EFFECTS OF PRESERVATIVES

IN FOODS ON THE GROWTH

OF

YOUNG ANIMALS

THESIS FOR DEGREE M. S.

CHARLES JAY OVIATT

1913

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M A S T E R O F S C I E N C E

Chas. J. Oviatt.

June, 1913.

THESIS

~~A Study of the~~ Effects of Preservatives in Foods on the Growth of Young Animals.

INTRODUCTION.

The prevailing high prices of dairy products and feeding stuffs have necessitated the rearing of calves and pigs on skim milk and supplementary feed. In many localities the milk is taken to a whole milk creamery or skimming station where the cream is separated from it, and the skim milk returned for feed. Ordinary methods of skimming present this milk to the young animal in poor shape for consumption and assimilation. Dirty cans, unsteamed skim milk vats, and infection of milk from preceding day, exposure to dust-laden air, and the subjection to the heat of a July sun on the stand in front of the farmer's house, tend to induce changes in the milk deleterious to its value as a food.

Since this milk is not to be used for human consumption, the possibility of preventing this loss of nutriments by the addition of chemical preservatives has presented a practical problem for investigation. Any such preservative might have an accumulative effect as time increased over many years of use in foods for human consumption, but it was thought that no such ill effects would result during the comparatively short feeding period of the young animal. A study of these problems, as they presented themselves, is submitted on the following pages.

Preservative Studied.

The most common preservatives found in milk are: boric acid and borax; formaldehyde, hydrogen peroxide, sodium carbonate and salicylic acid. Many proprietary preservatives are on the market, but their effects are due solely to one or more of the above or like chemicals which may be found in them. Since it was impossible to test all of them, the one appearing of most practical value was chosen. Hydrogen peroxide is oft mentioned, but a few trials in the laboratory showed it to be, not only expensive, but productive of a disagreeable odor and taste in the milk. This may be eliminated by heating the milk to 160 degrees F. but this would be utterly impractibable.

Formaldehyde was chosen because it has a much greater germicidal action than any other and is cheap. It is an oxidation product of methyl alchohol and has a chemical formula of HCOH. Normally it is a gas, but commercially it is known as formalin which is a 40% solution of the gas in water. It is very popular as a disinfectant and is also one of the chief preservatives of food stuffs. For this work formalin was used in the proportion of 1-6000 parts (1-14000 Ius,,0075% of formaldehyde). The investigation resulting in the adoption of this amount as a preservative in this study may be found later in this treatise.

Summary of Previous Work.

In preparing this summary of the work done elsewhere, previously, an effort has been made to include only such results as have a practical relation to the subject in question. Considerable work has been done, especially by foreigh experimentors, upon the chemical changes due to the use of preservatives, but in most cases a preservative other than formaldehyde was chosen. Boric acid and salicylic acid were the ones largely used in their experiments, and since formaldehyde differs from these very greatly in its action no definite conclusions can be drawn by comparison.

In reviewing the literature on the effects of formaldehyde upon the digestion of foods it is found that there exists but little of practical value in this study. Much work has been done, but usually with formaldehyde in the proportions of from 1:25 to 1:2000 which is more than is needed or is ever used; hence the results have but little bearing on our study at this time. These investigations have been with amounts sufficient to insure an interference in digestion with no apparent attempt to learn the minimum amount which could be used without a retardation of the processes and factors of digestion.

The following experiments have been chosen as those of most value in this study. The results have been questioned, in some cases, but since the knowledge is so limited a summary is presented. It is imperative that the student should bear in mind, continually, the

fact that all these experiments had but one object in view, viz., the effect of formaldehyde upon the human system. The effects upon the animal used in the experiment were noted only that a comparison might be made. Furthermore, these animals received several times as much of the preservative as is needed to produce complete preservation of food for reasonable periods of time. Under these exaggerated conditions some very definite conclusions have been drawn, but a close study of the work oft suggests a prejudice in disfavor of the formaldehyde. The data is presented in chronological order, and not in reference to its value in this study.

Annette.

One of the earliest experiments with formaldehyde and its effects was carried on by Dr. Annette (Lancet 1899), a professor in University College of Liverpool, England. He experimented with different preservatives and their effects upon young kittens. His results with formaldehyde, only, are summarized here.

Kittens of about three weeks of age were used. This was considered the least age at which a kitten would exist on cow's milk alone. According to Dr. Annette, they were divided into lots of equal size and weight. No record of original weights is given nor the amounts of milk consumed by each lot of kittens. This omission, together with no record of appetites or conditions suggests a rather serious error on the part of the investigator. A record of gains is given in the table on following page.

Gains in Grams.

Kittens receiving 1-50,000 HCOH.			Control Kittens.	
1st week	Av.5 kittens	8.2	Av.4 kittens	14.7
2nd "	" 5 "	3.6	" 4 "	10.0
3rd "	" 5 "	28.4	" 4 "	89.2
4th "	" 4 "	8.2	" 4 "	96.7
5th "	" 2 "	145.2	" 4 "	60.5
Total increase		177.2		251.1
Kittens receiving 1-25,000 HCOH.			Control Kittens.	
1st week	Av.4 kittens	38.0	Av.3 kittens	77.6
2nd "	" 4 "	114.2	" 3 "	129.6
3rd "	" 4 "	0.5	" 3 "	61.3
4th "	" 4 "	30.5	" 3 "	104.5
5th "	" 4 "	7.2	" 3 "	38.0
6th "	" 4 "	7.2	" 3 "	0.6
Total increase		196.6		325.7
Kittens receiving 1-12,500 HCOH.			Control Kittens.	
1st week	Av.5 kittens	20.4	Av.3 kittens	46.5
2nd "	" 5 "	25.4	" 3 "	68.3
3rd "	" 5 "	18.0	" 3 "	80.6
4th "	" 3 "	13.3	" 3 "	63.0
5th "	" 3 "	55.3	" 3 "	34.0
Total increase		96.4		312.5

It will be noticed that each lot of kittens fed on preserved milk had a control lot in comparison. Out of the ten kittens fed as controls no deaths occured, while with fourteen fed on formaldehyde treated milk five died during the course of experiment. In the lot fed formaldehyde in proportion of 1-50,000 one died the fourth week and two the following week. It is recorded that one of the remaining two died soon after the experiment closed. In the lot fed formaldehyde in the pro-

portion of 1-25,000 no deaths occured, but a celeterious effect may be noted by a comparison of gains made. In the lot fed formaldehyde in the proportion of 1-12,500 two died the fourth week and the total increase was relatively small. In the control kittens in the second division of the table we find some very peculiar gains in the last three weeks. The gain for the fourth week was 104.6 grams, the next week it had fallen to 38.0 while there was practically no gains in the week following. Notes or comments in Dr. Annette's work are lacking to show causes for such extraordinary changes in growth.

In justice to this study it might be well to state that Dr. Annette received some very severe criticism from eminent men of that time. One of the chief criticisms was that no record was kept of the amounts of milk consumed. This made it impossible to even approximate the amount of the preservative taken into the system. In order to draw any definite conclusions, it has been argued, the amount of preservative consumed in proportion to the body weight must be known. Dr. Annette was called before the British Commission to give testimony on his work. At this time he was severely criticized by Dr. Rideal, an English scientist of much note, who claimed that kittens of the age mentioned could not be reised on cow's milk alone. But on the other hand we have the results of Dr. Annette, whose honesty apparently never was questioned, to show that he did not lose a single control kitten.

While the exceedingly varied gains from week to week make his work less positive, the results stand as the work of an eminent man in a position of trust and honor and must not be discarded. It is, however, the only experiment showing a marked deleterious effect by the use of formaldehyde as a preservative when fed in small quantities.

A. G. R. Foulerton. (English Investigator)

This experiment with formaldehyde is described in the "Lancet" 1899 (p1427). In testing the effects of formaldehyde upon living tissues he placed several goldfish, as are found in small aquariums, in water containing the preservative in the proportion of 1-40,000. A close study showed no ill effects even after several days in the water. Furthermore, he tried a proportion of 1-10,000 upon frogs with no apparent ill effects. It would not be illogical to suppose that if this preservative is very active in the living tissues that it would affect the fish and frogs forced to live in it for periods of time.

At another time this same man reports that formaldehyde in the proportion of 1-40,000 had but very little effect upon the digestion of proteids. The percentage given was four while in the digestion of starch there was a slight retardation. In the case of rennet coagulation there was no effect when the amount stated was used.

A. Bach. (Chem. Central Blatt 1897 p.429)

This investigator reports that albumin in an aqueous solution treated with formaldehyde, the amount of which was not stated, could not be percipitated by heat and the same was percipitated with more difficulty by treatment with 95% alcohol.

Bliss and Novy (Jour. of Expt. Medicine 1899 IV.)

Experiments with formaldehyde treated milk resulted in the conclusions that this preservative in the proportion of 1-2500 prolonged the action on the cassin and rendered it more difficult to digest and to coagulate with rennet. When smaller amounts were used there was no effects apparent.

Dr. T. Landon Brunton (Eng.)

In conclusion of experiments and results with the use of preservatives in milk and other foods this chemist advised that "the poisons are actually formed by the decomposition of food products and the question is whether formaldehyde or the products of decomposition are the least harmful." In his opinion the preservatives are the least harmful to the system. His experimental data have little bearing upon the condlusion given.

Chako. (reference by Savage)

By using a bacterial count on milk treated with formaldehyde in the proportion of 1-10,000, Chako decided that there was a slight killing off of bacteria during the first twenty-four hours. During a short period following there was a slight increase and then

growth increased very rapidly--apparently ignoring the presence of the formaldehyde. When a proportion of 1-20,000 was used a falling off in number of bacteria resulted for a very short time and then the increase was exceedingly rapid as in the case of untreated milk. In the proportion of 1-40,000 the formaldehyde had little or no effect upon the rapidity of growth.

Dr. von Behring.

This eminent scientist has done much with the tubercule bacillus in milk. Although it has been impossible to secure absolute data of experiments he advises that the use of formaldehyde in milk in the proportion of 1-10,000 will destroy any germs of tuberculoses which might be in the milk by secretion from the udder direct or by an aerial route. A further discussion of this will be found in the laboratory tests by the writer.

Jensen. Denmark College at Copenhagen.

In Dr. Jensen's book, translated by Person, is found the following:- "The presence of .008% formaldehyde (.02% formalin or 1-12,500 formaldehyde) is able to check coagulation of milk one hundred hours. The addition of formalin causes no changes in the fats. Formaldehyde has, however, a decided effect upon albumin and as it has a deadly effect upon the protoplasm of bacteria, so it is a powerful poison for the animal cells and tissues." How Dr. Jensen came by these conclusions is not given, but since he is considered quite an authority, these statements must be considered of

practical value.

D. S. Hall and H. S. Hammond of S. E. Ag. College of Wye, Eng.

These men were appointed by the British Commission to investigate the effects of preservatives upon the growing tissues and the ultimate effects by continual consumption of food products. This is the only experiment on record, so far as the writer could determine, where children were used as subjects. In this case only three were used and hence the results are not very positive or substantial. Two boys, one five and the other two and one-half years of age, both being strong and healthy, and one girl four years of age, "delicate, poorly nourished and developed" were used in these tests.

The younger boy consumed 200 grams of bread, 550c.c. of milk, 20 grams of butter, 30 grams of sugar, 50c.c. of water and 5 grams of taffy. The other children were fed a similar ration varying somewhat in amounts. The following table shows the amounts fed and the effects upon the children.

Healthy Boy, two and one-half years old.

	N. of Food.	N. of Faeces	Fat of Food	Fat of Faeces.
1st 7 days-No preservative	48.20	4.14	303.95	18.27
2nd 7 " 1-18000 HCOH	46.62	4.49	277.12	19.52
3rd 7 " 1- 9000 HCOH	44.62	3.54	272.33	15.51
4th 7 " none	46.69	3.78	366.05	17.52

Healthy Boy, five years old.

1st 7 days-No preservative	53.76	4.36	307.89	18.36
2nd 7 " 1-18000 HCOH	57.61	4.32	299.62	19.06
3rd 7 " 1- 9000 HCOH	55.66	3.83	294.65	18.74
4th 7 " none	61.64	4.61	306.49	16.99

Sickly Girl, four years old.

1st 7 days-No preservative	46.56	3.78	272.68	12.44
2nd 7 " 1-9000 HCOH	49.11	5.42	263.13	21.71
3rd 7 " none	48.95	4.80	294.44	13.65

From the above table it appears that, in the cases of the healthy children, the formaldehyde had acted as a stimulant in the digestion of the foods. In fact the most complete digestion takes place in the children to which the greatest amount of formaldehyde was fed. In the case of the sickly girl there was an opposite effect. This, however, should not be considered as a point against the use of the formaldehyde for any change of diet might have caused the less complete digestion. It will be noticed that this formaldehyde was added to the diet very abruptly and that in amounts never found in the preservation of foods. In other words, this is a much more severe test than our study would necessitate. This sudden addition of formaldehyde or any other chemical might easily have caused digestive disorders without implicating

a deleterious effect by the chemical as such. The investigators, after careful study of data, concluded that formaldehyde in amounts even as high as 1-9000 had no ill effects upon the general health of yound children. Since these men were commissioned by the British Gov't to investigate this problem, their conclusions must be considered carefully.

A. D. Hall, Principal S.E.Ag.College of Wye,Eng.

This experiment was carried on by the use of young pigs, and included several of the common preservatives. The work with formaldehyde is interesting and of value in this study. Six pigs were used and they received from 2 to 4c.c. of the preservative (about 1-1,500 which is ten times more than is necessary for preservation of foods) in the milk which was a part of their diet. These pigs remained in good health and appetite and gained steadily throughout the seven weeks of the experiment. During this time they gained 94 pounds, nearly 2 pounds per day, and apparently suffered no inconvenience from the consumption of this preservative. Analyses were made of the food and faeces to determine whether there had been a change or interference of digestion. No positive results were obtained and no data is given. The final conclusion was that formaldehyde even in a proportion of 1-1,500 did not have ill effects upon digestion.

Doane and Price, Maryland Bul. 86.

These men made an exhaustive study of the effects of preservatives and their results are, perhaps, the most valuable quoted in this study. The experiment was started when the claves were two weeks old. Since the excreta was to be collected it was necessary for the calves to remain standing. In order to provide a chance for rest a bran sack was placed under the body of the calf and fastened to the sides of the crate in which it was confined. This allowed the calf some rest, and at the same time no excreta was lost. In the first experiment, formaldehyde was added to the milk in the proportion of 1-10,000 just before feeding.

The general plan of the work was to feed the calf a preliminary period of three days which was considered a sufficient length of time for all food to pass through the digestive tract, as nothing but milk was given the calf. Then the faeces were collected for three days, the calf meanwhile being confined in a crate. A longer period for collecting the faeces would undoubtedly have been better, but it is likely that three days was about the limit of time a young calf could be confined in an upright position without becoming so fatigued as to influence results. The faeces were collected every day, weighed and sampled, the size of the sample being in direct proportion to the amount excreted. These samples were acidized and added together for analysis for fat and protein.

