# STUDIES ON BOVINE MASTITIS

By

Thomas Hugh Prescott III and Jane Huffman

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Approved

Dean

Approved

Head of the Department
of Biology

Approved

Course Adviser

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# I

## INTRODUCTION

Mastitis, commonly known as garget, is considered by animal health departments as one of the most important of all the bovine diseases. It is of importance to the dairyman, as an economic problem, and to the public health department as a source of disease in the human family.

Cases of septic sore throat, caused by a hemolytic streptococcus which is often found in mammary glands affected with mastitis, have been traced to cows showing symptoms of mastitis. However, modern dairy barn construction along with everyday practice of cleanliness and sanitation tend to minimize the danger of such infection. Isolation of clinical cases of mastitis and the pasteurization of a large percentage of the milk are also of great value.

The significance of the disease lies in the economic loss to the dairyman; not only are cows showing clinical symptoms of mastitis removed from the milking line for the duration of the attack, but there is also danger of infecting the entire herd from such carriers of the disease. In acute cases, if the cow survives she frequently looses one or more quarters of the mammary gland.

At the annual Veterinary Conference of the New York Veterinary College, Holfort (41), the chief veterinarian of Borden's, found that of 1,330,000 cows in the state of New York, 57,190 are eliminated from milk production annually due to mastitis. This figure represents a loss of 3,236 million pounds of milk or about 49 million dollars per annum. The total loss of cows and milk would be, according to these figures, approximately 72 million dollars a year. Such a loss is colossal and can only be explained with the ideal in view of the cow as the perfect milk producing machine.

The New York Farm Bureau (34) expressed the opinion that the detection and eventual elimination of cows affected with either chronic or acute mastitis in the advanced form, from dairy herds, is highly desirable and will pay good profits to the dairy farmer. This committee recognized that the condemnation of infected cows without some knowledge of prophylactic measures is ineffective. The three steps suggested by them are first, education, then research and finally, regulation by law.

# II

## REVIEW OF MASTITIS

The incidence of mastitis is evidently on the increase, and at the same time steadily gaining importance as evidenced by the experimental work reported. Rosell (41) summarized the prevalence of mastitis in many countries by quoting statistics tabulated by important investigators. In Germany and Austria the loses were estimated to be one-third of the annual milk production. Udall stated that in all probability the losses from mastitis exceeds that of tuberculosis or abortion. M. Porcher director of the French Agricultural Journal (1931) writes: "the healthy cow is actually the exception, since the infected cow is almost the rule". Steck (48) stated that a latent infection is present in the udders of all normal milk cows.

A careful review of the literature on the subject showed that a large percentage of all cows tested were found to be infected. In most of the infected cows, the disease was of a chronic nature. However, milk from cows infected with chronic mastitis may change both in appearance and reaction with the progress of the inflammation and the kind of organism responsible for the disease (17).

Stader (46) stated that the predisposing factors which lower the resistance of individuals seered to be of major importance and that it was highly probable that the organisms gained access to the gland tissues by the way of the blood stream. Most investigators, however, consider the hands of the milker, milking machines, contaminated bedding or generally unsanitary conditions as the principle mode of transmission. Artificial inoculations have failed to produce the disease when given per orum, injected into the blood stream, or subcutaneously, except in a few instances (31). Injections into the teat canal has produced the disease in several instances (31, 12). Jones and Little (1934) in a paper presented at the Twelfth International Veterinary Congress (28), stated that in a series of inoculations into the mammary gland; the first inoculations were uniformly negative, subsequent inoculations with approximately the same number of streptococci, produced one or the other of two types of mastitis, either acute or sub-acute, both being characterized by destruction of the glandular tissue. These investigators were of the opinion that the first injection sensitized the gland with the result that the subsequent injections produced clinical mastitis.

Derrick (10) stated that the transmission of the disease seemed to be by infected milk contaminating the end of the teat of a clean quarter and migrating through the teat canal. Christiansen and Nielsen (8) stated that under natural conditions there must be some factors as yet unknown, which govern the entrance of bacteria by way of the teat canal.

Nocard and Mollereau (1884-1885) isolated a long chain streptococcus from the milk of cows infected with mastitis. Since that time streptococcus mastiditis has been recognized as a constant etiological factor in mastitis (46).

Hansen and Hucker (21) have classified mastitis into four groups according to the nature of the infection, and stated that it is probable that different types of bacteria are responsible for each. Other investigators have classified the strains of streptococci found in mastitis infected mammary glands (36, 42, 40, 22, 11, 55, 32, 38, 37].

