# CONTAMINATION OF RUBBER MILK TUBES OF MILKING MACHINES AS AFFECTED BY DETERIORATION OF THE INSIDE SURFACES

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**Studies were made on the susceptibility to bacterial contamination of rubber milk tubes as affected by deterioration of the inside surfaces. Even with used tubes having only microscopic breakdown, contamination was much greater than with new tubes. The average contamination in the used tubes averaged 15 times as great as in new tubes under laboratory conditions and 12 times as great under practical operating conditions with dry storage. When lye storage was used bacterial counts were lower and differences were smaller. Even inconspicuous deterioration of the interior surfaces of rubber milk tubes is a potential hazard in milking machine sanitation.**

In milking machine sanitation, increased attention is being given to the rubber parts as factors in contamination[1, 2, 3, 4, 5, 6]. Although obvious breakdown of rubber is recognized as a sanitary hazard, little consideration has been given to the influence of less evident conditions. It has been shown that inconspicuous deterioration of the interior surfaces of teat-cup liners is an important factor affecting the susceptibility of the liners to contamination[2]. It might be expected that the same would prevail for the rubber milk tubes. On the other hand, since the tubes are not subjected to flexing and squeezing action and there is less likelihood of fat and other milk materials being worked into the rubber, the condition might be less significant. Accordingly, studies were made to determine what effect breakdown of the inner surfaces of milk tubes had on their susceptibility to bacterial contamination.

## METHODS

During the study, 10 used milk tubes, currently in service, were obtained from three grade-A dairy farms. The tubes were from bucket type machines, and were of one brand, since others were not readily available in the area. They had undergone various sanitizing treatments and had been in service for periods ranging from 5 to 18 months.

In the laboratory the tubes were thoroughly cleaned by soaking in organic acid solution[b], washing, boiling in 2% lye solution for 15 minutes, and again washing. They then were inspected for general physical condition. Since the tubes were to be used in experimental work, they could not be cut open at this time for close inspection of the interior surfaces. Examination was made as fully as possible by viewing from the open ends against a light source. Following experimental work, the tubes were cut and the interior surfaces examined under a stereoscopic microscope at 85X magnification.

*Bacteriological studies* — To determine the importance of surface condition of the rubber milk tubes in sanitation problems, bacteriological studies were made on the used tubes in comparison with new tubes of the same type and under the same conditions. In some laboratory trials, new plastic tubes were included for comparison. At the start of each comparison, the cleaned new and used tubes were steam sterilized. They then were contaminated either by experimental laboratory procedures or by usage under practical operating conditions.

In experimental contamination, a 35 ml. quantity of incubated raw milk was added to each milk tube stoppered at one end. After stoppering the other end, a vigorous and uniform shaking procedure was used to distribute the milk over the interior surface. After draining off the milk, the tube was rinsed briefly under a water faucet to flush out free milk droplets, again drained and then examined bacteriologically.

When contamination was achieved under practical operating conditions, a used tube was placed on one milker unit and compared with a new tube on a second unit of the same type under the same routine conditions at the College dairy barn. In the initial comparisons, tubes were brought into the laboratory immediately after evening milkings, flushed with lukewarm water under a faucet and held dry overnight. Bacteriological examinations were made the following morning. In the remaining investigations, washing was done as usual at the dairy barn.[c] In one phase the teat-cup assemblies and tubes were stored in a lye rack after washing and in another they were hung to drain and stored dry. Milk tubes were removed before the afternoon milking and taken to the laboratory for bacteriological examination. Where lye storage had been used, the tubes were first rinsed briefly under a water faucet to remove excess lye solution.

Bacteriological examinations were made by adding 35 ml. of sterile water to each tube, suitably stoppered with sterile stoppers. After a uniform shaking procedure, the rinse water was plated immediately, using Standard Plate methods and tryptone glucose extract agar. Although it is recognized that a considerable proportion of the bacteria would remain on the surfaces of the rubber, this rinse procedure was considered satisfactory for comparative studies.

## RESULTS

*General physical conditions of milk tubes*—The condition of the interior surfaces of the cleaned, used tubes, as viewed from the open ends against a light source, varied from dull and slightly scratched to definitely rough and broken surfaces. Some tubes appeared little different from new tubes. Upon cutting open the used tubes, deterioration of the inner surface sometimes was evident to the unaided eye as a roughened, finely cracked surface. In other tubes the surfaces appeared clean and smooth with no obvious breaks. None of the tubes showed softness or tackiness of the interior surfaces as is sometimes evident in teat-cup liners as a result of fat absorption.