The milk fed the calves the first year of experiment, was taken from the same two cows throughout the period. It was thought that this would be better than taking from the entire herd, as milk from different cows is supposed to differ in digestibility.

No faeces were collected from calves that commenced to scour after the beginning of the feeding. In such cases the feeding was stopped untill the calf regained its normal condition or was continued a sufficient length of time after the scouring ceased to insure that it would have no effect on the results of the test. In cases where calves were not satisfactory through refusing occasionally to drink the full amount of milk they were discarded and other calves substituted. About the quantity of milk to keep the calf in good growing condition was fed, the amount being always the same for all calves.

Tables X and XI show the composition of the milk fed, and the faeces collected in the different tests. These can be identified with the table showing the per cents digested in the different trials by the index numbers. The Babcock test was used for fat determinations of the milk. Results with preservatives other than formaldehyde are included to show that formaldehyde is more practical than others suggested.

Composition of Milk used in Expt.

Index No.					Water %	Protein %	Fat %
2581	Whole	milk	plus	Boric acid.	87.00	3.36	3.80
2589	"	"	"	" "	87.00	3.20	4.00
2597	"	"	"	nothing	86.50	3.70	4.40
2604	"	"	"	Salicylic acid	86.50	3.32	4.20
2611	"	"	"	Formaldehyde	86.50	3.36	4.10
2627	"	"	"	"	86.50	3.59	4.60
2633	"	"	"	Salicylic acid	86.50	3.32	3.80
2539	"	"	"	nothing	86.50	3.52	4.40
2645	"	"	"	Boric acid	86.50	3.41	4.10
2661	"	"	"	Formaldehyde	86.50	3.55	4.60
2664	"	"	"	"	86.50	3.55.	4.60
2666	"	"	"	nothing	86.50	3.64	3.60
2673	"	"	"	Borax	86.50	3.18	3.80
2679	"	"	"	"	86.50	3.22	4.00
2683	"	"	"	nothing	86.50	3.64	4.60
2685	"	"	"	Salicylic acid	86.50	3.18	3.80
2701	"	"	"	nothing	86.50	3.70	4.50
2709	"	"	"	Boric acid	86.50	3.40	4.60

This simply shows, aside from being of value in further study of succeeding tables, that the chemical composition of the milk is not materially changed by the addition of chemicals even in the case of formaldehyde which is said to be fifty times as strong a germididal agent as boric acid. On the following page will be found a table showing the composition of the faeces in the digestion experiments.

Composition of Faeces in Digestion Expt.

Index No.	Description of sample					Water %.	Protein %.	Fat %.
2589	Calf	1	Whole	Milk plus	Boric acid	81.82	8.59	2.32
2592	"	2	"	"	" "	83.26	6.72	3.40
2600	"	2	"	"	nothing	63.72	6.45	4.94
2607	"	2	"	"	Salicylic acid	73.64	7.26	8.80
2614	"	2	"	"	Formaldehyde	88.98	5.95	2.00
2630	"	3	"	"	"	79.59	11.69	4.43
2636	"	4	"	"	Salicylic acid	73.71	12.10	6.61
2642	"	4	"	"	nothing	83.43	7.96	2.47
2648	"	4	"	"	Boric acid	75.49	6.97	2.25
2663	"	5	"	"	Formaldehyde	72.37	10.87	5.24
2666	"	4	"	"	"	80.72	5.33	1.96
2671	"	5	"	"	nothing	79.42	8.99	4.55
2676	"	4	"	"	Borax	85.29	5.56	1.40
2682	"	5	"	"	"	[illegible]	[illegible]	[illegible]
2686	"	4	"	"	nothing	81.21	4.71	2.14
2689	"	5	"	"	Salicylic acid	84.96	6.60	3.10
2704	"	5	"	"	nothing	84.88	4.39	1.61
2712	"	5	"	"	Boric acid	80.15	4.80	1.15

It is evident from the data above that formaldehyde does not materially interfere with the digestive processes. In fact, an average of the above shows that more protein and fat is digested with this preservative than with others or no preservative. Since our study is, not to prove its value as a stimulant to digestion, but rather, to show its lack of ill effects when used as a practical preservative, the above data is of great value.

On the following page may be found a table showing the digestibility of whole milk preserved with formaldehyde.

Digestibility of Whole Milk Preserved with Formaldehyde.
(Preservative added just before feeding).
Duration of preliminary period 3 days.
" " digestive " 3 days.

Index No.		Fresh substance grms.	Dry substance grms.	Water %	Prot. grms.	Fat grms.
	Calf 9, age 35 days					
2540	Milk fed	18714	2432	87.00	632.53	711.13
2534	Total excreted	535	151	70.56	32.56	14.60
	Digested				599.97	696.53
	% digested				94.85	97.94
	Calf 8, age 38 days					
2547	Milk fed	18714	2936	86.50	640.01	767.27
2551	Total excreted	750	131	82.54	30.80	18.82
	Digested				609.11	748.45
	% digested				95.17	97.56
	Average-2 tests of milk preserved with HCOH				95.01	97.75
	" whole milk untreated				94.79	96.82
	Difference in favor of milk preserved with formaldehyde				.22	.93

The first trial was with Calf 9 which was five weeks old. He was in normal condition at commencement of experiment, but when placed in crate his manure was hard and dry. The table shows that 94.85% of the protein and 95.17% of the fat consumed was digested.

The second trial was with Calf 8 which was a trifle older than Calf 9. Everything during the period was normal. Data shows that 95.17% of the protein and 97.56 of the fat consumed was digested. The average for the two trials was 95.56% of the protein and 97.75 of the fat. Comparing this with the digestibility of untreated milk we find .22% of the protein and .93% of the fat in favor of the milk treated with formaldehyde.

In the following table is found the gains or losses in weights during the three days of the feeding experiment.

Gains or Losses.

Calf No.		Index No.	Preservative	Gain
1	Age 14 das.	2584	Boric acid.	3
2	14	2592	" "	5
2	22	2600	none	2
2	28	2607	Salicylic acid	3
2	34	2614	Formaldehyde	5
3	14	2730	Formaldehyde	9
4	25	2636	Salicylic acid	4
4	31	2642	none	2
4	37	2648	Boric acid	-4
5	14	2663	Formaldehyde	4
4	43	2666	Formaldehyde	2
5	20	2671	none	0
4	49	2676	Borax	-1
5	26	2682	Borax	5
4	55	2686	none	0
5	32	2689	Salicylic acid	2
5	44	2704	nonw	2
5	50	2612	Boric acid	-2
	Average gain for		Boric acid	.5
	" "	"	No Preservative	1.0
	" "	"	Salicylic acid	3.0
	" "	"	Formaldehyde	5.0

In the four trials with formaldehyde there is shown a steady gain. Unless this preservative acts as a stimulant of digestion it is hard to understand why it should show greater gains than from untreated milk in which case the gains were only one-fifth what they were in the case of formaldehyde treated milk. At least the results show no ill effects from the use of this preservative.

Realizing that artificial experiments give less positive results than actual feed tests these men outlined an experiment with two calves. These calves were in perfect condition at beginning of the test. The following table is self explanatory.

	Calf A	Calf B.
March 11	88#	109#
" 26	101	124
April 1	102	125
" 15	124	146
" 24	131	151
Gain in 44 days	43#	42#

Thus it is seen that these calves made very satisfactory gains in weight. Their condition remained good and no ill effects were apparent. From these results it "would be hard to conclude that preservatives are harmful and therefore should be prohibited". In any case there are good gains shown and that is all concerned with this particular study.

In general conclusions Drs. Doane and Price advise that there is a certain stimulation, caused by the use of formaldehyde, in the digestive processes. As has been stated 1.31% of the protein and .99% of the fat is digested in excess of that when no preservative is used. The final conclusion is that formaldehyde does not have ill effects upon the growth of young animals.

T. M. Price, Assistant in Biochemistry, B.A.I.

This experiment by Price is detailed in the report for 1903. It has, probably, the greatest value in this study of any laboratory experiment cited. While it is impossible to give in detail all the results a few of the important ones are tabulated.

Action of Calf's Rennet.

All enzymes were secured from fresh extracts from young animals' glands. Milk was treated with varying amounts of formaldehyde and rennet was added at intervals as recorded in table.

From Flask	Amount HCOH	Mixtures of Milk and HCOH Tested. After 10 min.	After 24 hrs.	After 48 hrs.
1	Control	Solid 7 min.	Solid 7 min.	Solid 7 min.
2	1-50	No Coag. 18 hrs.	Same	Same
3	1-125	" " "	Same	Same
4	1-250	" " "	Same	Same
5	1-500	" " "	Same	Same
6	1-1250	Solid in 10 min.	Same	Same
7	1-1875	" " "	Same	Same
8	1-2500	Solid in 7 Min.	Same	Same
9	1-3750	" " 7 "	Same	Same
10	1-5000	" " 7 "	Same	Same
11	1-7500	" " 7 "	Same	Same
12	1-12500	" " 7 "	Same	Same

It is therefore concluded that HCOH in the proportion of 1-2500 or less does not retard coagulation of milk by rennet. Proportions of 1-1875 retards and 1-500 inhibits coagulation for eighteen hours.

Pepsin.

The pepsin used was extracted from the stomach of a young pig. Dr. Price says, "It has been demonstrated by Hammarsten and others that milk, when subjected to the action of pepsin hydrochloric acid, is broken up into calcium paracasein and a small amount of albumose-like albumin. The paracasein is percipitated, and may be recognized after digesting the milk for a short time; the paracasein separating and finally decomposing with the formation of Paranuclein. Any foreign matter that interferes with the formation of paranuclein interferes with the digestion of the milk. This feature was taken to determine when the formaldehyde affected the digestibility of the milk. It was found upon the addition of formaldehyde in the proportion of 1-50 the pepsin digestion was retarded, while in a stronger solution or in the proportion of 1-25 the digestion was materially interfered with; and in the proportion of 1-125 or less the digestion was normal with the control."

In the case of steapsin a similar experiment brought out the fact that formaldehyde in the proportion of 1-35 entirely prevented the action of this enzyme. In the proportion of 1-50 its action was retarded, but in weaker solutions the enzyme acted identical with the control.

In the preceding experiments enzymes were used, whose activity if interfered with, would materially

affect the digestibility of the food. Since formaldehyde is used often in food containing much starch Dr. Price tested the effects of it upon the ptyalin and amylopsin, important enzymes in the conversion of starch. The food in our particular study is skim milk, but since whey which contains a large amount of starch and sugar, might also be treated with formaldehyde to advantage, the results are included. He found that the action of ptyalin was not in the least retarded until a strength of 1-1250 or more was used. In less strengths the action was perfectly normal. Since the strength necessary for retardation is about one-tenth that proposed for use, there can be no danger of affecting this enzyme.

A similar experiment with amylopsin showed that formaldehyde had no effect upon the action of this enzyme on starch until a proportion of 1-1000 was added. This is also much above the proposed addition hence the effects on amylopsin may be disregarded.

In summarizing the results of the above experiments it is evident that formaldehyde may be added to milk or other foods in the proportion of 1-2500 without affecting the action of any of the enzymes of digestion studied.

However, there is an enzyme, about which little is known except that it is important to perfect digestion. It is a proteolic enzyme known as galactase. A detailed account of the experiment may be found in

the report mentioned. From the data given, it is evident that the action of this enzyme is retarded only when a very concentrated solution of formaldehyde is used as a preservative. A proportion of 1-1000 or less has no effects of retardation of action. Babcock and Russell (Wis. Ann'l Rpt for 1896) claim that the addition of formaldehyde prevents the action of this proteolytic enzyme, but they give no experimental data to prove their conclusions. Since Dr. Price's work is so thorough, it may be concluded that his data is reliable. A complete summary of the report follows:-

"(1). Formaldehyde added to milk in the proportion of 1-2000 preserved it for 48 hours.

(2). Formaldehyde added to milk in the proportion of 1-2500 has no effect on the activity of the fresh enzymes, rennet, pepsin, pancreatin, and steapsin.(in vitro)

(3). Formaldehyde added to starch in the proportion of 1-2500 has no effect on the conversion of starch by the enzymes ptyalin and amylopsin (in vitro).

(4). Formaldehyde added to milk in sufficient quantity to preserve it 48 hours—1-2000—does not interfere with the action of the enzyme galactase (in vitro).

(5). Formaldehyde added to milk in the proportion of 1-2000 prevents the development of the more common bacteria found in milk, and when added in the proportion of 1-1560 kills them.

(6). Formaldehyde may be added to milk in sufficient quantities to prevent the development of some of the more common bacteria and therefore preserve it, and still have no deleterious effect upon the digestibility of the milk (in vitro)."

The value of this work in our study must not be underestimated. His work has been very exhaustive and his conclusions have a direct bearing upon our study. From them we can see that there would be no danger to the action of any of these enzymes when formaldehyde is added in the proportion of 1-15000 as is the amount used in our work.

H. W. Wiley, Ex-Chief of Bureau of Chemistry.

Under title of "Influence of food preservatives and artificial colors on digestion and health", Dr. Wiley records some investigations for the U. S. Gov't in 1908. The record of these tests is too long to reprint here, hence, a general summary only will be included.

Dr. Wiley chose for the subjects of his tests several healthy young men and with the aid of his staff of chemists he made careful studies of the effects of preservatives on health. The amount of food and excreta was carefully determined and recorded. These were analyzed and notes taken concerning the condition of the young men in the test. As is usually the case, much more formaldehyde was used than is necessary to preserve.

After making several conclusions, he says, "The medical data plainly indicate that formaldehyde, even when given in small quantities, is an irritating substance to the mucous membrane, and therefore, the normal organs are at first actively stimulated to rid themselves of the irritating foreign substance. It is not strange, therefore, that this preservative had a marked stimulative action on those organs and cells secreting the various digestive juices. It is evident that when the digestive and secretory organs of the body are excited to unusual activity by such extraneous body having no condimental value, they act in

self defense and it would be wholly illogical to conclude from this increased excitation that these bodies are helpful to digestion and conducive to health.***** ********* The final conslusion, therefore, is that the addition of formaldehyde to foods tends to derange metabolism, disturb the normal functions and produce an undue stimulation of the secretory activities, and therefore, is never justifiable."

Since Dr. Wiley, at the time of these experiments, was making a fight for pure food, it would not be amiss to suspect a prejudice in disfavor of the preservative used. The work has been done in detail and the data seldom varied sufficiently to justify positive conclusions to be drawn. In cases where a better digestive effect was produced by the use of the formaldehyde, the excuse is offered that this was simply an irritation on the part of the glands of the digestive organs. This may or may not be true, but since our desire is to ascertain the effects on successive gains in the case of domestic animals, his conclusions in this case have little bearing, for it is desired to feed this preservative only a short period of time, and that for actual gains. Furthermore, there is the "personal" element entering his tests which he has seen fit to disregard. The knowledge of the fact that one is being fed a chemical to ascertain its effects would tend to induce digestive disorders. In this respect, an experiment with human beings is not as

valuable as one with animals that have no knowledge of the surrounding conditions. In the report of the condition of these men from time to time, it is stated that some were "feeling generally ill", and it would not be illogical to suspect that part of this feeling was purely mental. On the other hand, the results stand as a standard work, and aside from the fact that much more formaldehyde was used than is necessary, there is little reason for questioning the conclusions.