Investigators are uniformly of the opinion that the hemolytic streptococci are of the greatest importance; streptococcus epidemicus (40, 19, 20), staphlococci, E. coli, B. pyogenes and other organisms are of less importance as the causative organisms of mastitis.

Acute cases of mastitis are usually accompanied by loss of appetite, temperature, congestion of the gland, tenderness, redness and swelling of the gland. There is usually almost a complete cessation of milk flow. The milk that can be obtained is watery or flaky . It may be stringy with the presence of large clumps, which can be removed from the teat with difficulty. The skin over the gland sometimes becomes very dark and may show evidence of necrosis. Physical examination in chronic mastitis shows the mammary gland to be lumpy or indurated and also it may be asymmetrical. In chronic mastitis the symptoms are less acute, except at periods of intensity or "flare-ups". Kingman (29) stated that chills, rough coat, arched back, loss of appetite, temperature and stiffness of gait are the clinical symptoms of acute mastitis.

Meyers (30) recommended a county dairy and milk inspector, who is a veterinarian, to inspect all dairies. This inspector should be familiar with the various physical characteristics of mastitis, and should make the inspection equal for all of the dairies of the state. Most investigators recommend sanitation, removal of contaminated bedding and extreme cleanliness of the barn as the best method of control. Stader (46)

stated that sanitation is not necessary since the organism is already present and the disease is due to some predisposing factor. Udall and Johnson (53) stated that sanitation in the barn and precautions while milking are the most effective preventatives. Several investigators recommend frequent milking and application of heat to infected quarters, along with isolation of infected cows as the best means of control (52, 16, 5, 6).

Udall and Johnson (54) in a recent experiment have been able to control mastitis in a herd by placing all cows in one of four groups and milking them in order of degree of infection. Seeleman and Hadenfeldt (44) have in no case found that separation alone halts the spread of the disease. Steck (50) in a paper presented at the Twelfth International Veterinary Congress stated that the control of mastitis can be rendered effective by the combined application of three procedures; elimination, separation of all infected animals and chemotherapy. Plastridge, Anderson, White and Rettger (37) and Hucker (26) have recommended similar plans for the control of the disease.

Minnett (32) stated that chronic streptococcus mastitis can be controlled by systematically milking infected cows last.

Autogenous vaccines have been used by several investigators. Seddon and Rose in Australia (45) showed that vaccination with autogenous vaccine was of great value as a preventative measure. Frost (13) Bryan (1) DeCamp (9) and Stader (47) have also found that autogenous vaccines halt the spread of the disease in a herd. Bryan (3) stated that the spread of the disease in a herd was halted by use of lacto-vaccine preserved with gentian violet.

Seeleman and Hadenfeldt (44) used entozon infusion as a treatment for mastitis, while Steck (49) has found tryptaflavine to be a good method of treatment for mastitis.

Gildow, Hansen, and Cherrington (15) used ultra violet light and found that it removed clinical symptoms but did not affect the bacteria count, thus the causative agent remained. They found that formalin per os, which was recommended by Frost, gave negative results. Colloidal carbon injected intravenously, which was recommended by Concklin, increased the number of leucocytes, but the bacteria count was not decreased. No consistent results were obtained by the use of an autogenous vaccine. Thus the methods of treatment did not have any effect on the causative agent.

# III

## EXPERIMENTAL PROCEDURE

In a survey of mastitis in the dairy herd of the Virginia Polytechnic Institute, a series of tests, which were recommended by several investigators for the detection of mastitis, were made and the results compiled. Samples from each quarter of fifty-one cows were used in the experiment. The samples were collected in three ounce bottles used only for this purpose. Before each series of samples were collected, the bottles were thoroughly washed, dried and plugged with cotton. They were then placed in an autoclave and sterilized at fifteen pounds pressure for forty minutes. The metal covers were placed on sterile cheese cloth and sterilized with the bottles. The covers were placed on the bottles with sterile forceps before removal from the autoclave. The samples were collected directly after the cows had been washed for the milking line. Each teat was bathed in seventy per-cent alcohol. The metal cap was carefully removed from the bottle and the cotton plug was thrown away. The milk was milked directly into the bottle. Care was taken not to spread any contamination from cows showing clinical mastitis to the remainder of the herd.