The interior surfaces of new tubes viewed from the ends did not appear so smooth as expected. Some surface irregularities and apparent

[a]Contribution No. 224 Department of Dairy Husbandry, Kansas Agricultural Experiment Station.

[b]Commercial preparation used as directed for removal of milkstone.

[c]The teat-cup assemblies and tubes were promptly flushed with tepid water. A detergent solution at about 140° F., followed by clear water at the same temperature, was then drawn through the assemblies.

TABLE 1—EXAMPLES OF BACTERIAL COUNTS ON USED AND NEW TUBES AFTER EXPERIMENTAL CONTAMINATION FROM THE SAME LOTS OF MILK.[a]

| Trial[b] | Counts per ml. water rinse[c] | | |
|---|---|---|---|
| | Rubber tubes | | Plastic tubes |
| | Used | New | New |
| 1 | 44,000 | 9,000 | 1,100 |
| 2 | 2,400 | 110 | 95 |
| 3 | 14,000 | 1,200 | 450 |
| 4 | 75,000 | 700 | 600 |
| 5 | 10,000 | 1,800 | 220 |
| 6 | 6,600 | 260 | 140 |
| Log av. | 13,940 | 855 | 310 |

[a]Each trial involved a different lot of milk of different quality.

[b]Each trial included a different used tube, but the same new tubes were usually, although not always, used.

[c]35 ml. water rinse used per tube.

TABLE 2—BACTERIAL COUNTS ON USED AND NEW RUBBER TUBES AFTER OPERATION UNDER THE SAME PRACTICAL CONDITIONS.[a]

| Trial[b] | Days in operation | Counts per ml. water rinse[c] | |
|---|---|---|---|
| | | Used tube | new tube |
| 1 | 1 | 78 | 4 |
| | 2 | 2,000 | 150 |
| | 3 | 1,760 | 16 |
| 2 | 1 | 10 | 5 |
| | 3 | 710 | 10 |
| | 4 | 2,500 | 150 |
| Log av. | | 410 | 20 |

[a]Before making bacteriological examinations, the tubes were flush washed and held as described under "Methods".

[b]Each trial involved a different set of milk tubes.

[c]35 ml. water rinse used per tube.

TABLE 3—BACTERIAL COUNTS ON USED AND NEW RUBBER TUBES AFTER OPERATION AND SANITIZING UNDER THE SAME PRACTICAL CONDITIONS (DRY STORAGE)

| Trial[a] | Days in operation | Counts per ml. water rinse[b] | |
|---|---|---|---|
| | | Used tube | new tube |
| 1 | 1 | 24,000 | 350 |
| | 2 | 700 | 300 |
| | 4 | 1,200,000[c] | 120,000[c] |
| | 5 | 1,000,000[c] | 100,000[c] |
| | 6 | 1,500,000[c] | 100,000[c] |
| 2 | 1 | 1,300 | 240 |
| | 2 | 4,000 | 600 |
| | 3 | 10,500 | 410 |
| | 5 | 300,000[c] | 25,000 |
| | 7 | 14,000 | 400 |
| Log av. | | 38,380 | 3,070 |

[a]Each trial involved a different set of milk tubes.

[b]35 ml. water rinse used per tube.

[c]Estimates, plates crowded.

scratches or creases were evident. On cutting open, the surfaces showed no breaks or roughness, but did not have the smooth, even surface usually seen in new teat-cup liners.

On microscopic examination the interior surfaces of the used milk tubes were found to have the same general types of breakdown as previously found in used teat-cup liners and illustrated in an earlier report[2]. Surfaces that appeared dull and rough to the unaided eye were intensely cracked and disintegrated. Even those surfaces that appeared unbroken macroscopically were eroded and pitted, giving a spongelike effect. Combinations of these two general types of breakdown were common.

With new tubes the microscopic appearance of the interior surfaces supported the impression gained macroscopically. Although no cracking was evident and the surfaces were much better than those of the used tubes, the new tube surfaces were irregular and uneven and gave the impression of being "unfinished".

