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The Detection of Mastitis In Dairy Herds

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The Detection of Mastitis in Dairy Herds

D. H. Jacobsen and T. M. Olson

Summary

The tests considered in this work were applied to 1812 samples of milk taken from 54 cows. The tests showed a much more frequent occurrence of abnormal milk than was expected. Before the experiment was started very few noticeable indications of garget or mastitis were detected by the milkers and herdsman. The study herein reported deals with the occurrence of abnormal milk in the college dairy herd and a comparison of the results of various tests including the brom thymol blue test, the catalase test, the leucocyte count and the examination for long-chain streptococci.

Of the 54 cows tested only 14 produced milk which was positive to all of the tests used and was physically abnormal i.e. contained clots, sediment and was thick or serous in consistency. Twenty-seven individuals showed positive brom thymol blue and catalase tests and high leucocyte counts. Of these 27 there were 24 which showed long-chain streptococci in the milk. Three cows gave positive brom thymol blue and catalase tests and showed high leucocyte counts but failed to show long-chain streptococci in the samples examined.

Of the 14 cows which produced milk of abnormal consistency the following number of abnormal milk samples were noted, cell count 79.0 per cent, catalase test, 77.8 per cent, brom thymol blue test, 73.5 per cent, and long-chain streptococci 46.3 per cent.

Thirteen cows which were termed suspicious to positive but never showed milk of abnormal consistency during the experiment gave positive laboratory tests as follows:

Of 540 "quarter" samples tested

174 or 32.2 per cent were positive to the brom thymol blue test.

192 or 35.6 per cent showed more than 500,000 leucocytes per cc.

252 or 46.7 per cent produced more than 1.5 cc. gas by the catalase test.

88 or 16.3 per cent showed long-chain streptococci in incubated samples.

Causes and Control of Mastitis

Mastitis is perhaps of more economic importance than any of the diseases common to dairy herds. Decreased milk production and discarded milk causes enormous losses to the dairyman. It has been estimated by good authorities that around twenty-five per cent of all cows culled from dairy herds are sold because of udder trouble.

To prevent spread of infection it is very necessary to understand the causes and symptoms of mastitis. The present work deals with the detection of mastitis by testing for abnormal milk which is associated with the infection in the udder .

A number of factors have been considered as causes of mastitis. In most herds, however, it is a combination of factors which contribute to the incidence and spread of the disease. The two main causes of mastitis, however, are the badly infected cow and unsanitary stable management. Udall (15) lists as causes of mastitis: primarily infection from active cases, also injuries, rough manipulation, incomplete milking, high plane of protein feeding and improper use of milking machines. Although the direct cause of mastitis usually is infection from active cases the other factors may act as contributing causes in making the udder tissue more susceptible to infection.

Some recommended methods of preventing the spread of mastitis are as follows:

1. Test every quarter of every cow at regular intervals to locate sources of infection.
2. Isolate badly infected cows by placing them at the end of the milking line and use extreme precautions at milking time.
3. Destroy all stringy or badly infected milk. Never strip infected milk into the bedding.
4. Wash the hands after milking each cow if infected individuals are milked with normal cows.

5. If milking machines are used, dip the teat cups in a disinfectant solution to prevent carrying infection from one cow to another.

6. Careful feeding and management.

Avoid exposure during inclement weather.

Avoid crowding in sheds and stanchions.

Avoid excessively high protein rations.

Use plenty of bedding.

7. When badly infected individuals become unprofitable, remove them completely from the herd.

The detection of infected cows through testing seems to be an important part of any system of control. Therefore practical methods of testing should be given greater consideration. The tests considered in this report are laboratory tests which may be used in conjunction with a barn test such as the strip cup.