Senior Theses 1911 and 1912.

During the years above mentioned, some seniors in the Michigan Agricultural College made a study of the effects of formaldehyde when fed to pigs. While this work was far from exhaustive, the results are interesting. In the first year, formaldehyde was fed in the proportion of 1-2500 in skim milk to five young pigs. Careful records were kept and compared with records of pigs fed sour skim milk, and another lot fed a grain mixture. Computation shows that the lot fed formaldehyde treated milk retained much more of the protein fed than did either of the other lots. The final conclusion of their studies is "That milk kept sweet with formaldehyde is a much more valuable feed than milk allowed to sour, and fed in that condition.

In the second experiment, which was similar in its scope, formaldehyde in the proportion of 1-6000 was used. This amount is about two and one-half times more than is necessary, according to other investigators. The data shows that the cost per pound of gain was less in the case of the lot fed formaldehyde treated milk. The percentage increase in weight of this lot also exceeded that of the other two lots fed in comparison. They conclude, therefore, that "skim milk kept sweet by the use of formaldehyde is a little better for feeding pigs than skim milk that is sour."

Results of other experimentors might be given, but they would not vary greatly from those preceding. From the work reviewed, it is evident that there is a wide difference of opinion in regards the effects of this preservative on digestion and growth. With the exception of Dr. Wiley's results, the more recent the investigation, the more favorable is the conclusion toward the use of formaldehyde. As has been previously stated, there was a prejudice, apparently, against the use of it, and all results were made to show its ill effects. Dr. Annette's work is largely quoted by all other early investigators, but it is evident that his conclusions have but little value in our study, for the amounts used were many times that proposed in our work. The work by Hall and Hammond with the young children

offsets the results obtained by both Wiley and Annette, while Boone and Price have shown that formaldehyde, in amounts necessary for preservation only, is not harmful to the digestion or health of young calves. In fact, a general summary of the work done previously leads one to believe that this preservative, when care is exercised in its use, may become a very valuable aid in the retention of nutrients in some of our easily decomposed foodstuffs, and, without having a deleterious effect on the growth of the animal concerned.

Preliminary Work.

Before starting the laboratory experiments, or practical tests, it was decided to make a thorough study of the problems under Michigan conditions. The work, which is very limited, done in other stations was carefully stucied in order that work would not be duplicated. A careful survey of all previous work was made and data studied. Later, inspection trips to some of the best adjacent creameries and skimming stations were made, that a better knowledge of actual conditions might be gained. A letter of inquiry was sent to practical creamery men of the state. A portion of the letter follows:

"It is proposed to make a study of the advisability of the use of preservatives in milk, especially during the summer months, from the time it is skimmed until it is fed to the growing calf or pig. We hope to prove that the preservative will prevent the loss of valuable nutriments in the milk without having a deleterious effect upon the growth of the young animal. Please note this has nothing to do with milk for the human family, where its use extends over a period of many years. In your experience, has anything of this nature been tried, and with what results? In actual practice, is there anything to prevent its use? Any information or suggestions will be appreciated."

Over two hundred of these letters were mailed, and about seventy answers were received. A general summary of these answers may be gained from the portions, included below, of letters from prominent creamery men. "We feed considerable butter-milk, and know that we get better results when fed fresh than when held two or three days. If we could use something that would keep it as good as when made for a few days, with no harmful results, we would be much pleased, and the discovery would be of much value." Another says, "I see nothing to hinder its use if you could find something that would not injure the animal. I believe if you could discover something of this kind, it would be a great help in bringing back the whole milk system of running creameries, which we know would improve the quality of the butter."

From the many answers, it was concluded that there existed nothing in actual creamery practice to prevent the use of a preservative, and hence, the problem was limited to the determination of the correct amount of the preservative necessary, and the actual effects on the growth of young animals.

Laboratory Work.

Considerable of the laboratory work was done in the bacteriological department. The first work of any consequence was the determination of the amount necessary to inhibit the development of bacterial growth in the milk under the temperatures which would be prevalent in actual practice.

This determination was made by the use of litmus lactose agar plates. It was found, after much experimentation, that in the proportion of 1-500 formaldehyde rendered the milk sterile, even when exposed to the air for several weeks. A greater amount killed the acid enzymes completely. These results were gained by the following method:- Skim milk, six hours old, was plated in order to ascertain the initial number of bacteria per cubic centimeter. Samples of this milk were treated with various known strengths of formaldehyde solutions, and placed in a constant temperature (78 F.) room. Each twenty-four hours a bacterial count was made. Careful notice, also, was made of the nature of the surviving bacteria from day to day. The time of loppering and the acidity was also recorded. Data regarding a few of these tests are given in the following table, and they may be regarded as fairly typical of the full number of tests.

Bacteria in Formaldehyde Milk.

Sample No.	Age.	Initial Pacterial Count.	Amounts of HCOH	Bacteria in HCOH Milk.	Acidity of Milk.	Remarks.
1.	6 Hrs	20000	1-500		15 deg.	
	24			2000	15	
	48			200	16	
	72			50	16	
	96			00	12	Milk sterile.
2.	6	9600	1-750		20	
	24			3500	16	
	48			700	17	
	72			60	15	
	96			10	14	Milk sterile.
6.	6	26000	1-1000		19	
	24			6000	15	
	48			300	15	
	72			100	15	
	96			40	15	Did not curdle.
7.	6	15000	1-1000		16	
	24			4600	17	
	48			500	17	Did not lopper
	72			60	15	in 30 days.
	96			0	15	
10.	6	10000	1-2500		14	
	24			7000	14	B.Subtilis Group
	48			1000	14	and Molds remain.
	72			125100	15	Did not lopper
	96			3200000	17	in 30 days.
12.	6	28000			22	B. Sub. Group and
	24		1-2500	1300	18	molds remain.
	48			600	16	Did not lopper
	72			75	16	in 30 days.
	96			Few	16	
14.	6	15000			17	B. Sub. and
	24		1-5000	1600	17	molds remain.
	48			700	15	Curdled in 15 days.
	72			10000	18	
	96			250000	20	
16.	6	26000			20	
	24		1-5000	700	20	Curdled in
	48			2000	17	18 days.
	72			12500	21	
	96			320000	20	
18.	6	35000			20	
	24		1-5000	2000	20	
	48			1200	19	Curdled in
	72			16500	21	12 days.
	96			255000	20	
	120			1400000	--	

Bacteria in Formaldehyde Milk. (Cont.)

Sample No.	Age.	Initial Bacterial Count.	Amounts of HCOH	Bacteria in HCOH Milk.	Acidity of Milk.	Remarks.
21.	6 hrs	15500			16 deg.	
	24		1-750	1800	17	
	48			7600	16	Curdled in
	72			10000	15	10 days.
	96			265000	17	
	120			46000000	20	
26.	6	22000			15	Curdled in
	24		1-7500	750	16	13 days.
	48			1200	16	
	72			1500000	20	
27.	6	31000			15	
	24		1-10000	6500	18	Curdled in
	48			37000	18	7 days.
	72			4630000	22	
	96			Innumer.	26	
30.	6	17500			15	
	24		1-10000	2400	15	Curdled in
	48			51000	18	13 days.
	72			2000000	18	
	96			10000000	20	
32.	6	9000			18	
	24		1-10000	3000	18	Curdled in
	48			16500	18	8 days.
	72			200000	18	
	96			26500000	20	
36.	6	17500			16	Curdled in
	24		1-10000	8500	17	
	48			56000	15	10 days.
	72			4800000	18	
	96			49000000	21	
37.	6	21000			15	Curdled in
	24		1-15000	950	15	6 days.
	48			31000	18	
	72			2500000	17	
	96			32000000	20	
38.	6	42000			14	
	24		1-15000	8000	15	Curdled in
	48			26000	17	4 days.
	72			1325000	16	
41.	6	10000		12000	15	
	24		1-15000	12000	16	Curdled in
	48			9000	15	7 days.
	72			960000	17	
	96			78000000	21	

(Continued on next page)

Bacteria in Formaldehyde Milk. (Cont.)

Sample No.	Age.	Initial Bacterial Count.	Amounts of HCOH	Bacteria in HCOH Milk.	Acidity of Milk.	Remarks.
42.	6	18500			17 deg.	
	24		1-15000	2000	18	Curdled in
	48			11800000	20	4 days
	72			Innumer.	--	
43.	6	25000	1-20000		16	Curdled in
	24			12000	16	3 days.
	48			3300000	21	
	72			-------	--	
46.	6	12200			15	Curdled in
	24		1-20000	15000	18	3 days.
	48			8000000	19	
48.	6	23000			17	
	24		1-20000	6300	18	Curdled in
	48			300000	19	4 days.
	72			166000000	22	
52.	6	12000			20	Curdled in
	24		1-50000	140000	21	3 days.
	48			2750000	26	
53.	6	30000			15	Curdled in
	24		1-50000	63000	16	3 days.
	48			13600000	22	
55.	6	18600			17	
	24		1-50000	17000	19	Curdled in
	48			3450000	22	3 days.
	72			Innumer	--	

From the preceding table it is concluded that formaldehyde in the proportion of 1-15000 is sufficient to inhibit bacterial growth for the necessary preservative period. This includes a generous factor of safety, for it is desired to keep the milk sweet under all conditions. If the milk was allowed to sour occasionally, the calves would be subjected to a sour milk diet, and digestive disorders would probably occur.

Formaldehyde in the proportion of 1-50000 had but little preservative effect, and the bacterial count was difficult after twenty-four hours. From bacterial transfers made, it was found that the group known as B. subtilis, a bacillus found universal on hay, and very resistent, and several kinds of molds remained when formaldehyde even as strong as 1-2500 was used. None of these, however, developed, and their presence in the inhibited growth condition, was probably not harmful. It was noted that the acidity of the milk increased but very little.

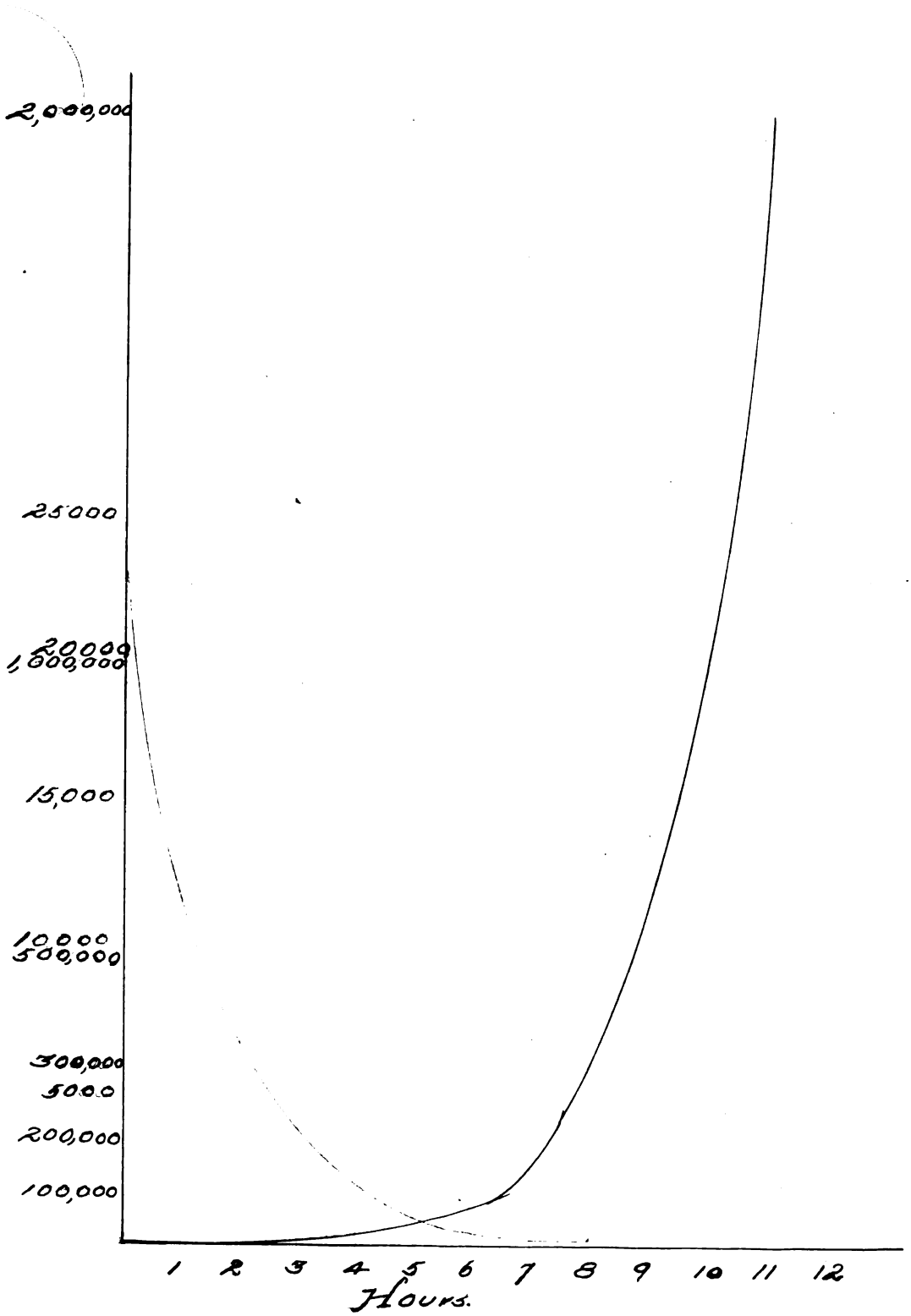
Work done later, in Dairy Bldg., with various amounts of milk with no reference to bacterial count, showed further that a proportion of 1-15000 was sufficient. The temperature there was fairly uniform at about 78 degrees F. It was found that the time for loppering varied within

quite wide limits even under identical conditions. This is shown very definitely in Dr. Marshall's work (Michigan Special Bulletin No. 33). He introduced a known number of bacteria into milk from day to day, and kept same at given temperatures, and the results show very irregular times for curding. In the present test, the milk was simply set aside and records kept of loppering. The following table gives data covering same:-

Formaldehyde	Limits of Time for loppering
1-500	Sterile
1-750	Sterile
1-1000	25 days
1-2000	20 to 30 days
1-3000	16 to 25
1-5000	7 to 20
1-7500	7 to 18
1-10000	5 to 15
1-12500	5 to 14
1-15000	3 to 9
1-20000	2 to 5
1-50000	1 to 3

A combination of the results shown in the two preceding tables show that bacteria behave differently, depending, apparently, on the individual samples of milk. It is evident that in milk treated with formaldehyde in the proportion of 1-5000 or more that there is a distinct decrease in the numbers of bacteria, especially during the first few hours. As the amount of formaldehyde is decreased, the falling off of bacteria is decreased, which is followed by a normal increase.