## A. FIELD TESTS

### 1. Physical Examination

Physical examination is used in correlation with other tests by most investigators. It is at the present time the test that the Federal government uses, almost exclusively, in their method of detecting animals to be eliminated from the herd, for mastitis (4). Switzer and Gates (51) stated that the physical examination is a very efficient method of detecting mastitis under practical field conditions. Udall and Johnson (53) stated that the value of the physical examination rates above any other single method for the detection of the disease.

Procedure.-- This test was carried out by the senior author. Each quarter of the mammary gland was examined by manual manipulation to find if any induration or fibrosis were present. The symmetry of the gland was noted ; each quarter was lifted in both hands and the consistency, size and weight were compared to the other quarters.

Results.-- Out of a total of 200 quarters examined, 33 or 16.5 per cent of the quarters gave a positive reaction.

## 2. Strip Cup

The strip cup was first developed by Moak (33) as a means of detecting infected quarters. Hucker (24) stated that the presence of flakes in the milk should be interpreted as indicating that a pronounced infection is present. All quarters giving positive reaction have been shown to be infected, although, not all infected quarters gave a positive reaction. The strip cup is used by most dairy farmers producing certified milk. It has a moderately high degree of accuracy in detecting quarters that are in an early as well as in an advanced stage of the disease.

Procedure.-- The strip cup is essentially a metal container over which a 100 to 120 mesh wire screen has been incorporated within a concave cover. Several streams of the fore milk were milked on the screen. The detection of infected quarters was noted by the appearance of clotted, stringy or materated and flaky masses on the screen. The strip cup was washed between each milking.

Results.-- Out of two hundred quarters tested, 11 or 5.5 per cent showed positive reaction and 6 or 3 per cent showed a slight reaction.

### 3. Brom Cresol Purple

This test was recommended by Jensen (27) as more accurate and less expensive than brom thymol blue. The brom cresol purple indication paper used in this experiment was prepared and used in the manner described by Jensen.

In brief, the brom cresol purple impregnated paper was made by dropping brom cresol purple dye (0.1 grams of the powder dissolved in 100 cubic centimeters of distilled water) onto filter paper (Whatman Ashless No. 40) until thoroughly saturated. The paper was dried in an electric oven at a low temperature. When dry this impregnated paper has a jersey-yellow color. The paper was then cut into strips one and one-half inches by one-half inch. One end of four of these filter strips were sealed between two strips of gum sealing paper. The number of the cow was placed on one end of the paper, and the quarters of the cow were designated above each strip of impregnated paper as follows: R F, R R, L F, L R. The papers were kept in an envelope until used, after which they were placed between the leaves of a book. The test was graded 4, 3, 2, or 1 positive or negative according to degree of reaction.

Procedure.-- A small stream of milk, three or four drops, was milked on the brom cresol purple papers; the results were read immediately and the papers were kept for a comparative reading in the laboratory. The indicator papers were kept dry and were protected from the barn atmosphere as much as possible. This test was carried out only on dry days as dampness was found to affect the reaction. A deep purple color indicates a diseased quarter and a grayish tan color indicates normal milk. Varying degrees of purple were judged as partial or slight reaction.

Results.-- Out of a total of 200 quarters examined, 45 or 22.5 per cent of the quarters gave a positive reaction and 66 or 33 percent of the quarters gave a slight or partial reaction, 89 or 44.5 per cent of the quarters were negative. This gives a total of more than half or 55.5 per cent of the quarters showing some degree of reaction.

## B. LABORATORY TESTS

### 1. Chlorine

Rosell (39) stated that the chlorine test discloses abnormalities in the glandular tissue with a high degree of accuracy, and that an increase in chlorine is caused by an inflammatory exudation of serum from the blood. Hucker (25) stated that fibrotic quarters usually secrete milk containing more than 0.14 per cent chlorides, and the presence of this fibrosis probably indicates past or present infection. More than 0.14 per cent chlorides is indicative of an infection.

An abbreviated method of determining the percentage of chlorides in the milk was suggested by Hayden (23).

Procedure.-- Five cubic centimeters of silver nitrate solution (1.3415 grams of silver nitrate in one liter of water) was accurately measured into a test tube. Two drops of a ten per cent chromate solution were added, which produced a brick red color. Exactly one cubic centimeter of the milk to be tested was added. This is a quantative test and is so adjusted that a yellow color develops in the mixture if the concentration of the chlorides is greater than 0.14 per cent. The test was graded 4, 3, 2, or 1 positive or negative according to the degree of reaction.

