toxicity to high K would not be expected.

E. W. Swanson

### GENETICS AND BREEDING

N. L. VAN DEMARK, SECTION EDITOR

**84. A recurrence of the semi-hairless gene in cattle.** J. F. KIDWELL and H. R. GULBERT, Univ. of Cal., Davis. *J. Heredity*, **41**, 7: 190-192. July, 1950.

From meager evidence the authors conclude that 5 semi-hairless calves from a Polled Hereford herd were caused by the same gene as described by Craft and Blizzard (1934). The affected

calves had but little hair, a thick hide, small amount of internal fat, were small for their ages and were somewhat wild. This non-lethal trait found in both sexes fits the explanation of a single autosomal recessive. L. O. Gilmore

**85. Dwarfism, an hereditary defect in beef cattle.** L. E. JOHNSON, G. S. HARSHFIELD and W. McCONE, S. Dakota Agr. Expt. Sta. *J. Heredity*, **41**, 7: 177-181. July, 1950.

A new type of dwarfism, in which the body dimensions were proportional to those of the normal except for greater width and blockiness, was analysed genetically from data provided by the station herd. Affected calves could be identified at birth. The calving difficulty experienced, especially in first-calf heifers, was attributed to the increased width and resulted in some stillbirths. Dwarfism and bloat became marked by 3-4 mo. of age. At 2 yr. reduced spermatogenesis was observed in a bull and heat occurred in a heifer.

All affected calves traced several times through each parent to Prince Domino. Because of frequency and distribution it was concluded that the trait was caused by a single autosomal recessive gene. A proposed testing program of breeding prospective sires to 10 heterozygous cows would yield 94.4% (instead of 93.5) of bulls free of the undesired gene provided no affected calves were sired.

L. O. Gilmore

### HERD MANAGEMENT

H. A. HERMAN, SECTION EDITOR

**86. The measurement of milk volume.** W. G. WHITTLESTON and S. A. VERRALL, Ruakura Animal Research Sta., Dept. of Agr., Hamilton, N. Z. *N. Z. J. Sci. Technol.*, **30**, 2: 65-70. Sept., 1948.

A labor-saving, milk-measuring and sampling apparatus is described for use with the releaser type of milking machine. The results indicate that the device is as accurate as the usual weighing and sampling procedure.

W. C. Frazier

**87. Teat cup.** E. J. GIERL. U. S. Patent 2,531,266. 2 claims. Nov. 21, 1950. *Official Gaz. U. S. Pat. Office*, **640**, 3: 1002. 1950.

A vacuum-operated teat cup for a milking machine is described which has a rigid outside wall, a flexible inner wall and an expandible and contractible circular chamber at the end attached to a pulsating source of vacuum.

R. Whitaker

### ICE CREAM

C. D. DAHLE, SECTION EDITOR

**88. Ice cream clinics and their relationship to sales.** E. GOLDSTEIN. *Can. Dairy Ice Cream J.*, **29**, 11: 84. Nov., 1950.

Placing the results of clinics in the hands of the salesmen and the sales department can and does build confidence and morale. The sales forces are informed at all times of the products' rating in the clinics, whether favorable or unfavorable. When reports are unfavorable, the trouble is corrected as soon as possible.

H. Pyenson

**89. Body and texture preferences vary with regional markets.** L. GALLIKER. *Can. Dairy Ice Cream J.*, 29, 10: 66-68, 80. Oct., 1950.

The particular type of body and texture that is acceptable in one market may not be received with equal popularity in another. The manufacturer should not be the final judge of his own product. A consumer clinic will help to establish the body and texture desired. Body and texture can be regulated by composition, such as serum solids, sugar, stabilizers and emulsifiers, by mechanical perfection, by fast hardening and by proper refrigeration. H. Pyenson

**90. Ice cream's "eye appeal" is a colorful sales producer.** F. KIMBLE. *Can. Dairy Ice Cream J.*, 29, 10: 62-64. Oct., 1950.

There is a marked difference in consumers' demands for coloring ice cream. The public should be the judge of the amount of color to use in various ice creams, sherbets and ices. It would be an expensive though profitable endeavor to take frequent polls of consumer acceptance on every basis in any given territory. Ice creams must appear alive and vital; a drab, lifeless appearance never catches the consumer's attention. H. Pyenson

**91. Vorschläge für eine neue Verordnung über Speiseeis. (Suggestions for new regulations on ice cream.)** W. SCHULZ. *Die Milchwissenschaft*, 5, 2: 56-58. Feb., 1950.