*Effect of deterioration of the inner surface of the tube on susceptibility to contamination* — When new and used rubber tubes were experimentally contaminated in the laboratory under the same conditions, the bacterial counts on the used tubes were much higher in every case than those on the new tubes. In 17 trials involving 25 comparisons of 10 used tubes with new tubes the counts on the used tubes ranged from four to more than 100 times as great as the corresponding counts on the new tubes. The log average count on the used tubes was 13,200 and on the new tubes 880. The log average count on new plastic tubes used for comparison in 14 of the trials was 420. Results of six representative trials are presented as examples in Table 1. Different lots of milk used in the different trials accounted for the range of contamination among trials.

When contamination of the tubes occurred under practical operating conditions, with the tubes being stored dry following the sanitizing procedure, counts on the used tubes again were higher than those on the new tubes. After laboratory flushing and dry storage, counts were low to moderately low

with both used and new tubes (Table 2). However, in the six comparisons with two different sets of tubes, the log average count on the used tubes was 20 times as great as the log average count on the new tubes. Although the trials were of short duration, there was a tendency for the counts to become higher in both new and used tubes as the trials progressed.

When tubes were sanitized at the dairy barn under routine conditions and held dry, the bacterial counts were frequently high. (Table 3) As in previous trials the used tubes were more heavily contaminated in every case than the new tubes, ranging from slightly more than twice as great to almost 70 times as great. In the 10 comparisons, involving two different sets of tubes, the log average count on the used tubes was about 12 times as great as the corresponding count on the new tubes. The wide range of contamination among the different comparisons presumably reflected the variation in efficiency of the sanitizing operations.

When the teat-cup assemblies and milk tubes were stored in a lye rack following routine washing operations, the counts on both new and used tubes were usually low (Table 4). With two exceptions the counts on the used and new tubes were approximately the same. In the 11 comparisons with two sets of tubes, the log average count on the used tubes was slightly more than four times as great as the log average count of the new tubes. There was no particular tendency for the counts to become higher as the trials progressed.

TABLE 4—BACTERIAL COUNTS ON USED AND NEW RUBBER TUBES AFTER OPERATION AND SANITIZING UNDER THE SAME PRACTICAL CONDITIONS (LYE RACK STORAGE)

| Trial[a] | Days in operation | Counts per ml. water rinse[b] | |
|---|---|---|---|
| | | Used tube | new tube |
| 1 | 1 | 47 | 43 |
| | 2 | 1,200 | 18 |
| | 4 | 29 | 20 |
| | 6 | 25 | 1 |
| | 12 | 100 | 110 |
| | 25 | 5,800 | 130 |
| 2 | 1 | 65 | 8 |
| | 2 | 410 | 28 |
| | 4 | 20 | 9 |
| | 5 | 18 | 35 |
| | 7 | 36 | 38 |
| Log av. | | 98 | 22 |

[a]Each trial involved a different set of milk tubes.
[b]35 ml. rinse water used per tube.

## DISCUSSION

The experimental contamination procedure was a useful means of measuring the effect of deterioration of rubber milk tubes on susceptibility to bacterial contamination. The results obtained generally were supported by later trials where contamination occurred under practical operating conditions and where dry storage was used.

It is evident that surface breakdown that occurs in tubes while they are in service magnifies the contamination problem. Observations indicated that tubes appearing in reasonably good condition and in which cracking was largely of microscopic nature were often as susceptible to contamination as were those tubes having more obvious breakdown. In other words the length of life of the tubes from a sanitation standpoint was less than when determined by clearly evident physical deterioration or by operating efficiency. This relationship was generally similar to that demonstrated with rubber teat-cup liners[2].

Although the study was not designed to compare the effects of sanitizing treatments on the contamination of used and new tubes, it appears that lye storage is a means of reducing the differences in bacterial counts. However, such a conclusion should be made with reservations since the trials were made at different times than the dry storage trials and other conditions may have varied. The further fact that, in two of the comparisons, counts on the used tubes were considerably higher than counts on the new tubes indicates that lye storage is not an infallible means of counteracting the effect of surface breakdown. Nevertheless, the results suggest that such treatment helps to reduce the contamination hazard of milk tubes having surface deterioration.

The tendency on farms is to use milk tubes until they will no longer remain attached to the pail head or claw, or until the ends have been trimmed back so that the tubes are too short for further use. Observations made during the study indicate that this stage is far past the point where deterioration of the inner surfaces constitutes an increased contamination hazard. With the increase in pipeline milking installations where the milk tubing is usually longer, it is likely that surface breakdown in the tubes will be a proportionately greater problem.