Results.-- Out of a total of 194 quarters examined, 124 or 64 per cent were positive, 42 or 21.7 per cent were slight or partial and 28 or 14.3 per cent were negative.

## 2. Catalase

Rosell and Miller (43) stated that catalase ranks next to chlorine in efficiency in detecting mastitis. Infection in the mammary gland is accompanied by an increase in leucocytes and proliferation of epithelial cells. This test is based on the fact that cellular tissue contains a certain amount of catalase which breaks down hydrogen peroxide to hydrogen and water (24). The test then becomes a rough index as to the number of cells contained in the milk.

Procedure.-- Ordinary glass slides were painted on one side with black enamel paint. A large drop of the sample to be tested was placed on the mirror surface of the black slide and one drop of 10 per cent watery solution of hydrogen peroxide was added. Infected quarters were noted by the appearance of bubbles in the drop. To aid the detection of bubbles a reading glass was used. This test was graded 4, 3, 2, or 1 positive or negative according to the degree of reaction.

Results.-- Out of a total of 200 quarters tested 39 or 19.5 per cent were positive, and 100 or 50 per cent showed a slight or partial degree of reaction, and 61 or 30.5 per cent were negative.

## 3. pH

Bryan (2) stated that there are normal physiological factors that are responsible for the variation in the pH of freshly drawn milk. It has been found by a number of investigators that an alkaline reaction in milk nearly always indicates an abnormal quarter, although normal pH (6.3-6.6) does not necessarily indicate a normal gland.

Procedure.-- The pH of freshly drawn milk was determined by use of a Hellige hydrogen-ion comparator. A number of samples were checked by a Leeds and Northrup type K potentiometer, to determine the accuracy of the readings by the colormetric method, which was found to be reasonably accurate. Brom thymol blue (0.5 cc.) was used as an indicator, and was added to 9.5 cubic centimeters of neutral distilled water, to which was added 0.5 cubic centimeters of the sample to be tested.

Results.-- Out of a total of 200 quarters examined, 16 or 8 per cent were found to be more alkaline than 6.8. The remainder were within the range generally considered normal.

## 4. Rennet

The rennet test for the detection of mastitis was developed by Hadley (18). This test is reliable only when applied to individual samples of fresh milk. The test is based on the fact that abnormal milk does not coagulate readily, with rennet. This may be due to the fact that the optimum hydrogen-ion concentration is about pH 6.0. As pH 7.0 is approached a measurable destruction of the enzymes occurs rather suddenly (14). The optimum temperature for rennet action is 40 to 42 degrees Centigrade.

Procedure.-- A solution of rennet was prepared by mixing one part of fresh cheese-makers rennet to fifty parts of distilled water. One tenth cubic centimeter of the solution was accurately measured and added to 10 cubic centimeters of the freshly drawn sample. Thus the final dilution was 1:5000. The mixture was thoroughly shaken and allowed to stand at room temperature for one hour. Within this period normal milk will coagulate. Difference in degree of reaction was noted by reading at fifteen minute intervals. This test was carried out at a later date than the other tests, and was made in correlation with a second brom cresol purple test. All milk which coagulated within the sixty minute period was considered to be normal. Milk which did not coagulate within the

hour or on prolonged standing was considered to be abnormal.

Results.-- The total number of quarters tested was 172 and out of this number 15 or about 9 per cent did not coagulate and were considered as abnormal. The results of the brom cresol purple test in comparison with rennet are given in Table II.

## 5. Bacteriological Examination

Bacteriological examination of the milk was made to detect the relative number of bacteria present in the milk of each quarter as well as the causative agent producing mastitis. This test is important because the invasion of streptococci obviously precedes any physical change which may take place in the mammary gland. Thus the relative number of organisms were calculated to determine the purity of the milk.

Procedure.-- Agar slant method: The agar slant method consists essentially in using a loop in measuring the milk and spreading the contents over the surface of a dry agar slant. The following medium was used:

| | |
|---|---|
| Veal Infusion | 1000 cc. |
| Bacto agar | 20 grams. |
| Glucose | 2.5 grams. |

The reaction was adjusted to a pH 7.2.

The slants were allowed to stand in an oblique position until hardened and incubated for sterility. A platinum wire loop measuring 1 mm. in diameter was used to inoculate the slants. A loop of the milk was secured from the sample, on the first opening of the sample bottle, and touched to the surface of the slant in three places. The inoculin was then

spread over the entire surface of the slant. The slants were incubated for forty-eight hours, after which the colonies were counted. The types of colonies were noted.