From the preceding data, the curve following was constructed, which will show at a glance the behavior of bacteria in milk treated with 1-5000 formaldehyde. In contrast, the other curve was made from data secured elsewhere, and this shows the action of bacteria in untreated milk. In the former case, skim milk was used, and in the latter, whole milk, but this should not affect results. It will be noted that the curves are just opposite in every particular. The most rapid decrease in the one case occurs during the first three hours, while in the other the most rapid increase occurs in the last few hours. In the case of the formaldehyde treated milk, the decrease shows the rapid germicidal power of this preservative, and also the fact that this strength of formaldehyde not only inhibits growth, but actually destroys. This strengthens the conclusion that a less amount is sufficient for practical work.



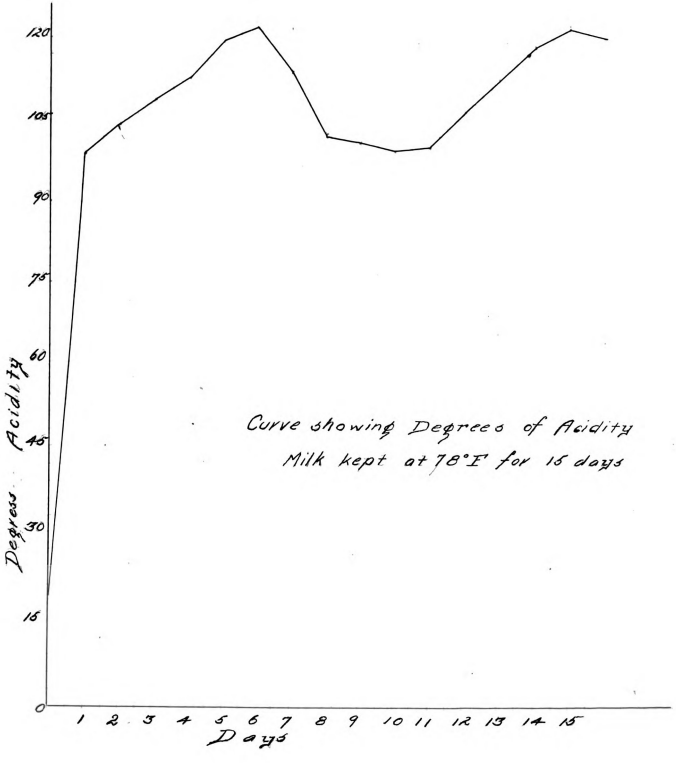
Red curve shows decrease of bacteria in formaldehyde milk (1-5000) during the first 8 hours at 78° F.

Black curve shows increase of bacteria in untreated milk at the same temperature.

Normal Souring of Milk.

During this investigation a careful study has been made of the action of the acid-forming bacteria. Several tests were made with sweet skim milk at a temperature of about 75 degrees F. to determine the effects these bacteria had upon the milk, and also upon themselves after rapid multiplication had taken place. In all twelve, samples were tested, and each sample was from a different day's milking. These were placed at 75 F. and the acidity taken each day for fifteen days. An average of the results are best shown by the curve plotted on the succeeding page.

This curve shows the rapid action of acid-forming bacteria during the first twenty-four hours under rather high temperatures. This is of value, for, in practice, the milk (skimmed) is placed in the hot sun and before it reaches the calf or pig, it is unfit for feed. Hence, the necessity of a preservative. Fresh milk has an acidity usually of about 15 degrees. Although the bacterial count may decrease during the first few hours due to a peculiar germicidal action in the milk, the acidity seldom decreases. Loppering is reached when the acidity is between 50 and 60 degrees acid. When the acidity reaches 90 degrees, the curd is solid. The normal action is shown on next page.



Curve showing Degrees of Acidity
Milk kept at 78°F for 15 days

Normal Souring of Milk.

Open Can on South Side of Brick Bldg.

Date.	Time	Temp. of Air.	Temp. of Milk.	Relative Humidity	Acidity of Milk.	Sky.
May 1.	6A.M.	48	47	71	58	
	12 N.	77	80			
	6P.M.	75	76	54	83	Clear.
2.	6A.M.	52	50	54	60	
	12 N.	78	81			
	6P.M.	78	78	49	74	Clear.
3.	6A.M.	59	57	64	35	
	12 N.	81	84			
	6P.M.	75	77	56	96	Clear.
4.	6A.M.	61	58	58	40	
	12 N.	77	80			
	6P.M.	76	77	44	93	Pt. Cloudy.
5.	6A.M.	65	62	58	56	
	12 N.	72	74			
	6P.M.	72	73	75	89	Cloudy.
6.	6A.M.	52	51	79	24	
	12 N.	65	68			
	6P.M.	58	63	54	82	Pt. Cloudy.
7.	6A.M.	35	34	71	25	
	12 N.	57	60			
	6P.M.	57	58	46	57	Clear.
8.	6A.M.	40	40	65	32	
	12 N.	60	62			
	6P.M.	61	62	70	85	Clear.
9.	6A.M.	41	40	67	25	
	12 N.	54	57			
	6P.M.	50	51	51	65	Clear.
10.	6A.M.	29	22	72	21	
	12 N.	50	53			
	6P.M.	51	52	47	46	Clear.

A comparison of the data in the foregoing table with that in table under "Feeding Operations" shows that there exists a wide variance in acidity under different conditions. Where milk was soured by heat from a radiator, the temperature was nearly constant, but when the souring took place in the open air beside a brick building, the temperature and atmospheric conditions cause great variance in the souring of the milk from day to day. On May 10th there was a heavy frost, and the acidity in the morning was only 21, while other mornings it had risen to 60 or more. Records show a variance in mornings milk from the 21 mentioned to 76 on later date.

This milk is, at the present time, being fed to an experimental lot of calves to determine the effects of sour milk. The results will finally be shown in the conclusion of this work.

Retardation of Souring.

In making a further study of the effects of formaldehyde in preserving skim milk, an experiment was made with different strengths of the preservative and at different temperatures. The data below shows the amounts of formaldehyde and the lengths of time those amounts will keep the milk sweep on a basis of acidity being reached at 30 degrees.

No. of Trials	Temp. F.	Formaldehyde. None	1-50000	1-20000	1-15000	1-10000
10	60	55 hrs	57 hrs	95 hrs	110 hrs	125 hrs
12	70	40	45	65	92	100
10	85	20	30	40	63	70

These results do not exactly check with work done previously and compiled in first table under laboratory data. This may be accounted for by the fact that the experiments were performed in different laboratories under very different atmospheric conditions. The results, however, being the average of several trials on different days may be taken as fairly accurate. A sliding scale of preservation by formaldehyde is thus established.

Are all Forms of Bacteria Affected Alike?

The growth of different bacteria differ greatly depending upon numerous conditions. It is not odd then that different bacteria should be affected differently by formaldehyde. In order to ascertain the types surviving the germicidal action of this preservative, considerable work was carried on with but few positive results. Laboratory conditions prevented exhaustive work, but it was determined that those bacteria usually resistent to heat are also resistent to the action of formaldehyde. One of the most resistent is the "hay bacillus" (B. Subtilis) which is found in quite large quantities in milk. This bacterium, however, develops more slowly in milk than other ones common to that media, and since it is desired only to inhibit growth, further consideration in unnecessary. Tests brought out the fact that molds are not as much affected by this preservative as are bacteria.

The effects on lactic bacteria have been shown in this work, but acidity is no real test as to the development of bacteria in milk. Lactic bacteria retard the development of many other forms, and it is possible that the inhibition of lactic growth might induce greater development of these other forms. However, the presence of formaldehyde also retards the growth of most of these other forms. It has been found

in the laboratory that the first few hours witnesses a decrease in total numbers of bacteria in milk. This is probably due to the dying off of bacterial forms that find milk a poor media in which to develop. The lactic bacteria are most vigorous in their development, and hence, usually bring about quicker changes in milk. Formaldehyde perfectly checks this organism and renders others less virile. In amounts excessive of 1-15000 there is an actual killing of the bacteria. B.Subtilis and molds are the most resistent.

Tubercule Organisms.

One of the objections to feeding skim milk from the creamery is the danger of infection by the tubercule organism from milk from infected herds supplying that creamery. Pasteurization has been used with good results, but this is costly, and unless great care is exercised, it is inefficient. It was thought that the addition of formaldehyde in a proportion of 1-15000 would be sufficient to kill tubercule organisms. After much study on the advisability of such a proceedure, an experiment was outlined. This necessitated the use of guinea pigs and several weeks time. At that time, the former could not be secured, so the tests were never made.

Letters to prominent scientists advise that, while no data is obtainable, it is probable that the

formaldehyde in the proportion fed would have but little effect on this very resistent organism. Dr. M. J. Rosenau of Harvard Medical School, is an authority on the behavior of tubercule bacteria, and he concludes his letter as follows: "Whether the small amounts of formaldehyde you use interferes with the virulence of the tubercule bacilli is doubtful, although I have no data upon the subject."

It seems probable, however, that the presence of formaldehyde in sufficient quantities to inhibit the growth of all bacteria would tend to induce a decrease in the virulency of this organism. Tubercule bacilli do not develop in milk as a media--a fact which probably accounts for the resistance to germicides. Formaldehyde has little effect upon fats, and if it is true that tubercule bacilli in spore form are surrounded by a film of fat, it is possible that this preservative could have no effect on the virulency.

The presence of this preservative, however, would in no way increase the virulency. At the same time, it is possible that the virulency might be greatly decreased by its presence and it would be a valuable aid in the control of this disease.

Rennet Coagulation.

Since rennet is so important a factor in the digestive processes, much time was spent in ascertaining the effects of formaldehyde in various strengths on the action of this agent. As usual in this work, skim milk was used. Checks were run continually so as to avoid errors due to changes in lots of milk, temperature, acidity, etc. The work was done at a temperature of 87 degrees F., and the formaldehyde was added just previous to the addition of rennet, in a strength of 1-1000. Time was recorded when solid coagulation--when all milk ceased to flow upon change of position--had taken place. The results are given in the table on the following page.

Effects of Formaldehyde on Rennet Coagulation.

Time for coagulation at 87 degrees F.

No. Trials	Solution of Formaldehyde	Time for Check.	Time for Form.	Remarks.
8	1-100	5 min.	Not in 21 days	
10	1-200	5 "	Not in 21 days	
15	1-500	5 "	Not in 21 days	
11	1-1000	5½ "	18 min.	Very solid.
20	1-1250	5 "	13 "	
10	1-2500	5 "	7 "	Solid.
16	1-3000	5 "	5 "	No action.
15	1-2500	7 "	7 "	70 deg. F.
20	1-3000	7 "	7 "	70 deg. F.
10	1-5000	5 "	5 "	
17	1-10000	5 "	5 "	
12	1-12500	4½ "	4½ "	Older milk
20	1-15000	5 "	5 "	
5	1-25000	6 "	6 "	Temp. Varied.
5	1-50000	5 "	5 "	

The data above shows that formaldehyde in milk has no ill effects on coagulation by rennet until a strength of 1-2500 is used. It will be noted that the line of demarkation between effects and no effects is very clearly drawn. In the proportion of 1-500 there was no coagulation even after 21 days, but when twice that small amount was added, there was a decided retardation in time of

the action of this ferment. In all cases of coagulation, the curd was as solid as that in the cases of the untreated checks. In the proportion of 1-2500 there was still a retardation, but at 1-3000 there was no effect produced. On the proportion used in actual practice a great many tests were made, and there was, in no case, a retardation of coagulation. This was true as the amount of formaldehyde was decreased. It was then condluded that formaldehyde did not retard coagulation of milk by rennet until a strength of 1-2500 had been reached.

Coagulstion by Pepsin.

A test similar to the above was made with the digestive agent, pepsin. Sufficient of this ferment was used to produce normal coagulation in about ten minutes with untreated milk. The same amount was used each time so a direct comparison could be made. The results may be found on the page following.

Effects of Formaldehyde on Pepsin Coagulation.

Time for coagulation at 87 degrees F.

No. Trials	Solution of Formaldehyde	Time for Check	Time for Form	Remarks.
8	1-100	10 min.	Did not coag.	
15	1-250	10 "	Did not coag.	
12	1-500	10 "	Did not coag.	
10	1-1000	11 "	20 min.	Solid.
15	1-1500	10 "	16 "	
20	1-2000	10 "	10½ "	Little action.
10	1-2500	10 "	10 "	
5	1-5000	10 "	10 "	
5	1-7500	11 "	11 "	Varied Temp.
14	1-10000	10 "	10 "	
20	1-15000	10 "	10 "	
5	1-25000	10 "	10 "	
5	1-50000	10 "	10 "	

From the above data it is evident that formaldehyde in the amount fed in our practical experiment has no effect on coagulation of milk by pepsin. It is therefore justifiable to believe that it would in no way interfere with the action of this ferment. Not until a strength of 1-2000 was reached was there any evidence of retardation of the action of this digestive factor. The curd was solid and apparently did not differ from that of the checks.

The Disappearing Action of Formaldehyde.

Hydrogen peroxide has been suggested as a preservative in milk because the addition of heat will entirely remove it. When left exposed to the air, formalin will rapidly lose strength, but my experiments have shown that even in closed vessels, that formaldehyde disappears from milk. This may clearly be shown by studying the former tables showing the inhibiting power of this preservative for a few hours followed by a rapid increase in bacterial development. Just what becomes of this has not been fully determined.

It is known that certain liquids have a "binding" capacity for the constituents of other liquids or substances. For instance, it would be expected that the addition of NAOH to blood would make the resulting liquid more alkaline, but such is not the case when small quantities are added. Certain elements of the blood "bind" the NAOH and it has no chance to function. The same may be true of formaldehyde in milk. On the other hand, it may be used up in the killing of bacteria, and it stays in the bodies of the organisms. Corrosive sublimate, in known amounts, added to an infected liquid shows a less quantity when filtered out later. The difference was probably used up in the killing of bacteria.

In the study of this disappearing action, a

powerful detector was used. It is known as Denige's Reagent, and it can detect the presence of formaldehyde in a solution of 1-500000. While not a quantitive test, by the rapidity of the detecting color, various quantities may be approximeted. The following table gives the results of milk treated with 1-15000 HCOH.

Disappearing Action of Formaldehyde.

Color Test by Deniges Reagent.