The strip cup is simply some form of cup over which either a black cloth or fine screen (100 mesh) is spread. The strip cup is used by stripping the first four or five streams from each teat to find cows which show milk containing clots, flakes or stringy masses. Such milk should never be included in the supply and it is also dangerous as a source of infection to the uninfected cows in the herd. Cows showing abnormal milk by the strip cup test require no further testing. However, many cases never reach such an acute stage and yet they harbor the infectious organisms and are a dangerous source of infection in the herd. It is to locate such cows that we resort to laboratory tests such as are reported in this work.

The Problem

The tests studied in this work were selected as the most practicable for use in control work. These tests were applied to the college dairy herd with the purpose of obtaining more information on the relation of these tests to the occurrence of pronounced and suspicious mastitis infections and to assist the dairyman in the selection of a test which can be used in a practical system of control.

There is a great deal of confusion among dairymen as to the conditions which indicate the udder disease known as mastitis. This is not surprising, however, in light of present knowledge that mastitis occurs in many forms differing in type and degree

of infection. The type of mastitis which is easily recognized is clinical mastitis. In this type of mastitis the udder becomes inflamed and little or no milk is produced. This form of mastitis is comparatively rare.

The type of mastitis which is recognized as most common is chronic mastitis which is characterized by mild but recurrent inflammation of the udder and abnormal milk. Udall (15) defines chronic mastitis as follows: "Chronic mastitis is a progressive inflammation of the udder characterized by frequent or occasional acute activity. Either constantly or at intervals the milk contains flakes or clots, shows increased alkalinity, reacts to various biological tests and reveals streptococci on blood agar plates. At all times indurations may be found on physical examination of the udder. Pathologically there is an atrophy of the glandular tissue, suppurative infiltration or abscess formation of fibrosis. The chief bacterial agent is *Streptococcus mastitidis*."

The extent of infection in dairy herds has been estimated to range from twenty to fifty per cent, and by far the greatest number of cases have been shown to be of the type known as chronic mastitis. Measures designed to cure mastitis have not proved successful; therefore the disease must be controlled. This can be done only by finding the infected individuals and adopting measures to prevent the spread of infection from one cow to another. This is not a simple problem because in the mild form there is little or no outward indication of infection. It is to locate these mild but potentially dangerous cases that we must resort to laboratory tests of the milk.

That laboratory tests can be relied upon to indicate pathological conditions in the udder has been proved by a number of investigators. The purpose of the present work was to show the correlation between a number of these laboratory tests for the detection of mastitis.

Historical

Some of the more recent studies on the chemical and bacteriological tests for abnormal milk and their relation to mastitis are presented here. The tests devised for detecting abnormal milk caused by mastitis are reviewed to show the background on which certain conclusions have been developed.

Most dairymen judge the presence or absence of mastitis in the herd by the occurrence of stringy, clotty or flaky milk. When these signs appear, however, the disease has reached an advanced stage and extensive infection of other cows in the herd may have occurred. Also there are many udders harboring the mastitis infection in which stringy or flaky milk is never found. These cases are, nevertheless, a source of infection in the herd. The flakes or floccules in mastitis milk are composed of masses of leucocytes or cells and mucus according to Jones and Little (7). They found that such milk contained an increased leucocyte count as well as the causative organisms, hemolytic streptococci and staphylococci.

A number of workers including Baker and Breed (1); Tweed (14); Jones and Little (7); Johnson and Trudell (8); Hucker et al (5); Hucker and Udall (6); Halversen et al (3) and Prouty (10) have attached considerable importance to the leucocyte count in milk as a means of detecting mastitis. Although they do not agree on definite limits for normal and abnormal milk, their work shows very definitely that high cell counts indicate mastitis; and further that more severe stages of the infection are reflected by relatively higher leucocyte counts.

Hucker and Udall (6) and Johnson and Trudell (8) found that increased leucocyte counts were associated with physical changes in the udder. They considered the leucocyte count to be a good indication of the degree of fibrosis developed in the udder. Johnson and Trudell (8) also stated that "In mastitis there appears to be an increase in leucocytes which precedes the appearance of streptococci or staphylococci in milk . . . When the cell count reaches millions per cc. and persists it indicates infection, advanced mastitis and pus in the udder."