Veal agar plate method. The following medium was used:

| | |
|---|---|
| Veal infusion | 500 cc. |
| Distilled water | 500 cc. |
| Bacto agar | 20 grams. |
| Peptone | 5 grams. |

The reaction was adjusted to a pH 7.0.

The milk was diluted 1:100 and 1:1000 to insure greater accuracy in counting the colonies. The dilution bottles were six ounce size and were cleaned and sterilized in the same manner as the three ounce sample bottles used for collecting the milk. Distilled water was sterilized in large flasks at 15 pounds pressure for forty minutes. The water was transferred to the dilution bottles with sterile 100 cc. pipettes.

A separate sterile pipette was used to transfer the milk from the sample bottle to the dilution bottle. The dilution of the milk was plated out by the veal agar plate method. The plates were incubated for forty-eight hours and the number of colonies were counted. The colonies that had the characteristic appearance of streptococci were transferred to blood plates.

Blood agar plates.-- Ten cubic centimeters of sterile citrated (5.0 cc of 20 per cent sodium citrate to 100 cc. of blood) horse blood was added to a 50 cubic centimeter flask of melted agar (40 to 45 degrees centigrade) and rotated to insure thorough mixing. The plates were incubated to insure sterility. Typical streptococci colonies were transferred to the blood plates, which were incubated for forty-eight hours before further examination. At the completion of the incubation period the typical streptococci colonies were inoculated into sterile veal peptone broth. Slides were prepared from the broth cultures, which were stained with Grams stain and studied microscopically.

Results.-- Out of the 51 cows used for the experiment, streptococci were isolated from 34 quarters of 21 cows. The majority of the organisms isolated were short chained streptococci of the viridans type. Long chained streptococci were isolated from one of the cows after an attack of acute mastitis. There were no hemolytic streptococci isolated from any of the cows. The type of bacteria encountered were those classified by Hucker as the agent causing sub-chronic mastitis.

## C. Methods of Control

In an attempt to control the spread of mastitis in the herd, an autogenous vaccine was prepared and used. The most severely infected cows were sold or isolated from the herd. Several investigators have obtained fairly successful results from use of an autogenous vaccine, however the results are somewhat confusing due to the fact that no controls were used. In this investigation an autogenous vaccine was prepared in the laboratory from the organisms isolated from the herd. The following organisms were used in the proportions indicated; short chained streptococci, 50 per cent; staphlococcus albus, 25 per cent; and staphlococcus aureus, 25 per cent. The suspension of bacteria was heated to 60 degrees centigrade for one hour to destroy the organisms, and sterility test was made and found to be negative. The vaccine was standardized to tube Number 2 McFarland Nephelometer. The vaccine was inoculated into twenty-five of the fifty cows tested, and the remaining twenty-five were kept as controls.

To determine the efficiency of the autogenous vaccine as a measure of prophylaxis six heifers were included in this portion of the experiment. Three of them were inoculated as were the cows in the milking line and three others were

kept for controls. This gave a total of 28 injected cows and 28 controls.

The cows were selected at random and a series of three injections, 2.0 cc. 5.0 cc. and 10.0 cc. were injected subcutaneously at weekly intervals.

Control measures recommended by many investigators are described in the foregoing part of this paper under the heading Review of Mastitis.

All of the cows on this experiment were examined daily by use of the physical examination and strip cup for symptoms of clinical mastitis. A comparative chart was kept to show the number of outbreaks of clinical mastitis in the vaccinated and control groups. These results are given in Table 3.

A physical examination was made of all the cows tested, after completion of laboratory tests, and they were graded according to the method recommended by the United States Bureau of Animal Industry for mastitis control. Two cows placed in group four were sold, the remainder were isolated and used as nurse cows. The cows that were placed in group three were isolated for breed cows. Groups one and two were placed at the end of the milking line, to be eventually eliminated from the herd.

Acute mastitis may be readily noted by all the cardinal symptoms of inflammation, that is,heat, pain, redness, swelling and cessation or abnormality of glandular secretion.

The diagnosis of sub-acute, chronic and latent mastitis, although not classified as such, require a more careful and systematic examination. The right and left rear quarters are first examined by comparison. They are weighed and palpated simultaneously to better note differences in size, weight and texture or any deviation from the normal. The front quarters are then examined in the same manner.