A modification of the existing standards and regulations in Germany concerning ices, ice cream and other frozen dairy products is described and recommended for consideration. Primary attention should be given to (a) sanitation, (b) ingredients and (c) methods of manufacture.

Four defined products are proposed to replace the large no. of existing frozen products, namely: (a) Ice cream containing 10% B.F., 8% S.N.F., 12% sugar. The use of eggs, egg powder, nuts, fruits, cocoa, natural and artificial flavors and harmless coloring matter is permitted. Maximum overrun 100%. (b) Ice milk containing 3% B.F., 10% S.N.F., 12% sugar. Other regulations as for ice cream. (c) Fruit ices containing 11% S.N.F., 14% sugar. Other regulations as for ice cream, except max. overrun 80%. (d) Artificial ices of undefined composition, but which shall meet the sanitary requirements (coliform count 100 or less/ml.) of above products. Only naturally occurring ingredients such as sugar, fruit acids, lactic acid, etc. are permitted. I. Peters

**92. Ice cream container insert.** L. C. ROWE. U. S. Patent 2,529,816. 1 claim. Nov. 14, 1950. Official Gaz. U. S. Pat. Office, 640, 2: 520. 1950.

A liner for an ice cream container carrying a split ring in a hemlike arrangement on the outside bottom edge, to prevent upward movement of the liner in the container is described. R. Whitaker

**93. Extrusion nozzle for intrafold ice cream.** A. E. HERSHEY and J. F. GRISSINGER (assignors

to Hershey Creamery Co.). U. S. Patent 2,531,127. 2 claims. Nov. 21, 1950. Official Gaz. U. S. Pat. Office, 640, 3: 965. 1950.

An extrusion head for injecting fruit purees, syrups or ice cream into a continuously flowing stream of semi-frozen ice cream to produce variegated ice cream is described. The puree or syrup is introduced under slight positive pressure through a side arm of small bore with a series of small holes and short nipples therein, inserted at right angles in a cylindrical fitting through which ice cream from a continuous freezer is passed. R. Whitaker

Also see abs. no. 94.

## MILK AND CREAM

P. H. TRACY, SECTION EDITOR

**94. Sanitary milk and ice cream legislation in the United States.** A. C. DAHLBERG and H. S. ADAMS. *Natl. Research Council Bull.* 121. 59 pp. 1950.

The provisions of the laws of 48 states and the ordinances of those of 92 cities with populations exceeding 100,000 which replied to the questionnaire are presented in a series of tables and a brief discussion. The authors point out that this material is based upon the printed laws and ordinances and thus gives little information on what the actual enforcement situation may be, the latter constituting another portion of the study. A number of shortcomings in present regulations are indicated, particularly the relative lack of provisions for the sanitary control of ice cream. The amount of material covered makes it necessary that those interested examine the original to obtain any appreciable value from the information presented. F. E. Nelson

**95. Metallisch-schmirgelige Milch. (Metallic-oily milk.)** M. E. SCHULZ. *Die Milchwissenschaft*, 5, 2: 38-39. Feb., 1950.

This flavor may be due to (a) direct sunlight, (b) excessive feeding of beets, (c) presence of Cu or Fe in milk, (d) oxidation of unsaturated fatty acids or phospholipids. Preventive measures include: (a) avoid exposing milk to direct sunlight, (b) abstain from use of Cu or Fe equipment, (c) use higher pasteurization temperatures (to obtain sulfhydryl compounds), (d) cool milk to minimum low temperature and (e) add vitamin C at from 10-30 mg./l. I. Peters

**96. Dairy cases.** A. MOLDAVAN. *Can. Dairy Ice Cream J.*, 29, 11: 45-46. Nov., 1950.

Cases represent an investment on par with the value of the bottles and bottled milk that they carry to and fro. Industry needs a case that is sturdy, resistant to rough handling, lightweight, easy on bottles, durable, economical in price and maintenance, practical to handle by conveyor or by manual operation and sanitary. After a few years, cases take on a weather-beaten appearance but if taken care of properly may last over 2,000 trips. The cost of the case proper per average trip in an efficient dairy is about 0.035¢ (not including washing, repair and cost of handling).