Trial No.	0 hrs.	Acidity	24 hrs.	Acidity	48 hrs.	Acidity	No Color at	Acidity.
1	2 min.	15 deg.	3 min.	15 deg.	7 min.	17 deg.	60 hrs.	90
2	2	14	3	15	6	16	60	81
3	2	15	3.5	16	7	18	62	75
4	2	16	3	18	8	21	58	96
5	2	15	2.5	16	9	20	65	94
6	2	16	3	19	6	17	70	86
7	2	14	4	18	7	25	55	96
8	2	15	3	15	6	16	70	87
9	2	16	3	17	7	18	65	90
10	2	15	3	16	6	18	70	88
15	2.5	18	4	17	6	20	65	97

Presence of Formaldehyde in Faeces and Urine.

An effort was made to determine the channels of disappearance in the animals themselves. By the use of Denige's Reagent, it was hoped to ascertain if the preservative was present in the excreta. Urine was obtained from calves fed formaldehyde milk in the experiment and also from calves to which only three feeds of it had been given. In the case of the latter, sufficient time was given to allow for natural excretion. In neither case was formaldehyde detected even in slight quantities. By adding a known amount to the urine, its presence could be detected in amounts as small as 1-500000; hence, it was concluded that the preservative did not pass from the body in the urine. In the case of the faeces, a scant water solution was made and treated with the detector, but without results.

From these numerous tests, it was concluded that formaldehyde disappears in the milk itself, or at least, completely changes its form and becomes less active as time increases.

Effects of Formaldehyde on Nitrogen in Skim Milk.

Four 500 gram samples of the skim milk fed to the calves were taken and treated as follows:-

1. Raw skim milk untreated.
2. Raw skim milk treated with 1 gram pepsin.
3. Raw skim milk treated with 1-1000 pepsin and HCOH 1-14000.
4. Raw skim milk treated with 1-1000 pepsin and HCOH 1-2500.

The raw skim milk sample was used as a check. No. 2 was merely treated with pepsin and so acted as a second check. No. 3 was treated with the pepsin and then HCOH (1 C.C. 40% HCOH was diluted to 100 c.c. with distilled H_2O and 9.4 c.c. of the diluted solution was added to No. 3.) No. 4 was treated with pepsin, then HCOH, 1-2500. (1 c.c. of 40% HCOH was added to the 500 c.c. of milk).

Analysis.

1. Casein (unchanged)

   Acetic Acid precipitate.

2. Caseoses and peptones

   Precipitate of Acetic Acid filtrate with $CuSO_4$.

3. Amino Acids and Ammonia

   Filtrate.

As the nitrogen is the most likely element in the makeup of these substances to be changed, the nitrogen was determined in the various samples as outlined above.

The milk was slightly acid when worked with, and the work was all duplicated.

First, 5 gram samples of 1,2,3, and 4 were taken, and the Kjeldahl determination made with them. This amount was taken as the 100% content of N. for later work. Then a 100 gram sample of each was taken and the Casein in each precipitated with 12.5 c.c.of 6% Acetic Acid, then filtered. The Kjeldahl was run upon the precipitate to determine the N. content of the Casein. The filtrate was halved and Kjeldahl run upon each half. To one half,a dilute solution of CuSO was added until the precipitate was all thrown down, and then 10 c.c. excess was added. The solution was filtered and the Kjeldahl run upon both filtrate and precipitate, giving the N. content in both Nos. 2 and 3. The following table shows changes in the nitrogen forms.

Nitrogen Content.

	Casein	Caseoses	Amino
Skim milk	82%	11.7%	6.3%
" " plus Pepsin	78.3	5.9	15.8
" " " " plus HCOH 1-14000	74.4	8.4	17.2
" " " " " HCOH 1-2500	66.6	6.2	27.2

To make these results more positive, many more samples should be run containing various amounts of HCOH.

It will be noted that the nitrogen in the form of casein and caseoses is decreased when pepsin is added to the skim milk. There is a corresponding increase in nitrogen in the form of amino acids. This decrease and increase continues in a respective order

when formaldehyde is added in increasing amounts. The sum of the difference between the nitrogen in the casein and caseoses in untreated skim milk, and the nitrogen in the casein and caseoses in the treated milk is equal to the difference of the nitrogen in the amino acids in the same cases. For instance: (82 — 74.4 = 7.6) plus (11.7 — 6.4 = 3.3) = 10.9. In the same case 17.2 — 6.3 = 10.9 This uniformity holds true in all of the cases, and shows the exactness of the chemical changes.

Since nitrogen in the form of amino acids is soluble, it is probable that it is even more digestible than in the original form. However, amino acids are found in the by-products of digestion, which might offset the increased digestibility of this form. At least, it cannot be concluded that the addition of HCOH makes the nitrogen less digestible, or in any way depreciates the value of nitrogen in the food. The addition of HCOH simply changes the form of the nitrogen in the milk.

Practical Experiment.

In a study of this nature the results of artificial experiments are not as valuable as those taken from actual feeding records of young animals, for there are certain conditions occuring in animal digestion which cannot be duplicated in artificial processes. In order to ascertain actual accumulative effects, both of formaldehyde-treated and sour milk, a feeding experiment was carried on with young calves. Owing to the extreme scarcity of calves in the vicinity of the college, only a very limited number could be obtained, and for the same reason, it was necessary to use dairy stock of three different breeds. Such a combination was far from ideal, but the results are of interest.

These calves, twelve in number, were divided into lots of three each. Division was made in reference to age, breed, appetite, sex, general condition and weight, and care was exercised that the lots were of equal growing capacity.

Lot I was fed on sweet milk as a control lot. Lot II was fed on skim milk allowed to sour, and Lot III was fed on milk kept sweet by the addition of formaldehyde. In lot II was placed a small Guernsey of delicate health and appetite, but this deficiency in this lot was offset by the presence of two calves larger and stronger than the average. In lot III was a fine Holstein grade to balance the expected lesser gains from two rather small Guernseys. During

the first week of the experiment, this calf suddenly developed pneumonia, and died at the end of the week. An autopsy by Dr. Hallman of the Michigan Station showed disease in the respiratory organs only. A complete examination of the digestive tract showed nothing abnormal, and absolutely no irritation. His report (Autopsy 61) shows bronchial pneumonia alone as the ailment. During the next week, the small Guernsey in Lot II began to show effects of the sour milk and soon died. This reduced these two lots to three calves each, so a young Brown Swiss of considerable breeding value was taken from Lot I, and the experiment was completed with the remaining three in a lot. The removal, however, of the best calf in Lot III and the smallest in Lot II gave the latter lot a decided advantage. Another grade Holstein calf was secured, and fed formaldehyde treated milk, but its gains are recorded, not in Lot III, but separately, as it was fed a different amount of the preservative than Lot III. The table following gives sufficient data relative to the lots and the individuals in each lot.

Individual Characteristics.

No	Milk	Age	Breed	Sex	Weight	Condition	Remarks.
45	Sweet	38 days	Holstein	F.	133	Fine	Good size
46	"	36 "	Jersey	F.	101	Good	Spare
47	"	24 "	Hol.Grde.	F.	98.3	Good	Fair size
	Average	33 "			110.6		
48	Sour	42 "	Hol.Grde.	M.	123.6	Fine	Large
49	"	38 "	Guer. "	M.	111	Fine	Large
54	"	35 "	Hol.Grde.	F.	97.3	Good	Fair size
	Average	38 "			110.6		
50	Form.	40 "	Guer.Grde	M.	114.3	Good	Small
52	"	40 "	Hol.Grde.	M.	114.3	Fine	Fair size
53	"	27 "	Guer.Grde	M.	99	Good	Fair size
	Average	35 "			107.6		
Extra on Formaldehyde.							
51		25 "	Hol.Grd.	M.	82	Good	Good size.

Lot I.

This picture of Lot I., fed on sweet skim milk was taken at beginning of experiment. The Brown Swiss, in the corner, was discarded, and the three facing finished the experiment. From left to right, they are:

No. 45 No. 47. No. 46.

Lot II.

A picture of Lot II. at the beginning of the experiment. The little Guernsey, nearest the door, died from the effects of the sour milk, apparently, and the experiment was completed with the remaining three.

The Holstein in front is No. 54, the Holstein in the rear is No. 48, and the Guernsey between them is No. 49.

Lot III.

Lot III. at the beginning of the experiment. The Holstein facing died of bronchial pneumonia, and the experiment was completed with the remaining three.

The Guernsey facing is No. 53, the Guernsey in the corner is No. 50, and the remaining Holstein is No. 52.

Preparation of Milk.

Experiments in the bacteriological laboratory, detailed later, were carried on to determine the amount of formaldehyde necessary to keep the milk sweet a sufficient length of time under summer (artificial) conditions. Through an error in computation, the amount was thought to be.02%, and for the first eight feeds this amount was fed. This is nearly three times more than is necessary, and as soon as the error was discovered, the correct amount, which is .0075% formaldehyde, was used.

Skim milk from the separator at 10:00 A.M. was cooled and saved for the next day's feed. At night two cans were prepared from this and set by the radiator where an almost constant temperature of about 74 degrees F. was maintained. Experiment started on Feb. 1, 1913, and was carried on for twelve weeks. The milk in one can contained formaldehyde in the proportion stated above, which was sufficient to keep the milk sweet under these conditions. The milk in the other can was untreated, and it became sour and almost invariably loppered heavily. The remainder of the milk was kept cool, and in the morning it received treatment as described immediately preceding.

Condition of Sour Milk.

The sour milk fed calves in Lot II. was secured by placing the receptacle beside a radiator, where a fairly constant temperature was maintained. The following table contains data taken at random, which will suggest the average condition of the milk when fed.

Date		Temp. of Milk.	Acidity.	Remarks.
	A.M.	80 F.	93 deg.	Heavily curded.
Feb. 5	P.M.	76 F.	81	Lightly "
	A.M.	77	96	Beginning to curd.
" 12.	P.M.	83	96	Heavily curded.
	A.M.	81	73	Lightly curded.
Mar. 6	P.M.	82	96	Heavily "
	A.M.	79	87	Heavily curded.
" 10	P.M.	73	40	No curd.
	A.M.	83	92	Heavily curded.
Apr. 3	P.M.	82	96	" "
	A.M.	82	93	" "
Apr. 15	P.M.	80	94	" "
	A.M.	79	96	" "
Apr. 16	P.M.	85	90	" "

When fed, this milk was warmed to body temperature, as was the other milk, and stirred until curd was well broken.

A grain ration made up of

3 parts corn flake,
3 " whole oats,
1 " bran, and
1 " oil cake,

was fed in amounts varying with the appetites of the incividuals. Bright alfalfa hay was fed in limited amounts as shown in feeding tables. Each lot was fed all the milk that it would consume readily, so as to exaggerate, if possible, the effects of the different lots of milk. Records of weights of feeds and gains were carefully kept, and notes taken as to the general condition of each animal. The entire work of preparation of feeds, feedings, care, etc., was done by the author in order that, in drawing conclusions, no detail might be overlooked. During the first three weeks too much milk was fed, and

scours resulted. As a further result, small gains were made. Upon reducing the amount of milk, the calves immediately responded with normal gains. Further notes may be found succeeding the feeding tables.

The following tables give in detail, the amounts of different feeds consumed per week by each individual in the three lots. They also include the same for the extra calf fed formaldehyde-treated milk, but not included in Lot III.

Milk Fed per Week in Pounds.

No:	Week: 2	3	4	5	6	7	8	9	10	11	12	Total	
Lot I. on Sweet Milk													
45:	92.5:	112:	115:	125:	126:	125:	126:	126:	126:	126:	141:	140:	1480
46:	92 :	109:	111:	109:	100:	94:	95	107	112:	112:	129:	126:	1300
47:	90 :	106:	112:	112:	110:	105:	112:	112:	112:	112:	126:	127:	1336
Lot II.	on Sour Milk										Average		1372#
48:	95 :	109:	117:	126:	119:	119:	126:	126:	126:	126:	140:	141:	1467
49:	94 :	109:	120:	119:	121:	118:	126:	126:	126:	126:	140:	140:	1465
54:	85 :	106:	113:	112:	110:	100:	98:	107:	120:	126:	126:	127:	1330
Lot III.	on Formaldehyde Milk.										Average		1421#
52:	78 :	102:	111:	98:	102:	87:	98:	98:	106:	112:	114:	126:	1232
50:	85 :	109:	112:	112:	110:	94:	98:	98:	98:	98:	112:	114:	1242
53:	83 :	106:	111:	83:	97:	74:	84:	98:	106:	112:	104:	104:	1166
Extra Calf on Formaldehyde Milk.											Average		1213#
51:	85 :	85:	84:	83:	87:	88:	93:	98:	98:	99:		900	

Grain Fed per Week in Ounces.

No	Week: 2	3	4	5	6	7	8	9	10	11	12	Total	
On Sweet Milk Lot I.													
45	18	30	31	112	112	140	140	154	175	140	175	1227oz	-77#
46	14	24	25	84	84	84	112	140	140	140	140	987	"-62#
47	14	24	25	84	84	112	119	126	140	140	140	1009	"-63#
											Average		67#
On Sour Milk Lot II.													
48	20	37	37	112	112	112	119	140	168	175	175	1207	"-75#
49	16	30	30	112	112	112	126	140	168	161	168	1177	"-73#
54	6	10	10	98	98	112	112	119	168	140	168	1041	"-65#
											Average		71#
On Formaldehyde Milk Lot III.													
50			40	42	35	70	35	42	49	84	105	502	"-31#
52			30	70	70	56	70	91	70	84	112	653	"-41#
53				70	70	49	70	84	98	84	105	630	"-39#
											Average		37#
Extra Calf on Formaldehyde Milk.													
51			7	70	70	49	56	70	56	105	112	595	"-37#
						10 weeks feeding.					Average		37#

Hay Fed per Week. (In ounces).

No.	Week 2	3	4	5	6	7	8	9	10	11	12	Total Oz.	Pounds
Lot I. on Sweet Milk.													
45	40	84	84	112	112	112	140	168	168	168	168	1384	86
46	40	70	70	84	84	112	140	168	168	168	168	1272	80
47		56	56	112	112	112	140	168	168	168	168	1280	80
										Average			82
Lot II. on Sour Milk.													
48	30	70	70	112	112	112	140	168	168	168	168	1338	83
49	40	70	70	112	112	112	140	168	168	168	168	1346	84
54	40	70	70	112	112	98	140	168	168	168	168	1334	83
										Average			83
Lot III. on Formaldehyde Milk													
50			56	70	70	98	140	70	105	140	140	889	55
52			56	70	70	70	140	140	140	140	168	994	62
53				70	70	70	140	140	140	140	168	938	58
										Average			58
Extra Calf on Formaldehyde Milk													
51			48	70	70	98	112	168	168	140	168	1042	65
						10 weeks feeding Average:							65

From the foregoing tables, it is noted that the calves in Lot II consumed the greatest amount of feed, while those in Lot III consumed only 85% milk, 52% grain, and 70% as much hay as Lot II. In consumption of milk, Lots I and II show greater uniformity in amounts. Notes taken, especially during early part of the experiment, show that the appetites of these lots were much better than Lot III. The extra calf on formaldehyde treated milk consumed a uniform amount from week to week, gradually increasing as the animal grew. In the consumption of grain, Lot III did not begin as early to consume as much as the other two lots, and also, this lot was less uniform in amounts eaten. The same was true in the case of hay eaten. Because of conclusions drawn later, special attention is called to the fact that the Extra calf, No. 51, consumed as much as any other individual in the experiment.