Following the general examination, each quarter is examined individually. The end of the teat is pulled down with one hand while the teat and gland is carefully palpated for evidence of spiders, tumors, indurated areas and hard nodules suggestive of abscesses. Each quarter is examined in the same manner.

Each animal is classified according to the physical findings as normal, or 1, 2, 3, or 4, depending on degree of abnormality. Animals classified as one and two show only slight evidence of mastitis and are placed at the end of the milking line and milked last. Those classified as number three are

considered as spreaders of the infection and must be isolated where they may be used as nurse cows or they may be sold for slaughter.

Animals classified as number four are not only considered as spreaders of the infection but are economical liabilities to the owner and must therefore be sold for slaughter.

Some animals in groups one and two may overcome the infection and be returned to the milking line. Those in groups three and four are considered incurable and must be permanently removed from contact with the remainder of the herd. All animals with one or more nonfunctioning quarters, when caused by mastitis, are automatically classified as number four.

Three cows died during the course of the examination of the herd, one showing chronic mastitis (No. 14 on the chart) died from metastatic tumors. Two cows, (No. 34 and No. 41 on the chart) died of acute mastitis, one was complicated with septicaemia and the other a gangrenous condition of the gland.

The distribution of the cows in the groups as described above were as follows:

| Group | No. of Cows | Percentage |
|---|---|---|
| 4 | 13 | 26 |
| 3 | 6 | 12 |
| 2 | 6 | 12 |
| 1 | 8 | 16 |
| Normal | 17 | 34 |
| | 50 | 100 |

The above figures alone would indicate an exceedingly high incidence of mastitis, however, the figures do not represent the entire herd but only fifty cows selected at random from it.

The sanitary conditions under which these animals are maintained is exceedingly good. The barn is well lighted and ventilated, has cement floors and metal stanchions. The barn is kept clean and the cows are washed before each milking. A DeLavel Combine Milker is used.

# DISCUSSION

## Strip Cup

The strip cup is very valuable to the herdsman who desires to follow his herd closely. He should use the strip cup before each milking, especially if a milking machine is used. Cows showing abnormalities in their milk should be placed at the end of the milking line and milked last or better still to isolate them from the remainder of the herd. The strip cup is the least sensitive of all the tests now available and also one of the most practical since its use requires no special training on the part of the operator.

It is most essential that the strip cup be used before each milking because abnormalities in the milk may not be apparent at every milking. With routine use of the strip cup these abnormalities will eventually be detected. The appearance of flakes or clotted material is the result of a glandular disturbance.

In this investigation the strip cup along with physical examination was found to be the most accurate method of detecting mastitis.

## Brom Cresol Purple

Brom cresol purple impregnated paper was found to be a fairly accurate method of detecting mastitis under ideal conditions, that is out in the open, free from barn atmosphere. On one occasion it was found that the indicator papers, on exposure to barn atmosphere, changed color indicating alkalinity, before the milk was added. This can be explained by the fact that on this occasion the humidity was high and there was noticeable ammonia in the barn, producing an alkaline atmosphere, thus causing the indicator papers to change color. Brom cresol purple paper has the additional disadvantage in that there is a possibility of inaccurate reading of the variations in color change. The accurate classification of the reaction of individual milk samples as 4, 3, 2, or 1 positive or negative depending upon the presence of slight degrees of color change requires considerable experience, otherwise errors may be made. In other words the personal opinion of the recorder enters into the results thus rendering the test inaccurate.

## Chlorine

The chlorine test, which is based on the fact that infected glandular tissue allows the salts of the blood plasma to filter through into freshly secreted milk, is exceedingly sensitive. This test may be accurate if carried out properly but is so delicate that many unrelated conditions of the mammary gland may give positive results. The test can only be carried out successfully in the laboratory by skilled and experienced workers.

Here again the personal opinion problem involves the classification of the results. There is a color range from a brick red color, to a deep yellow according to the degree of infection.

In this investigation, as our results show, the percentage of cows showing positive reactions to the chloride test is far too high. This may be explained by the fact that the test is much too sensitive and a positive reaction may only be an indication of some unrelated condition or the reaction may be an indication of past infection.

## Catalase

The catalase test, devised to determine the approximate proliferation of cells in the milk, was found to compare favorably with the other tests, especially the chlorine test. From a practical standpoint it is probably not feasible to use positive results secured by this test for the elimination of animals from the dairy herd. This test is too delicate to be used as the sole basis for the elimination or isolation of animals. The test is probably best used in connection with other tests.