On a qt. basis, the cost of the case proper is about 0.03¢/qt. delivered. Square milk bottle cases occupy 18.77% less space than the corresponding number of "round" cases. A ratio of filled cases to overnight empty cases is approximately 10:1. Mechanical cleaning of cases costs between 0.01–0.05¢/case. Hand washing is estimated to cost about 5¢/case. The advantage of lateral motion of cases is that it makes room for 23% more cases on the conveyor and allows side-handling which is safer for untrained men. H. Pyenson

**97. Milk crate.** W. L. RAWN, JR. (assignor to Kaiser Aluminum & Chemical Corp.). U. S. Patent 2,530,481. 5 claims. Nov. 21, 1950.

Official Gaz. U. S. Pat. Office, **640**, 3: 796. 1950.

A light weight milk crate made of aluminum is described. R. Whitaker

**98. Cream remover.** J. P. JONES (assignor to Dairy Specialties, Inc.). U. S. Patent 2,531,630. 9 claims. Nov. 28, 1950. Official Gaz. U. S. Pat. Office, **640**, 4: 1205. 1950.

This device is designed to permit the removal of the cream from the top of the milk in Pure-Pak type paper containers, providing a cream draw-off opening a vent for introducing air above the cream, the whole to be attached to the hinged flap opening of container. R. Whitaker

---

### ASSOCIATION ANNOUNCEMENT

Due to an unfortunate error, the date of the Annual Meeting was stated incorrectly in the January issue. The meeting will be held JUNE 6–8, 1951, at the University of Tennessee, Knoxville.

# EMPHASIZE REGULAR CLIPPING

## for the production of quality dairy products

**Here's what authorities say:**

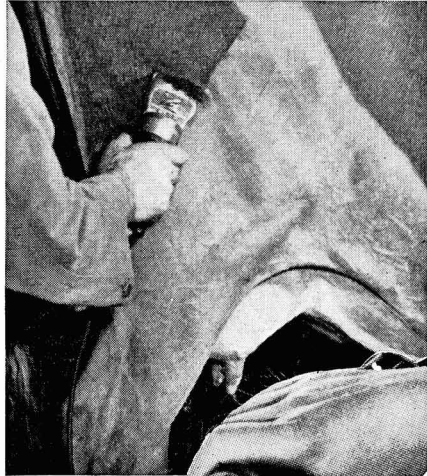
**National Dairies**—*"Clipping udders and flanks is the first step in clean milk production."*

**Univ. of Wisconsin**—*"Clipping saves time when preparing udders for milking."*

**Mich. Agri. Exp. Sta.**—*"Clipping the hair from flanks and udder will reduce the bacteria count of milk as much as 79%."*

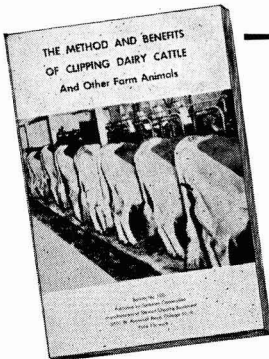
# Sunbeam STEWART ELECTRIC CLIPMASTER

Leading Health authorities say: "A regular clipping program means more wholesome milk. It is an essential step in the production of quality dairy products." Clipping reduces sediment, lowers bacteria, avoids contamination and helps in the control of lice, ticks, etc., which greatly affect milk production.



**Powerful  
air-cooled  
ball bearing  
motor inside  
the handle**

Encourage this good dairy management practice. Educational helps, circulars, and visual aids are available to help you in your program.



**NOW AVAILABLE . . . Bulletin 100—Simple 5-step method of clipping. Free upon request.**

This handy manual graphically illustrates the simple steps that can be easily and quickly learned by anyone. Contains no advertising.

NAME \_\_\_\_\_

ADDRESS \_\_\_\_\_

CITY \_\_\_\_\_ ZONE \_\_\_\_\_ STATE \_\_\_\_\_

**Mail to**

**Sunbeam CORPORATION** (formerly Chicago Flexible Shaft Co.)  
Dept. 141, 5600 West Roosevelt Road, Chicago 50, Illinois

**MINERALIGHT**  
*Ultra-Violet Light*  
**FOR DETECTING MILKSTONE,  
FATS AND OTHER SOILS**



*For Sanitariums, Field Men and Inspectors*

Mineralight is a compact portable long wave ultra-violet light which causes fluorescence in milkstone, fats, and other soils not readily seen by the eye. Used like a flashlight. Operates 110 V-AC or batteries. Adapter available for 110 V-DC. Carrying case optional but necessary for battery operation. Moderate cost. Valuable aid to any size plant. Indispensable in improving sanitary standards. Write for literature.