Notes on Feeding, etc.

As has been stated previously, about three times the correct amount of formaldehyde was fed to Lot III during the first eight feeds.

Formaldehyde was not added to the ration gradually, but the total amount in error was placed in first feed.

In the case of Calf No. 51, the whole milk diet was changed by the gradual addition of formaldehyde-treated skim milk at the rate of one half pound per

day. In this way there was not an abrupt addition of the preservative to the diet.

Lots I and II showed better and more uniform appetites than Lot III.

Calf No. 45 in Lot I was much more vigorous than the other two in that Lot. Calf No. 46 of the same lot was troubled with severe attacks of scours, and did not look well although it gained steadily.

Calves Nos. 48 and 49 in Lot II were very vigorous and healthy. These two experienced but little scouring, while Calf 54 in this lot had several attacks.

Calves Nos. 50 and 53 in Lot III scoured much during preliminary feeding period, but were troubled but little afterwards. These two were fed too much milk during first three weeks of the experiment and as a result, did not appear in vigorous condition. No. 52 of the same lot did much better, although it was "off feed" several times.

During preliminary feeding period, the calves were cared for in a rather dark pen, and during this time, became infested with lice. They were treated with tobacco dust, which resulted in the complete extermination of the lice by the end of the third week.

The calves in all the lots scoured some, probably due to overfeeding, but Lot III was less troubled.

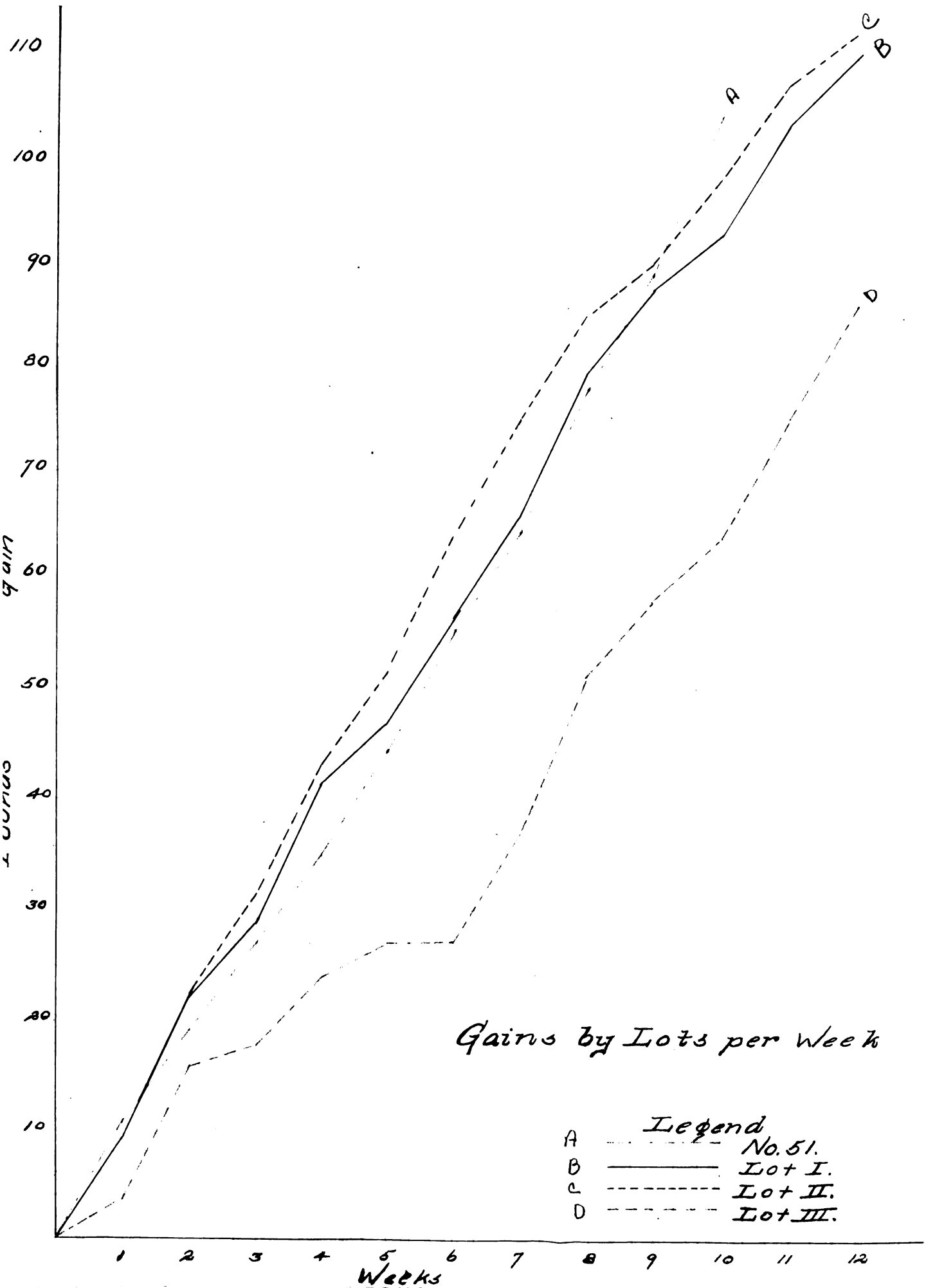
The calves in Lot II were of more sleek appearance, due to greater lustre of the hair.

Table Showing Gains of Individuals by Weeks.

No	Week: 2	3	4	5	6	7	8	9	10	11	12	Total	Average
Lot: I. on Sweet Milk.													
45	12#	15#	11#	16#	14#	11#	14#	14#	8#	7#	7#	134#	11.1#
46	9	8	5	13	-4	12	6	14	8	6	10	7	94	7.9
47	7	15	4	8	6	6	8	14	9	8	9	6	100	8.3
									Average:				109	9.1
Lot: II. on Sour Milk.														
48	12	14	8	11	12	11	13	10	0	10	10	5	116	9.6
49	5	16	9	9	6	14	10	9	9	8	10	6	111	9.2
54	11	9	11	14	8	12	11	11	6	6	7	3	110	9.2
									Average				112	9.3
Lot III on Formaldehyde Milk.														
50	1	15	2	10	0	-1	6	14	4	9	11	11	82	6.7
52	2	6	6	7	4	4	12	13	12	0	15	11	92	7.6
53	8	15	-2	1	5	-2	11	15	6	8	9	11	86	7.3
									Average:				87	7.3
Extra Calf on Formaldehyde Milk:														
51			11	8	9	8	9	11	9	14	11	15	105	10.5
							Ten weeks only							

A study of the preceding table shows that Lot II. made the greatest gains, and, also, the most uniform gains from week to week. The total gains were not excessive enough over those of Lot I. to justify the conclusion that sour milk is a better feed than sweet milk, but the gains were more uniform both by weeks and individuals. In Lot I, there was quite a variance in gains. Calf No. 46 was very irregular, and Calf 49, which made great gains at first, did not do so well during latter part of experiment. The cause for this is not known. In Lot III there was a great variance of gains, and the total was not as great as those of other lots. However, it will be noticed that the gains were greater and more uniform toward end of feeding period. The extra calf on formaldehyde, with the exception of Calf No. 49, gained more uniformly and a greater total than any other in the test. This suggests that the cause for the lesser gains in Lot III was due to the amount and manner of introducing the formaldehyde into the ration.

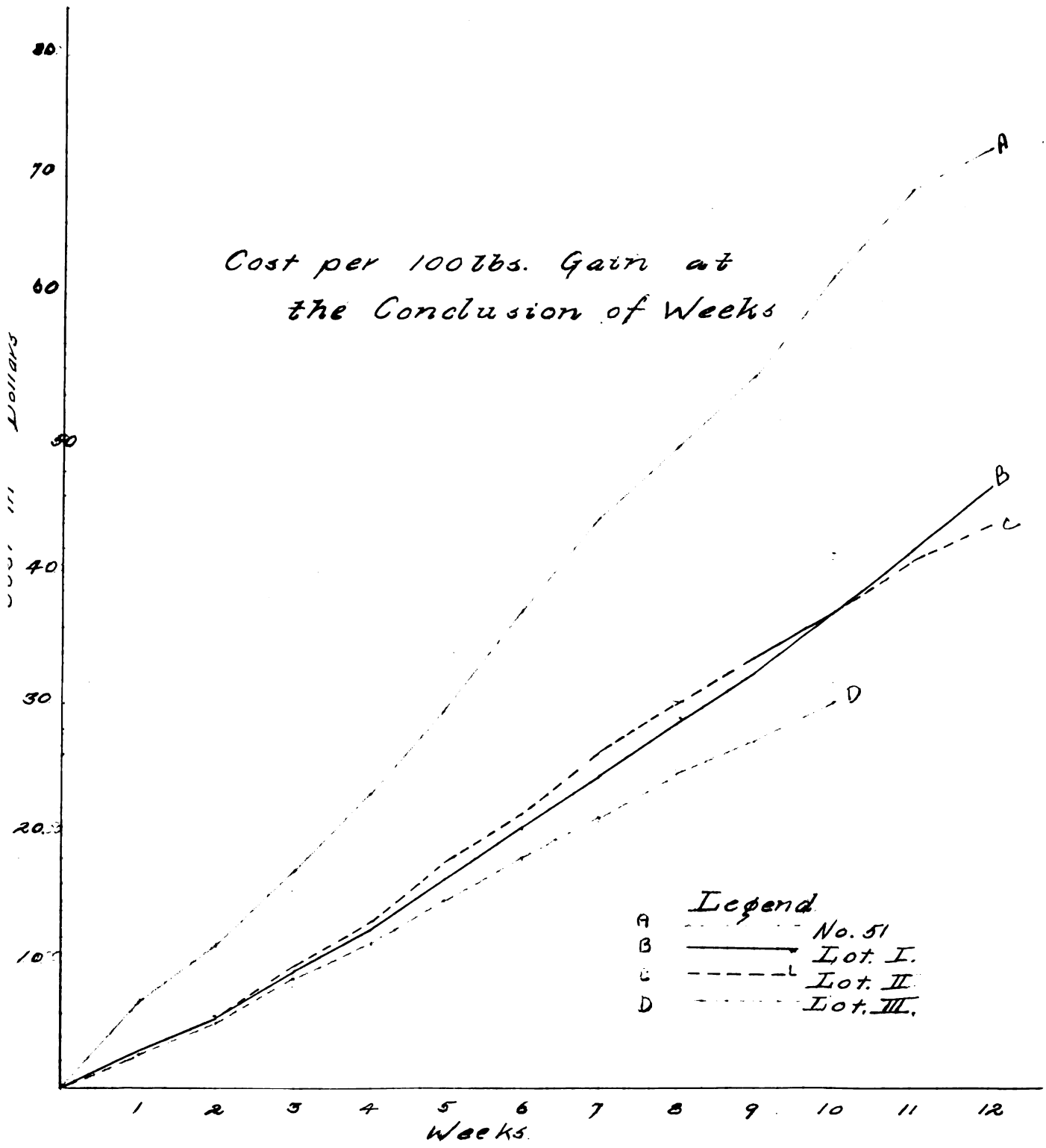
On the following page may be found curves showing the gains per week by lots. The curve for the gains of Calf No. 51 starts with the bthers, but is concluded at the end of the tenth week. In this way a comparison may be made.


110
100
90
80
70
60
50
40
30
20
10
Gain
A
B
C
D
1
2
3
4
5
6
7
8
9
10
11
12
Weeks
Gains by Lots per Week
Legend
A ..... No. 51.
B ——— Lot + I.
C - - - - - Lot + II.
D - - - - - Lot + III.

Gains by Weeks.

On the preceding page is shown graphically the gains made by the different lots during the twelve weeks of the experiment. Lot III shows a decided decrease in gains during the 3rd, 4th, 5th and 6th weeks. During this time it was decided that too much milk was being fed, and a lessening of the amount was probably accountable for the satisfactory gains made following this period. The other lots made very good gains and the curve shows these gains to be very uniform. Lot II. on sour milk, kept ahead of Lot I in gains, and finished in that order. The single calf on formaldehyde milk made gains in the total excessive of the other lots. The gains were very uniform, and toward the close of experiment this calf was doing better than ever before. The same may be said of the calves in Lot III, for their curve of gains begins to rise during the latter end of the test, and were it not for the bad weeks mentioned, their gains would equal the other lots. All of the calves gained an average exceeding a pound a day, and Calf No. 51 gained an average of exactly one and one-half pounds per day.

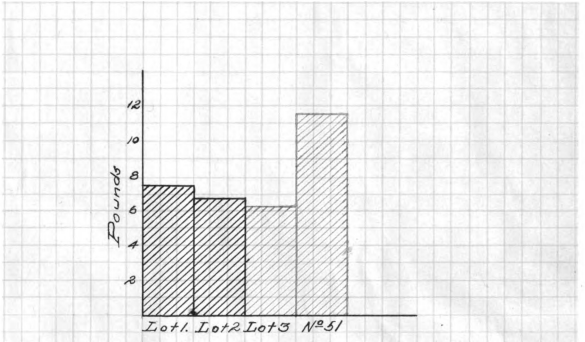
While the foregoing figures show the total amounts of gain, further computations show that the calves in Lot III, on a basis of economical gains, made more favorable returns. On the following page is shown the total cost of gains.


Cost per 100 lbs. Gain at the Conclusion of Weeks
Cost in Dollars
80
70
60
50
40
30
20
10
A
B
C
D
Legend
A No. 51
B Lot. I.
C Lot. II
D Lot. III.
1 2 3 4 5 6 7 8 9 10 11 12
Weeks

Some rather interesting facts are shown by the curves on the preceding page. These curves represent the cost of one hundred pounds gain total at the conclusion of each week. In the cases of lots producing normal growth, there is a gradual incline upward of the curve showing that as the animal increases in age, the cost per unit gain is also increased. This was less marked in the case of the single individual on the formaldehyde milk. Lot III on formaldehyde milk showed a great variation of cost from week to week. This suggests that the disturbing causes, whatever they were, acted more or less uniformly, or the gains would not vary so greatly in cost by weeks. During weeks 5 and 6, there was a rapid increase which was followed by a similar decrease during the next two weeks. This left the cost at the same figure it held one month before. It will be noted, however, that the curve for this lot begins to drop toward end of feeding period, while the others are increasing in cost. Lots I and II are very similar, but both are excessive of Calf No. 51, the gains of which were very uniform in cost.

Since all of the calves were fed more milk than would balance well in the ration because it was desired to exaggerate the effects of the different lots of milk, the gains per hundred pounds

of milk consumed have been computed. In studying the following graphic chart, it will be well to bear in mind the fact that Lots I and II ate more grain and hay in proportion to milk consumed, than did Lot III.