Here again the reading of the test involves the personal opinion of the examiner. The grade of reaction determines the class the cow is to be placed in. The difficulty encountered in determining the number of bubbles that is necessary to place an animal in a certain class complicates the test and renders it inaccurate.

The catalase test is primarily a laboratory test and is not adaptable to routine use by an unskilled herdsman.

## pH

The determination of pH is only adaptable to the laboratory where special equipment is available. For determination of the approximate pH, brom cresol purple or brom thymol blue are better adapted for general routine examination by the herdsman. The determination of pH by laboratory methods is only of value to the research worker.

## Rennet

The rennet test was found to be fairly accurate in the detection of infected cows. No satisfactory method for the determination of degrees of infection was derived, although it is not improbable that the scale used in this investigation came within the range of detecting infected animals, as compared with the other tests. This test could be used to advantage in the education of dairymen as to the importance of mastitis. Due to the complicated procedure of many of the tests they would have little practical significance to the average dairyman. However, most dairymen are familiar with the ordinary process of cheese making and the rennet test would have more significance and practical application for them.

As can be seen by inspection of Table II, rennet and brom cresol purple have a very marked correlation, and are close enough to be well within the range of experimental error. Therefore one could deduce from such information, that the range selected for the grading of the rennet test is comparable with the other tests used in this investigation.

## Bacteriological Examination

Due to the fact that streptococcic mastitis is caused by the invasion of streptococci into the mammary gland, it is important to determine the approximate number and type of bacteria present in the gland. However, the results obtained from a bacteriological examination of the milk are difficult to interpret since little information is available as to the exact significance of various types of bacteria present. Until more data is available on the procedure for classifying bacteria as to their etiological importance, it is advisable to use extreme precautionary methods in the handling of cows shedding large numbers of streptococci. Hemolytic streptococci are of importance as a potential source of danger to man when isolated from the mammary gland of cows. Many investigators consider long chained streptococci of more etiological significance than short chained streptococci. This is not supported in the present study.

Bacteriological examination presents certain obvious difficulties such as the large amount of labor and expense involved in the collecting of samples and necessary laboratory routine as well as the comparative obscurity of the results.

A cow which frequently sheds large numbers of streptococci in the milk should be eliminated from the herd since such cows not only raise the bacteria count and thus lower the grade of the milk, but also may be potential sources of infection to the remainder of the herd.

Bacteriological examination is of little value unless accompanied by physical examination and a case history of the animal. If fibrotic or indurated tissues are present it is reasonably safe to assume that the results obtained in a bacteriological examination are not a true indication of the normal flora of milk.

A cow which is positive to any one or more of the tests for mastitis infection and at the same time sheds a large number of organisms in the milk should be considered a source of danger and should be eliminated or isolated from the herd. Such a procedure is justifiable as a precautionary measure until the exact manner of infection is determined.

The period of lactation and the stage of lactation should be considered in interpreting the results of laboratory tests of milk in the dairy herd.

## Control Measures

Several investigators have reported some measure of success in the use of autogenous vaccine as a method of control of mastitis. In this study the autogenous vaccine was found to have little value as a control measure, since more inoculated animals developed the disease than control animals. One of the inoculated heifers developed mastitis in two quarters soon after freshening and was disposed of. Thus it would seem that the use of an autogenous vaccine is apparently of little value as a prophylactic measure. The conclusions drawn from this phase of the investigation might have been different had the vaccine been used over a longer period of time or had the injections been repeated at regular intervals, over a period of years. However, the vaccine appeared to have little effect and its further use was thought to be of little value in this investigation. The control cows and inoculated cows used in this study were observed for clinical symptoms of mastitis each day, for four months following the inoculations.

During the six weeks interval following the grouping of the cows and isolation of groups, only one cow in the milking line has shown any symptoms of mastitis, while all of the isolated cows have continued to show clinical symptoms of the disease.

# V

## CONCLUSIONS

In this investigation, physical examination along with daily use of the strip cup, and isolation of cows according to degree of infection, were found to be the best method for the control of mastitis. Under ideal conditions, brom cresol purple impregnated paper more nearly approximates the results obtained from physical examination and strip cup in the detection of infected quarters, than the other tests.

The rennet test was found to be the most accurate and least complicated of the laboratory tests. The simplicity of the rennet test makes it useful to the unskilled dairyman.