Prouty (10), in a comparison of the leucocyte count with the

brom thymol blue test and catalase test, found that as the leucocyte count increased there were successively fewer samples showing normal reactions to the other tests.

The upper limit for leucocyte counts in normal milk has been set at various points up to 500,000 per cc. Hucker and Udall (6) state that udders free from induration or scar tissue are free from demonstrable streptococci, do not show cells in excess of 500,000 per cc. and do not give a positive reaction to the brom thymol blue test. Halversen et al (3) concluded that milk from cows infected with chronic mastitis usually contains leucocytes in excess of 100,000 per cc. Cherrington et al (2) finds that normal udders usually contain less than 50,000 cells per cc. whereas milk from infected udders invariably contains more than 100,000 per cc. The difference between the limits set by various workers can no doubt be attributed to the different standards used in describing normal milk or normal udders.

The change in reaction of milk from pH 6.3-6.6 to neutral or alkaline has been used as the basis for the brom thymol blue test. The test has been given a great deal of study in recent years by Hucker et. al. (5), Hucker and Udall (6), Rosell (11), Horrall (4), and Prouty (10), who have shown the test to be very sensitive to milk which was abnormal by other tests such as the leucocyte count, catalase and chloride test.

For a confirmed diagnosis of mastitis, workers fail to agree on any one method but state that the results of a number of tests must be considered. Udall and Johnson (15) consider the physical examination of the udder to be the most important. They consider the development of fibrosis to be the final proof of mastitis infection. Rosell (13), however, states that "Long or short-chained streptococci in milk drawn aseptically or studied shortly after milking if found with a large number of leucocytes are always a sure sign of mastitis." In the absence of a physical examination of the udder, which requires a specially trained technician we can no doubt use the presence of long-chain streptococci as the confirmatory test as it is used in the present work.

The catalase test has been studied by a number of investigators including Halversen et al (3), Hucker et al (5) and Prouty

(10). Their results indicate a close agreement between this test and other laboratory tests used for indicating an active mastitis infection. The production of more than 1.5 cc. of gas from the 15 cc. of milk has been generally considered a positive test.

Method of Procedure

This study was made on the College dairy herd of 54 cows of the Guernsey, Holstein, Jersey and Ayrshire breeds over a period of sixteen months. Samples were taken at monthly intervals from each cow. Each quarter was sampled separately by first washing the udder with a chlorine disinfectant solution, then discarding the first four or five streams after which a 25 cc. sample was drawn into a sterile test tube for examination.

The tests compared in this work were the brom thymol blue test, the catalase test, the cell count, the long-chain streptococci count and observation and examination of the udder.

The brom thymol blue test was made according to Mills (9) procedure by mixing 5 cc. of milk and 1 cc. of brom thymol blue solution. The brom thymol blue solution was prepared by dissolving 0.4 gram of brom thymol blue in 50 cc. of 95 per cent alcohol and making the volume up to 200 cc. by the addition of distilled water. This solution was adjusted to a pH of 6.8 to 7.0 by the addition of N/10 NaOH.

The results of the test were recorded as follows: greenish yellow — normal; light green — suspicious; and green to blue green — abnormal, according to the color developed in the serum. It was found that chilling the samples to cause separation of the cream from the milk serum made reading much easier. This removed the effect of the fat which caused high fat samples to appear relatively lighter in color than a low fat sample of the same pH.

The leucocytes and long-chain streptococci were counted by the Breed method (13). At the beginning of this work counts were made of the fresh milk and also of the same samples after incubation at 37 degrees C. for 12 hours. The results, however, indicated that only a very few of the fresh samples contained enough long-chain streptococci to give significant results. The count of fresh samples was therefore abandoned and only the counts of incubated samples were included in this report.