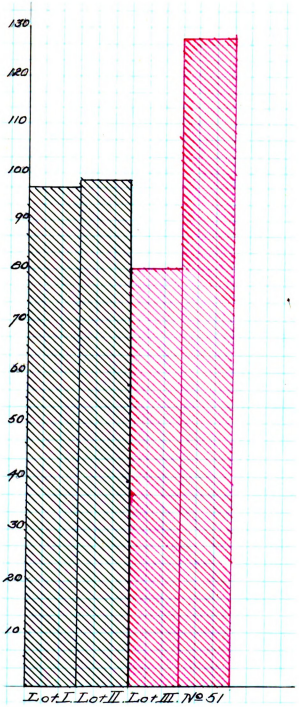


Graph showing gains per 100 lbs. milk fed.

It may be readily seen that the gains per hundred pounds of milk by lots are quite different in amounts. Lot III is least with an average of about 7.6#, while Lot II exceeds it but little with an average of 7.9#. The lot on sweet milk made an average of 8.5#, but this is much below gains made by

Calf #1 on formaldehyde. This would lead us to suspect that this calf was an extra good one in growth ability. Since this is a single individual, too much value should not be attached to the much better gains by an animal in comparison with the lots of three each.

On the following page may be found a graphic chart setting forth the per centage gains, of the gains per 100# original live weight. In this chart, we find that Lots I and II have changed from the positions they occupied in the chart just preceding, while the lots on formaldehyde remain as before--one showing much lesser gains, and the other much greater gains than Lots I and II. It will be noted that in this, as in preceding curves, etc., these two latter lots were very similar in their actions, while the lots on formaldehyde occupy the extreme positions. However, since percentage gains increase normally with age, the gains for Calf No. #1 are under a handicap because the test on this calf extended over a period of ten weeks instead of twelve as was the case with the other lots.



Graph showing gains per 100 lbs. live weight.

Conclusion of Experiment.

At the end of the experiment, all of the calves were looking vigorous and healthy. Calf No. 46, on sweet milk, and Calf No. 50, on formaldehyde milk, showed the least vigor. The calves on sour milk looked in better condition as a lot, but Calf No. 45, on sweet milk, was in much better condition than any other in the experiment. The following pictures were taken at close of tests.

Lot I. on sweet milk at close of feeding period. No 46 (Jersey at right) made the poorest gains, and was in poorest shape at close of test.

Lot II. on Sour Milk.

This picture shows calves fed sour milk at close of feeding period. They were the sleekest looking lot in experiment. The lustre of the hair was very marked, and they were in good growing condition.

Lot III. on Formaldehyde Milk.

This picture shows the calves fed milk treated with formaldehyde. The Holstein directly in the foreground is the extra Calf No. 51, also fed formaldehyde milk. These calves were in good condition, and, at the conclusion of test, were making good gains.

Post Mortem Examination.

At the close of the tests six of the calves were slaughtered and a careful examination made by Dr. Ward Giltner of the Michigan Station. His notes taken at this time follow:-

Calf No. 48 on sour milk. Contents of abomasom soft; no curd present; no excessive acid odor; mesenteric glands very large--abundant lymph exudation from cut surface. Rumen distended.

Calf No. 49 on sour milk. Contents of abomasom soft; few small granules of curd; mesenteric glands very large--abundant lymph exudation from cut suffface. Rumen distended and mucous membrane pink.

Calf No. 51 on formaldehyde milk. Contents of abomasom not abundant; large leather-like curd larger than man's hand, honey-combed with cavities. Very marked acid odor; mucous membrane pale, mesenteric glands small and firm with no lymph esudation from cut surface. Rumen only about half size Calves 48 and 49.

Calf No. 52 on formaldehyde milk. Only about one-half curd as No. 51, but having same consistency. Acid odor marked; mucous membrane pale; mesenteric glands about one-half size those of Calves 48 and 49.

Calf No. 50 on formaldehyde milk. Contents

of stomach soft; milk curd smaller than 52; acid odor marked, but less than No. 51. Mesenteric glands slightly larger than Nos. 51 and 52. Slight amount of lymph exudation from cut surface. Rumen mid-way in size between Nos. 46 and 51; mucous membrane pale.

Calf No. 46 on sweet milk. Small pieces of curd present; acid odor not marked; stomach nearly empty; mucous membrane pale; mesenteric glands small and firm.

Calf No. 53 on formaldehyde milk. Contents of stomach liquid; odor strongly acid; curd in large lumps and spongy; tough curd occurs in large lumps and honey-combed; mucous membrane pale and mesenteric glands small, relatively, but contained more lymph."

A study of this report shows that the calves fed sour milk and the single one fed sweet milk had much more rumen capacity. This suggests better appetites, as was actually the case. In the case of the calves fed formaldehyde, there was present in the digestive tract a strong acid odor that was almost completely absent in the other animals. In Calf No. 51, the extra calf on formaldehyde, there was a large leather-like curd, which shows that perfect digestion was not taking place. The sour

milk calves showed a pink (normal) mucous membrane, while the others fed formaldehyde appeared as pale. The mesenteric glands have at least two important functions, viz., to act as a filter for bacteria etc., and to produce white corpuscles. When an examination is made of these glands after animals are slaughtered, these glands may be taken as "barometers" of the condition of the animal. If the glands are small and firm, it is evidence that the animal has been poorly nourished. In the case of large glands, and when cut there is abundant exudation from the cut surface, it is evidence that the animal has been well nourished and is in good condition.

From the above evidence it may be concluded that the calves fed formaldehyde were not as well nourished--a fact already brought out by their lesser gains. The condition of the curd suggests that digestion was poor while the paleness of the mucous membrane indicates an inefficient functioning of the glands therein. The size of the rumen shows lack of digestive capacity in the calves fed formaldehyde milk, and the strongly acid odor suggests a retardation of the functions of the digestive enzymes.

Summary of Data.

Before drawing conclusions from the preceding data, it is thought best to group them in the table following:-

	Lot 1.	Lot 2.	Lot 3.	No. 51
Milk consumed	1372#	1421#	1213#	900#
Grain consumed	202#	213#	111#	37#
Hay consumed	246#	249#	174#	69#
Total gain	326#	337#	260#	105#
Average gain per calf per week	9.1#	9.3#	7.2#	10.5#
Percentage gains	97	96	80	120
Gain per 100# milk	8.5#	7.9#	7.6#	11.6#
Cost per hundred pounds gain.	$4.86	$4.95	$5.62	$3.35

*Note that Calf No. 51 was fed ten weeks.

Conclusions from Experiment.

So many factors have influenced the gathering of reliable data that few positive conclusions can be drawn from this experiment. Chief among these factors were variations of breeds, age, sex, and the necessary limited number of individuals in the various lots. Also the removal of one calf from each lot after a careful division had been made, gave advantages to certain lots.

After a careful study of the practical side of this experiment, a conclusion that formaldehyde, as fed, had no ill effects on growth would hardly be justifiable. The lesser gains, the poorer appetites, the smaller rumens, the condition of curd and digestive tracts, and the poorer condition of the animals at end of the feeding period, show but ill effects from the feeding of the preservative. However, it is believed that most of this was caused by the error in judgment in feeding the formaldehyde in the milk. Had the preservative been added in the correct amount at the beginning, and that gradually as in the case of Calf No. 51, it is believed that no ill effects would have resulted. The gains and general condition by this calf fully justify this conclusion. This, however, is a single animal, and while it made better gains in every way, it cannot be concluded that all individuals would be affected in like manner even though the treatment was the same.

One conclusion, however, can be definitely drawn from the work with these calves. The calves fed sour milk did fully as well as any in the experiment, and it is believed that sour milk, always fed when at the same degree of acidity, will give as good returns as sweet milk, and the calves eating it will be less

subject to digestive disorders. It will be noted from a preceding table that this milk was soured the same each day, and always fed in a loppered condition.

It is evident that formaldehyde is a powerful disinfectant agent, and will, unless fed judiciously, produce an effect on the growth of the young animal. It must be fed in the correct amounts and must be fed continually. Further consideration may be found in final conclusions.

Second Experiment.

In order to further ascertain actual effects of sour and formaldehyde-treated milk, a second experiment was carried on during April, May and June, when artificial heat was not necessary to produce souring of untreated milk.

As before, three lots were fed sweet, sour, and formaldehyde-treated milk respectively. Morning's milk was treated in the evening with .007% formaldehyde and placed in the open air for the next morning's feed. A can of untreated milk was placed with it. The latter was in a loppered condition by morning, and was fed in that condition. Milk treated in a like manner in the morning was placed in the sun and open air to be fed in the evening. The formaldehyde-treated milk never soured, and the other varied in its sourness with the temperature, humidity of the air, etc. In a preceding table may be found data covering temperature, acidity of milk, etc. from May 1st to May 10th inclusive. This shows that the average difference in acidity of night and morning feeds of milk was nearly 40 degrees. On the third day, there was a difference of 61 degrees. The morning's feed was 35 degrees, and the evening's feed 96 degrees, which would mean from a slightly soured to a heavily loppered condition. Hence, it is evident that the sour milk fed the second lot was much different than in the first experiment, when the

acidity varied but slightly.

A grain mixture, as in previous experiment, was fed, with good alfalfa hay. Data concerning amounts, may be found in an accompanying table.

Only two calves were available for each lot, Careful selection was made in order, if possible, to make the lots equal in growing capacity. The table following gives the individual characteristics.

Individual Characteristics.

No.	Milk.	Age.	Breed.	Sex.	Weight	Condition	Remarks
1.	Sweet	21 days	Grd.Hol-stein.	M.	116	Fine	Vigorous
2.	Sweet	22 "	Grd.Here-ford	M.	96	Fair	Heifer's Calf.
3.	Sour	21 "	Grd.Hol-stein.	M.	114	Fine	Strong.
4.	Sour	21 "	Grd.Short-horn.	M.	100	Fine	Heifer's Calf.
5.	HCOH	22 "	Grd.Hol.	M.	143	Fine	Strong
6.	HCOH	20 "	Grd.Hol	M.	95	Fair	Poor Appetite.

The preliminary feeding was the same in all cases. Each individual was started on the experiment when three weeks old. Changes from whole milk to the experimental milk were made at the rate of one-half pound per day. For instance, Lot III had formaldehyde skim milk added at this rate, and the whole milk decreased until the milk ration was entirely formaldehyde-treated skim milk. The amount of milk consumed by the individuals is shown in the following table.

Milk Consumed.

No.	Milk	Weeks 1	2	3	4	5	6	7	8	9
1	Sweet	87	96	96	96	96	90	96	96	102
2	Sweet	80	64	64	64	64	64	96		
3	Sour	88.5	96	96	96	96	96	96	91	99
4	Sour	88.5	96	96	96	96	96	67	90	102
5	HCCH	91	98	90	90	96	96	96	90	102
6	HCOH	84	90	90	67	70	70	60	78	

Grain Consumed in Ounces.

No.	1	2	3	4	5	6	7	8	9	Average per week.
1	30	35	65	80	112	123	115	120	136	91
2	45	56	68	65	110	130	125			88
3	34	60	71	92	112	116	132	120	115	94
4	40	55	64	90	114	126	115	130	116	94
5	45	60	75	106	120	116	112	126	131	99
6	30	45	60	75	100	106	112	112		81

It will be noted that the amounts consumed did not vary greatly. No. 5 and No. 6 consumed the most and least amounts of grain, respectively. The amounts of hay eaten did not vary enough to have any significance in our study, and hence, is omitted. Each individual averaged about one pound per day for the whole period.

Weights by Weeks from Three Weeks Age.

No.	Original	1	2	3	4	5	6	7	8	9
1	116	126	132	136	148	152	166	178	188	196
2	98	112	120	127	137	141	143	159		
3	114	120	128	141	150	160	170	167	175	
4	100	112	125	128	136	144	148	143	160	
5	143	157	165	175	188	200	213	220	231	252
6	95	106	117	124	127	140	144	152	162	

Lot I. on Sweet Milk gained 8.3# per week per animal.

Lot II. " Sour " " 7.5# " " " "

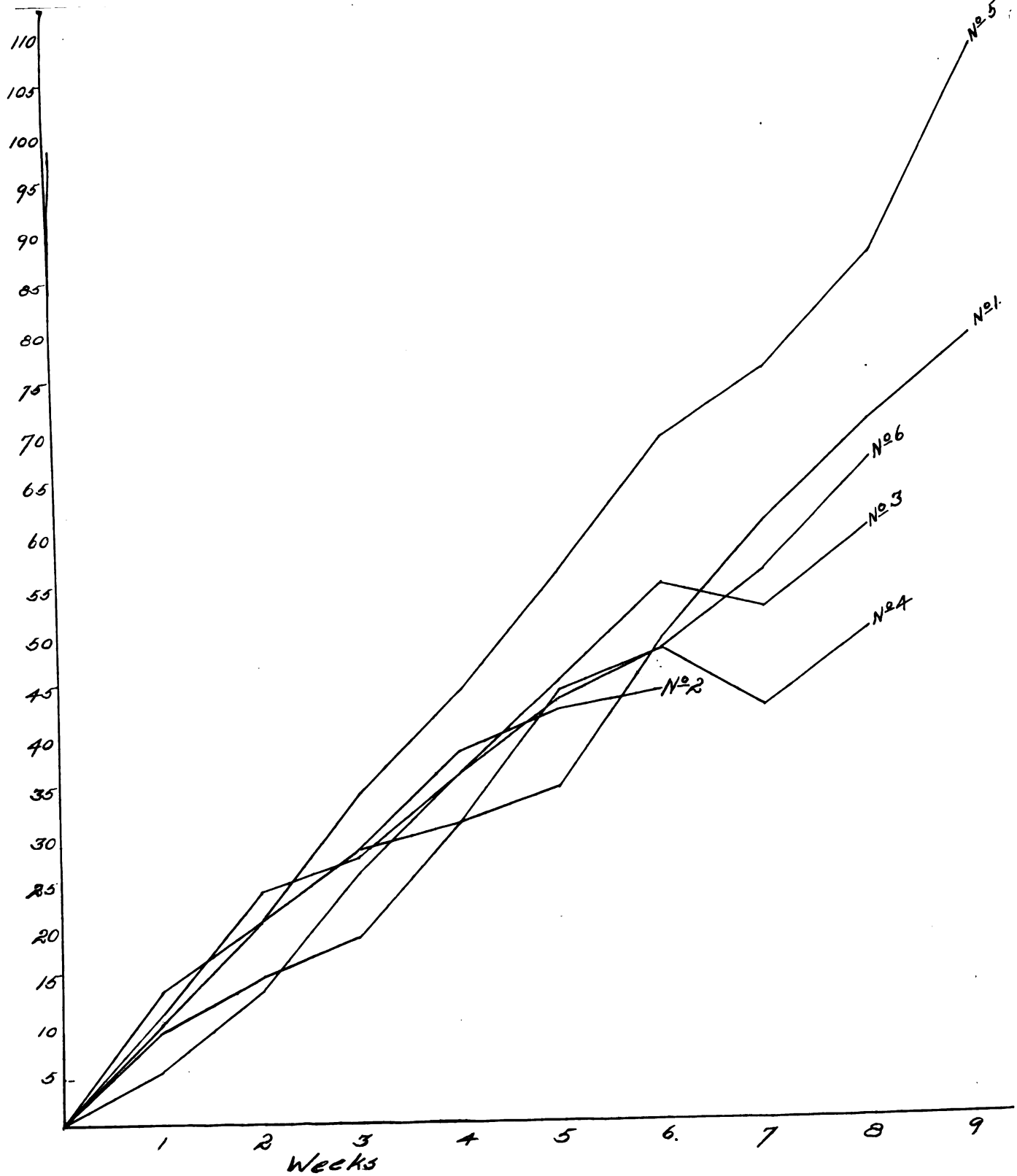
Lot III." Formaldehyde " 10.3# " " " "

Summary of Data.