The low counts obtained on new plastic tubes and the smooth nature of the inner surfaces suggest some advantages for this material and further studies are being made over longer periods of practical operation with this type of tube.

## SUMMARY AND CONCLUSIONS

Studies conducted on used rubber milk tubes in comparison with new rubber tubes revealed that deterioration of the inside surfaces increased the susceptibility of the tubes to bacterial contamination. This relationship existed even with tubes having only microscopic breakdown and otherwise appearing in satisfactory physical condition. With experimental contamination, the log average count on used tubes was 15 times as great as the corresponding count on new tubes. When contamination occurred under conditions of practical operation and sanitizing, followed by

dry storage, the log average count on the used tubes was 12 times as great as the log average count on the new tubes. Where lye storage was used in place of dry storage, counts were usually lower and the differences between used and new tubes were considerably reduced in most cases.

It is evident that deterioration of the interior surfaces of rubber milk tubes is a potential hazard in milking machine sanitation. The condition may develop while the tubes still appear to be in reasonably satisfactory operating condition. Modifications in rubber composition or utilization of suitable plastic milk tubes might minimize the problem.

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# THE COLIFORM BACTERIA OF STRAWBERRIES*

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**The positive presumptive tests obtained on the examination of strawberries undergoing processing for freezing are generally caused by Gram negative bacteria indigenous to plants and to the soil. The bacteria produce gas at the expense of the sugars naturally present in the berries. Washing the berries in chlorinated water or in water containing detergents does not eliminate the microorganisms. The incidence of coliform bacteria appears to be associated with dirty fruit, and soft and unsound fruit. A greater number of positive presumptive tests are obtained with lauryl tryptose broth than with brilliant green bile 2 percent broth. The former medium is more efficient in detecting the organisms of human and of animal origin.**

Tanner[1] has reviewed the literature concerned with the presence of coliform bacteria on strawberries. Early workers were generally agreed that these organisms are ubiquitous, and that their removal by chlorination of the wash water is uncertain. Recently Barber[2], discussing the bacteriology of raw fruits, stated that neither quaternary ammonium sanitizers at 200 ppm nor chlorine solutions at 1,000 ppm during an exposure of 15 minutes effected destruction of coliform bacteria. The raw fruit under discussion could well be strawberries, although not mentioned by name.

During experimental and routine bacteriological examinations of strawberries in the processing season, it was noticed that a large proportion of both unprocessed and processed berries yielded positive presumptive tests for coliform organisms. Generally, the true coliform bacteria could not be confirmed on the customary media, although the bacteria in the positive presumptive tubes were Gram-negative, and grew prolifically, although atypically, on eosin-methylene blue (EMB) agar.

The typical positive presumptive test also may be considered confirmatory for the presence of coliform organisms, according to *Standard Methods for the Examination of Dairy Products*[3]. Therefore studies were initiated to determine (a) the nature of the organisms responsible for the positive presumptive reactions, (b) the actual presence of organisms of human origin, and (c) the effect of washing berries in continuously chlorinated water (inplant chlorination) upon the incidence of these bacteria.

## EXPERIMENTAL PROCEDURES

Both lauryl tryptose (LS) broth and brilliant green-bile 2 percent (BGB) broth were employed, the latter because it is a standard medium for this purpose, and the former, because Mallmann and Darby[4] recommended its use for the presumptive isolation of coliform bacteria.

The strawberries were of the *Blakemore* and *Tennessee Beauty* varieties ,each of which has between 8 percent and 10 percent sugar, of which 75 percent is reducing sugar. All samples were obtained from the processing lines at the Food Technology Building of the University of Tennessee, or from two commercial plants, one in East Tennessee and one in West Tennessee. Samples were taken at various stages of processing, from the raw product in the crate through packaging. Berries were washed for approximately 75 seconds in soaker-sprayer washers with water which was chlorinated on request to provide inplant chlorination at the levels desired.

Approximately 1-pound samples were gathered in sterile jars and liquidized. Presumptive inoculations were made in decimal dilutions beginning with 1 ml of the fluid in 5 tubes of the presumptive media at each dilution. Positive tubes were streaked on EMB agar, and colonies were picked for further study. Plating media were Bacto violet red-bile agar and BBL desoxycholate agar.

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