**KLENZADE PRODUCTS, INC.**  
BELOIT • WISCONSIN

**"HANSEN'S"**  
**DAIRY PREPARATIONS**  
*For High Quality Dairy Products*

- CHEESE RENNET AND COLOR
- COTTAGE CHEESE COAGULATOR
- ANNATTO BUTTER COLOR
- DANDELION BUTTER COLOR
- CERTIFIED BUTTER COLOR
- STARTER DISTILLATE
- ICE CREAM COLOR
- LACTIC FERMENT CULTURE

- ODORLESS TYPE DAIRY FLY SPRAY
- CULTURE CABINETS
- TESTING SOLUTIONS

*Write for Literature*

**CHR. HANSEN'S LABORATORY, INC.**  
MILWAUKEE 14, WISCONSIN

*Marschall*  
**RENNET and COLOR**

Strong, clean Rennet is important in the making and curing of cheese. Use Marschall Rennet generously for maximum results.

**MARSCHALL**  
**DAIRY LABORATORY, INC.**  
MADISON 3, WISCONSIN

Copies of the 20-Year Index covering Volumes I to XX, inclusive are available at the following prices:

**MEMBERS**

- Cloth Bound ..... \$2.35
- Paper Bound ..... 2.00

**NON-MEMBERS**

- Cloth Bound ..... \$5.50
- Paper Bound ..... 5.00

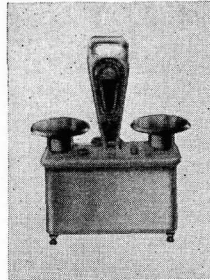
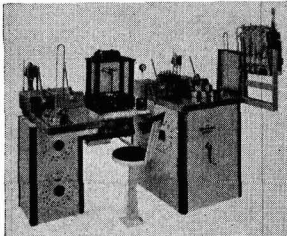
# For accurate results in YOUR laboratory

## Specify Mojonnier

The units shown here are typical of the Mojonnier line of dairy laboratory equipment that has earned a world-wide reputation for accuracy and speed.

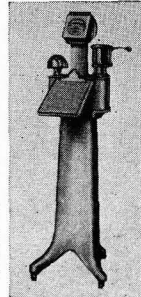
### MILK TESTER

Determines butter fat and total solids content of dairy products. Same accuracy as long, laborious "official" tests, but requires only 1/6th the time on butter fat and 1/17th the time on solids test.



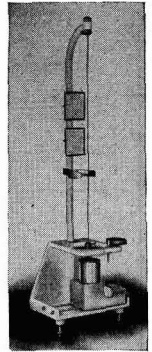
### BUTTER PRINT SCALE

Sensitive to 1/28th ounce. Recommended for weighing pound prints of butter, check-weighing paper containers of milk.



### OVERRUN TESTER

Shows instantaneously both weight-per-gallon and overrun percentage in ice cream. Tests can be made in 5 to 7 sec.



### MOJONNIER-DOOLITTLE VISCOSIMETER

Viscosity can be measured at any temperature. Torsion wire calibrated with extreme accuracy. For determining viscosity of ice cream mix, sweetened condensed milk, and evaporated milk. Simple enough for use on factory control operations.

## Mojonnier LABORATORY EQUIPMENT

#### OTHER MOJONNIER

#### PRODUCTS INCLUDE:

COMPOSITE SAMPLE BOTTLES

BOTTLE SHAKERS • BOTTLE HEATERS • TEST

BOTTLE BATHS • COMPOSITE TEST LIQUID • SAMPLERS,

TRIERS, DIPPERs • ACID BOTTLE TRUNNIONS • MILK WEIGH RACKS AND TICKETS

For specifications and prices, write for Bulletin 209 to:

**MOJONNIER BROS. CO.**  
4601 WEST OHIO STREET  
CHICAGO 44, ILLINOIS

It will PAY you to investigate—

## the GBI VALUE LINE

of Ice Cream Stabilizers and Emulsifiers

In these days of high ingredient and operating costs, GBI stabilizers and emulsifiers demonstrate their value in short order. They earn their way and produce the type of ice cream, ice or other frozen confection you want. Whatever your product or operating conditions, there's a GBI stabilizer and emulsifier to give top performance and profit to you. The GBI VALUE LINE includes the following outstanding products—

*Complete Stabilizer and Emulsifier*—BODY-GUARD and PRIDEX

*Straight Stabilizers*—HY-GUARD and PRIDE

*Straight Emulsifier*—EMEX

WRITE NOW for complete bulletins and prices on the GBI VALUE LINE. These basic formulas cover the field. You will be surprised at their low cost.

Reg. U. S.  Pat. Off.

Manufactured by one of the largest established suppliers in the field of biological sciences.

**GENERAL BIOCHEMICALS, INC.** • 20 LABORATORY PARK  
CHAGRIN FALLS, OHIO

**RESULTS  
SPEAK FOR  
THEMSELVES**

Cultured starters propagated from FLAV-O-LAC FLAKES are uniformly better in flavor, aroma and smoothness..



**YOGHURT CULTURE**

**FLAV-O-LAC FLAKES**

(shown) produce a quart of the finest starter on a single propagation. Single bottles \$2.00.

**SPECIAL FLAV-O-LAC FLAKES "40"** produce 40 quarts of starter on a single propagation. Single bottles \$3.75. Plus postage. Free Cultural Manual of Fermented Milk Products on request.

**THE DAIRY LABORATORIES** 

23rd & Locust Sts., Phila. 3, Pa.

BRANCHES

New York Baltimore Washington  
See our catalog in Dairy Industries Catalog

The Secret of making  
**GOOD  
CULTURED  
DAIRY PRODUCTS**  
is *YOURS* with



**FRIGIDRY**  
Reg. U. S. Pat. Off.

The original, the genuine  
**LYOPHILIZED DAIRY CULTURE**  
2 gm. bottle dry powder  
makes 1 qt. Mother Culture

Manufactured Exclusively by  
**GENERAL BIOCHEMICALS, INC.**  
20 Laboratory Park Chagrin Falls, Ohio

Mention

JOURNAL OF

D  
A  
I  
R  
Y

This helps  
both the  
Advertiser  
and you

S  
C  
I  
E  
N  
C  
E

When  
Answering  
Advertisements



**DIFCO**

*for Microbiological Assay*  
of  
**VITAMINS and AMINO ACIDS**

*Bacto* dehydrated media containing all the necessary nutriment and growth factors for the microbiological assay of vitamins and amino acids are now available from Difco. These basal media require only the addition of graduated amounts of the substance under assay to obtain linear growth of the test organism for construction of the standard curve. The vitamin or amino acid content of the material under assay is determined by adding appropriate concentrations of the test substance to the basal medium and comparing the growth response obtained with the standard.

BACTO-FOLIC ACID ASSAY MEDIUM

BACTO-NIACIN ASSAY MEDIUM

BACTO-PANTOTHENATE ASSAY MEDIUM

BACTO-PYRIDOXINE ASSAY MEDIUM

BACTO-RIBOFLAVIN ASSAY MEDIUM

BACTO-THIAMIN ASSAY MEDIUM

BACTO-TRYPTOPHANE ASSAY MEDIUM

BACTO-VITAMIN B<sub>12</sub> ASSAY MEDIUM

⌘ ⌘ ⌘

The method employed in carrying stock cultures of the test organisms and preparing the inoculum for microbiological assay is important. The following media have been developed especially for carrying stock cultures and for preparation of the inoculum.

BACTO-MICRO ASSAY CULTURE AGAR

BACTO-MICRO INOCULUM BROTH

BACTO-NEUROSPORA CULTURE AGAR

⌘ ⌘ ⌘

BACTO-VITAMIN FREE CASMINO ACIDS, dehydrated, is an acid hydrolysate of vitamin free casein prepared especially for laboratories investigating microbiological assay of vitamins.

*Descriptive literature is available upon request.*

**Specify "DIFCO"**

THE TRADE NAME OF THE PIONEERS

In the Research and Development of Bacto-Peptone and Dehydrated Culture Media

**DIFCO LABORATORIES**

**DETROIT 1, MICHIGAN**