Results of the examination for leucocytes were recorded as

- when less than 500,000 cells per cc. were noted. Samples showing from 500,000 to 2,000,000 were noted as + and over 2,000,000 as ++. This simple classification was based upon the results of numerous investigations which seemed to indicate a division between the normal and abnormal samples at about 500,000 with the more acute cases above 2,000,000 cells per cc.

The presence of long-chain streptococci was indicated by an S while their absence was indicated by the - sign. This seemed logical on the incubated samples because, after incubation, samples containing streptococci were easily differentiated from those free from mastitis streptococci.

The catalase test was performed by mixing 15 cc. of milk with 5 cc. of 1 per cent Hydrogen Peroxide solution in a Smith Fermentation tube. After 2 hours incubation at 37° C. the number of cubic centimeters of gas collected in the closed arm was recorded. Samples showing more than 1.5 cc. were considered abnormal.

When samples were taken notations were made of the occurrence of stringy or thick milk, inflammation or hard lumps in the udder and any other abnormal condition easily noted by the milker. These observations were made and compared with the results of laboratory tests to show what correlation existed between such an examination and the laboratory tests.

Another common method in use is the strip cup. It is composed of a metal cup of 12 to 16 oz. capacity over which is placed a black cloth or a fine screen. When the first streams of milk are drawn from each quarter the presence of flakes or clots can be detected. Although the strip cup was not included in this report it was used on the same individuals tested by other methods. Observations on the strip cup test compared with the appearance of the milk in glass sample tubes, i.e. when flakes or clots showed on the strip cup there was a sediment or other abnormal physical condition noticeable in the milk in the sample tube.

Discussion of Results

A comparison is made in Table 1 of the various tests used to show abnormal milk. This group of 14 cows was recognized as definitely infected with mastitis, basing the classification on the results of repeated tests including the microscopic exami-

nation for long-chain streptococci and the appearance at some time during the testing period of milk which was abnormal in consistency. The table is presented to show the frequency with which abnormal milk occurs as shown by chemical and bacteriological tests.

In two cases, Cows 109 and 215 in which the notation "staph." is used in the table, the milk failed to show long-chain streptococci during the testing period. Pus cells and staphylococci were found on microscopic examination. The quarters thus infected finally ceased to secrete milk and became blind or dry. These two cases were included in the tabulation as equivalent to finding the long-chain streptococci as the causative organisms.

Tests made during the month at the beginning and at the end of the lactation were not considered because of the occurrence of milk which is naturally abnormal at these periods.

The data in Table 1 show that 73.5, 77.8 and 79.0 per cent of the milk samples were abnormal to the brom thymol blue test, catalase test and leucocyte count respectively. In this table the results of the tests were considered as either positive or negative indications and the various degrees of reaction were not considered. There does not appear to be much choice between the three methods as indicators of infected udders. However, the cell or leucocyte count shows some advantage with 79.0 per cent of the samples detected as abnormal. The catalase and brom thymol blue tests can be ranked next in order with 77.8 and 73.5 per cent respectively detected as abnormal.

Long-chain streptococci occurred in only 46.3 per cent of the samples, indicating that this test alone could not be used as a means of detecting mastitis unless repeated testing was the rule. The appearance of the long-chain streptococci usually occurred immediately before stringy or clotty milk was observed. The other tests used regularly showed very abnormal milk on these instances.

It was noted in a number of cases that only one quarter of an udder would show evidence of abnormal milk while the other three showed normal on repeated tests. This would indicate that the testing should be done on quarter samples and that it would not be necessary to condemn the entire produc-