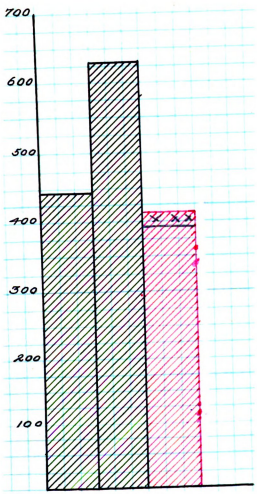
	Lot I.	Lot II.	Lot III.
Milk consumed	1467#	1735#	1523#
Grain "	90#	106#	90#
Hay "	97#	141#	121#
Total Gain	141#	121#	176#
Gain per calf per week	8.8#	7.5#	10.3#
Percentage gains	6[illegible]%	56%	74%
Gain per cwt. milk	9.5#	7.0#	11.5#
Cost per cwt. gain	$4.44	$6.34	3.63

From the preceding data, and graphs, it is evident that the lot consuming formaldehyde-treated milk made the greatest and most economical gains. Their general appearance and appetite was equal to those fed sweet milk. Those fed milk soured according to varying weather conditions lacked both growth and finish.

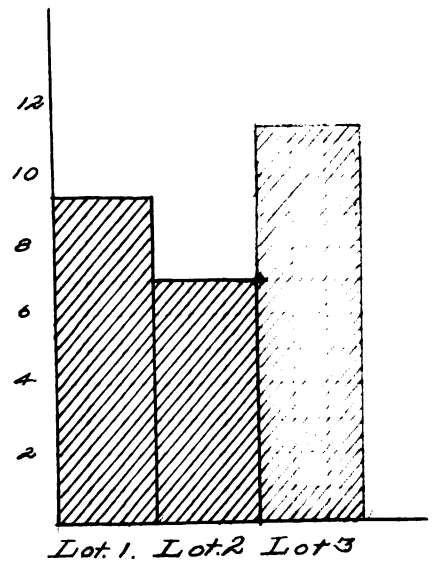
The graphs following were constructed from data recorded during experiment, and are self explanatory.



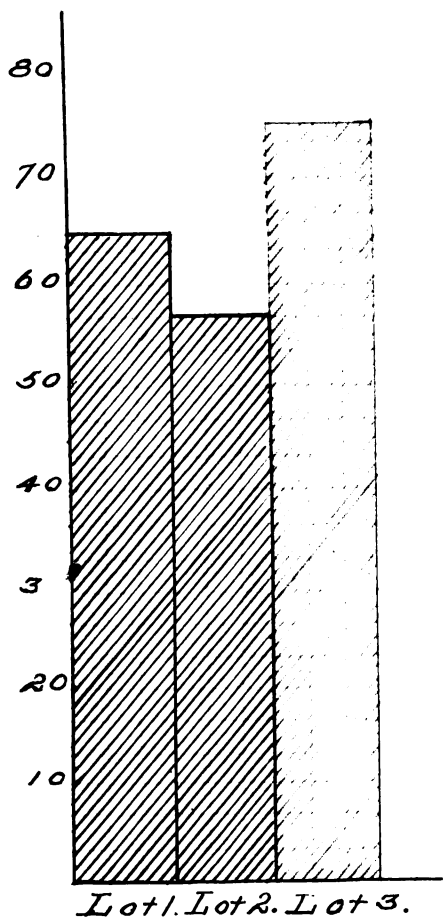
Gains of Individual by Weeks.



Cost per 100 lb gain.



Gains per 100 lbs. Milk fed.



Gains per 100 lbs. original weight.

Conclusions.

A careful review of the study in the foregoing, and an observance of the growth and condition of the animals, establish the following conclusions:-

1. Formaldehyde added to milk in the proportion of 1-15000 inhibits the growth of bacteria common to milk.

2. Formaldehyde added to milk in the proportion kills all bacteria, rendering the milk sterile.

3. Formaldehyde added to milk in the proportion of 1-15000 has no deleterious effect upon the digestibility of the milk.

4. In the proportion of 1-15000, formaldehyde and its action gradually disappears from milk.

5. Formaldehyde does not effect the coagulation of milk by rennet until a proportion of 1-2500 or more has been added.

6. Formaldehyde does not effect the coagulation of milk by pepsin until a proportion of 1-2000 or more has been added.

7. Formaldehyde added to milk in the proportion of 1-15000 has no effect upon the action of the digestive enzymes, pancreatin, steapsin, ptyalin, amylopsin and galactase.

8. Formaldehyde in amounts sufficient to preserve milk has no effects on the fat in the milk.

9. In the proportion of 1-15000, formaldehyde will keep milk sweet forty-eight hours, often longer, under summer conditions.

10. The Bacillus Subtillus group, Bact. tuberculosis and the molds are the most resistent, of organisms common to milk, to the action of formaldehyde.

11. Formaldehyde, as a preservative of milk for feeding purposes must be added to the ration gradually, or ill results may appear. The effects may vary with the individual.

12. Sour milk, the acidity of which varies but little from day to day, compares very favorably with sweet milk as a feed for calves.

13. Formaldehyde, fed in milk in the proportion of 1-15000, will not inhibit the growth of young calves.

Suggestions.

Formaldehyde in Calf Feeding.

From the work done this year, it is evident that this preservative may be of much value in the economical production of young animals. As has been described in this work, formaldehyde in the proportion of 1-15000 (1-6000 formalin) is sufficient for the preservation of milk for feeding purposes. Since formalin is the commercial product, computation of correct proportion has been made with it. This proportion may be secured in any one of the following ways.

1. 2. 5c.c. formalin to 33# of milk.
2. 7. 5c.c. formalin to 100# of milk.
3. Teaspoonful formalin to 65# of milk.
4. 2 parts water and 1 part formalin, teaspoonful to 20# of milk.
5. 1 liquid ounce of formalin to 400# milk.

This should be added as soon as milk comes from the separator, and mixed very thoroughly. In case of poor mixture, the formaldehyde seems to preserve the upper portions of milk, and will allow souring next to the can near the bottom. This preservative should not induce uncleanliness on the part of the dairyman since by its use he can cover up his unsanitary methods.

When introduced into the calf's ration, it should be done gradually, and at the time the calf is changing to a skim milk diet. Add one-half pound per day of formaldehyde-treated milk until the calf is on a ration of this milk, and no ill results will appear.

In the case of scours in calves, the author has used, with good success, the following:-

Add one-half ounce of formalin to 15½ ounces distilled or rain water. Of this mixture, add teaspoonful for every pound of milk fed. Two or three feeds of this should sufficiently check the disease.

Bibliography.

Facts about Milk. Farmer's Bulletin No. 42.

Ward and Jaffa
Pure Milk and the Public Health.1909 pp.218.

Milk and its Relation to the Public Health. Hygienic Laboratory, Bulletin 41.p.758.1908.

Dairy Herd Record and Creamery Notes Alabama Expt. Station. Bulletin No. 121.

C. F. Doane.
The Character of Milk during the period of heat. Maryland Expt. Station. Bul.No.95.

H. A. Harding.
Notes on Some Dairy Troubles. N.Y.Expt.Station Bulletin 183.

C. E. Marshall.
A Popular Discussion of Pure Milk Supply.Mich.182.

Law.
Tuberculosis in relation to animal industry and public health. N. Y. Cornell Bul. 65.

H. L. Bolley.
Cleanliness in Handling Milk. N.D. Expt. Station Bulletin 21.

R. W. Clark.
Care of Milk on the Farm. Utah Sta. Bul 96.

Cottrell.
Keeping Milk in Summer. Kans. Station Bul.88.

C. F. Doane.
Economical Methods of Improving the Keeping Qualities of Milk. Md. Sta. Bul. 88.

R. A. Pearson.
Care of Milk on the Farm. Farmer's Bulletin.63.

C. S. Plumb.
Dairy Experiments. Ind. Station Bul. 44.

Alvord and Pearson.
The Milk Supply of 200 Cities and Towns. Bur. An. Ind. Bul. 46.

C. F. Doane.
The Milk Supply of 29 Southern Cities.B.A.I.Bul. 70.

Lindsey and Smith.
Market Milk. Mass.Station.Bul.110.

W. A. Stocking.
Studies of Market Milk. Storrs Sta.Rpt.1905.

H. W. Conn.
The Relation of Temperature to the Keeping Property of Milk. Storrs Sta.Bul.26.

S. H. Ayers.
The Pasteurization of Milk.Bur.An.Ind.Cir.No.184.

A. R. Leeds.
The chemical and physical changes attendant upon the sterilization of milk. Jour.Am. Chem.Soc.1891.

C. E. Marshall.
Killing the Tubercule Bacillus in Milk. Mich. Station Bul. 173.

H. W. Conn.
Fermentations of Milk. Office Expt.Stations Bulletin No. 9.
Classification of Dairy Bacteria. Storrs Sta. Rpt. 1899.
Comparative growth of different species of bacteria in normal milk. Storrs.Rpt. 1901.
A Classification of Dairy Bacteria.Storrs 1906.

R. R. Dinwiddie.
Milk, its decomposition and Preservation. Ark. Sta. Bul.45.

Eckles and Barnes.
Bacteriological study of the college creamery supply of milk. Iowa Station Bul.59.

P. G. Heinmann.
The kinds of lactic acid produced by lactic bacteria. Jour. Biol. Chem. 1907.pp 602-612.

O. F. Hunziker.
Germicidal action in cow's milk. Cornell Bul.197.

L. L. Lewis.
Bacteriology of Milk. Okla.Station Bul. No. 40.

C. E. Marshall.
Associative action of bacteria in souring milk. Mich. Special Buls.29 and 33.

L. H. Pammel.
Notes on Dairy Bacteriology.
Iowa Station Bul.34.

Rosenau and McCoy.
The germicidal property of milk.Jour.Med.Research. March 1908.

W. A. Stocking.
The so-called germicidal property of milk. Storrs Sta. Bul. 37.

Rosenau and McCoy.
Public health and marine hospital service.U.S.A. Bulletin No. 41 1908.

The feeding value of skim milk. Mass. Annula Rpts. 1884, 85, 86, 87 and 89.

The feeding value of skim milk. Vermont Sta.Bul.18.

The Comparative value of skim milk and buttermilk. Wisconsin Annual Rpt. for 1886.

The comparative feeding values of buttermilk and skim milk. Mass. Annual Rpts. for 1884 and 85.

The comparative value of sour and sweet skim milk. Vermont annual rpt. for 1891.

Castle and Roberts.
The Chemistry of Milk. Hyg. Lab. U. S. Pub. Health and Marine Hospital Service.Bul.No.41.

The Dairy Calf at Meal Time. Wis.Bul.192.

Skim Milk for Pigs. Cornell Sta.Bul.141.

Germicidal Action of Milk. Cornell Sta.Bul.197.

Substitutes for skim milk for calves. Cornell Bul.269.

Milk in hot weather. Okla. Rpt. Vol.22.

Care of milk on the farm.B.A.I.Bul.63.

S. Henry Ayers.
A study of the bacteria which survive pasteurization. Bur.An.Ind.Bul.161.

C. B. Cochran.
Penn. Dept. of Agri. for 1899.p.277

Journal American Chemical Society No. 27 1905 p. 1060,1497.

" " " " pp.189, 1906.

Iowa Station Bulletin No. 32.

Babcock and Russell.
Wis. Annual Report 1898.

Van Slyke.
Rpt. N. Y. Expt. Station 1901.

Symons.
Journal American Chemical Society 1897.

Ridcal and Foulerton.
Public Health Bul. 1899.

A. R. Ward.
Pure Milk and Public Health, U. of Cal.

Farrington and Woll.
Testing Milk and its products.

Publow and Troy.
Questions and Answers on Milk and Milk Testing.

Ross.
Dairy Laboratory Guide.

Snyder.
Dairy Chemistry.

Russell and Hastings.
Outlines of Dairy Bacteriology.

Conn.
Practical Dairy Bacteriology.

Lipman.
Bacteria in Relation to Country Life.

Larson and White.
Dairy Technology.

Michael.
Market Dairying.

Van Norman.
First Lessons in Dairying.

Lane.
The Business of Dairying.

Woll.
Handbook for Farmers and Dairymen.

Michels.
Dairy Farming.

Monrad.
Pasteurization and Milk Preservation.

Belcher.
Clean Milk.

Wing.
Milk and Its Products.

Pearson.
Jensen's Milk Hygiene.

Spargo.
Common Sense and the Milk Question.

Henry.
Feeds and Feeding.

U. S. Dept. Year Books.
By Products. 1897.
Dairy Development. 1899.
Formaldehyde. 1905.

Aikman.
Milk. Its Nature and Composition.

Winslow.
Production and Handling of Clean Milk.

Grotenfelt.
Principles of Modern Dairy Practice.

Gurler.
American Dairying.

Stewart.
Dairyman's Manual.

Sheldon.
The Farm and the Dairy.

Lang.
Dairy Farming.

U. S. Consular Reports.
Cattle and Dairy Farming--3 books.

Report Wis. Dairyman's Ass'n.
1906 and 1907.

Dean.
Canadian Dairying.

Fleischman (Translated by Aikman and Wright.)
The Book of the Dairy.

Hoard's Dairyman.
Vol. 42 p 1037.
Vol. 31 p 976.
Vol. 31 p 1036.

Kellner.
The Scientific Feeding of Animals.

Wiley.
Foods and their Adulteration.

Rosenau.
The Milk Question.

Price.
The Handbook of Sanitation.

Swithinbank and Newman.
Bacteriology of Milk.

Dr. F. Lohn.
Handbuch der landwirtschafthchen Bacteriologie.

Savage.
Milk and the Public Health.

Annette.
Boric acid and Formalin as Milk Preservatives.

Kastle and Roberts.
The Chemistry of Milk. Bul.41 Pub. Heal.and
Marine Hospital Service. 1908.

Leach.
Food Inspection and Analysis. 1906.