TABLE 1.—Occurrence of Abnormal Milk as Indicated by Various Tests.*

Cow No.	No. of tests	BTB positive	Cell count over 500,000	Catalase over 1.5 cc.	Long-chain streptococci	Physical appearance
101	9	8	7	7	1	sediment and pus 2 quarters
107	10	6	7	9	1 staph	pus in one quarter
215	12	4	12	8	4	sediment in one quarter
217	12	9	11	11	9	sediment in one quarter
272	6	5	4	4	1	yellow, curdy one quarter
285	14	8	5	9	1 staph	thick, yellow one quarter
337	14	11	14	13	9	occasional clots occasionally
339	11	9	9	10	8	flaky sediment
345	11	8	8	9	1	one quarter thick, yellow
349	12	11	11	8	11	one quarter sediment all
355	14	9	12	13	8	quarters clots in
360	12	9	11	9	2	two quarters clots, sediment
4A	12	11	9	9	4	one quarter clots and serous
4B	13	9	8	7	5	fluid, one quarter
total	162	119	128	126	75	
Per cent		73.5	79.0	77.8	46.3	

* The positive tests recorded in this table indicate positive tests in one or more quarters.

tion of such an individual. The results of tests made on individual quarters are shown in Table 2.

During the course of the experiment none of the cows included in Table 2 gave milk which was abnormal in physical consistency. The tests indicated chronic infections in which long-chain streptococci were present in most cases and the milk was abnormal to the leucocyte count, brom thymol blue and catalase tests.

A larger number, 46.7 per cent, of these samples showed abnormal by the catalase test than by any other test. Of the 540 quarters tested 252 or 46.7 per cent produced more than 1.5 cc. of gas from 15 cc. of milk. One hundred ninety-two or 35.6 per cent of the 540 quarters gave cell counts of over 500,000 and 174 or 32.2 per cent were positive to the brom thymol blue test.

Only 88 of 540 or 16.2 per cent of the quarters showed long-chain streptococci in the incubated samples. Three of the thirteen cows did not harbor long-chain streptococci in any of the

TABLE 2.—Relation Between Various Tests Made on Quarter Samples From Cows Showing No Physically Abnormal Milk

Cow Number	Number of tests	B T B positive	Cell count over 500,000	Catalase over 1.5	Long-chain Streptococci
103	56	14	22	29	9
106	36	15	20	29	7
222	44	10	8	11	4
223	20	5	5	6	0
270	36	6	15	17	7
273	48	22	22	26	14
264	48	22	19	27	0
346	40	14	12	18	16
353	36	5	10	5	9
357	52	23	13	15	0
361	44	18	13	23	13
365	40	5	15	15	7
112	40	15	18	31	2
No. of Quarters	540	174	192	252	88
Per cent Positive		32.2	35.6	46.7	16.3

samples taken, although these samples were decidedly abnormal to the other tests used. This may be explained by the fact that the long-chain streptococci are not always present even though the udder is infected and has undergone physical change. The infection goes through various stages and it is only at certain times that the organisms are eliminated in the milk in numbers sufficient to isolate them by the method used in this work. The same irregular occurrence of abnormal milk is noted by the other tests.

It may be concluded that the presence of long-chain streptococci and positive chemical tests can be taken as positive evidence of infection although negative tests do not necessarily indicate the absence of infection.

Conclusions

1. Laboratory tests are a valuable aid in detecting cases of chronic mastitis which would pass unnoticed by the milkers.
2. Because of the mild and recurrent nature of the infection the tests must be used regularly to detect and eliminate dangerous sources of infection.
3. Infection and the occurrence of abnormal milk is frequently confined to a single quarter of an udder. That is, the cow may produce normal milk from the three quarters even though the one quarter is badly infected. It is important that these infected quarters be detected so that the milk may be destroyed and kept out of the regular supply and also that it be kept from coming in contact with the healthy udders.

4. The brom thymol blue test, the catalase test and the cell count all appear to detect abnormal milk effectively. The brom thymol blue test, however, is simpler as a control measure and can be recommended for use in the control of mastitis in the milking herd.

5. Microscopic examination of stained smears is not a good routine method of detecting "mastitis" milk when used alone because a relatively low percentage of the samples from abnormal udders contain long-chain streptococci.

6. Positive brom thymol blue, catalase tests and high cell count do not definitely prove mastitis infection but the tests are an indication of abnormal milk which is at least presumptive evidence of an abnormal udder condition.

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