The remainder of the laboratory tests used in this study are complicated and require laboratory equipment and highly trained personel, which are not available to the average dairyman. They have very little practical significance, except to the research worker.

The results obtained from the use of autogenous vaccine indicate that it is of little or no value in the control of, or the prevention of mastitis.

TABLE I--CORRELATION OF TESTS

| Cow No. | Quarter | Physical Examination | Strip Cup | Brom Cresol Purple | Chlorine | Catalase | pH | Rennet | Bacterial Count | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | | | 1-100 | 1-1000 | Slope |
| 1 | RF | - | - | + | ++++ | ++ | 6.1 | 15 | none | none | none |
| | RR | Injury | - | ++ | - | + | 6.3 | + | 900 | 1000 | 4 |
| | LF | - | - | ++ | + | + | 6.2 | 30 | none | 3000 | none |
| | LR | - | - | ++ | - | + | 6.3 | 30 | 800 | 2400 | 1 |
| 2 | RF | - | - | +++ | - | ++ | 6.3 | 15 | none | 7000 | none |
| | RR | - | Clots | +++ | - | + | 6.2 | 60 | 45200 | 21000 | none |
| | LF | - | - | ++ | - | + | 6.4 | 15 | 1200 | 1000 | none |
| | LR | - | - | ++ | + | + | 6.2 | 30 | 1400 | 1000 | 9 |
| 3 | RF | Indurated | Stringy | ++++ | ++++ | ++++ | 6.3 | 30 | 2200 | 4000 | 4 |
| | RR | Lumpy | Stringy | ++++ | ++++ | ++++ | 6.6 | 60 | 2100 | 2000 | 2 |
| | LF | Indurated | Stringy | ++++ | ++++ | ++++ | 6.5 | + | 3800 | 5000 | none |
| | LR | Lumpy | Stringy | ++++ | ++++ | ++++ | 6.8 | + | 2200 | 3000 | 4 |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 4 | RF | - | - | +++ | ++++ | + | 6.4 | 15 | 300 | none | 2 |
| | RR | - | - | +++ | +++ | + | 6.3 | 30 | 2300 | 5000 | none |
| | LF | - | - | +++ | ++++ | + | 6.4 | 30 | 700 | 3000 | none |
| | LR | - | - | ++ | +++ | + | 6.6 | 15 | 2600 | 55000 | none |
| 5 | RF | - | - | - | ++ | ++++ | 6.4 | 60 | 600 | 5000 | 1 |
| | RR | - | - | - | ++ | ++++ | 6.6 | 30 | 200 | none | 56 |
| | LF | - | - | - | ++ | ++++ | 6.6 | 30 | 900 | 8000 | 4 |
| | LR | - | - | - | ++ | ++++ | 6.8 | + | 800 | 3000 | 4 |
| 6 | RF | - | - | - | ++++ | +++ | 6.6 | 30 | 3000 | none | none |
| | RR | - | - | - | +++ | - | 6.3 | + | 13700 | 38000 | none |
| | LF | - | - | - | +++ | ++ | 6.3 | 30 | none | 1000 | none |
| | LR | - | - | - | +++ | - | 6.3 | + | 300 | 3000 | 2 |
| 7 | RF | - | - | - | ++ | - | 6.2 | 30 | 700 | 1000 | none |
| | RR | - | - | - | ++ | - | 6.2 | 30 | 100 | 3000 | none |
| | LF | - | - | - | ++ | - | 6.2 | 30 | none | 2000 | none |
| | LR | - | - | - | ++ | + | 6.3 | 15 | 7400 | 8000 | 4 |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 8 | RF | - | - | - | +++ | ++++ | 6.2 | 30 | none | 8000 | none |
| | RR | Hard | - | - | +++ | ++++ | 6.2 | 30 | 12100 | 3000 | 1 |
| | LF | - | - | - | + | +++ | 6.3 | 30 | 800 | 6000 | none |
| | LR | - | - | - | +++ | +++ | 6.2 | 30 | 900 | 17000 | none |
| 9 | RF | - | Clots | ++++ | ++++ | ++++ | 6.0 | + | 3700 | 4000 | none |
| | RR | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry |
| | LF | - | - | ++++ | ++++ | ++ | 6.3 | 15 | none | 4000 | none |
| | LR | - | - | ++ | ++++ | - | 6.2 | 15 | 200 | none | none |
| 10 | RF | - | - | ++++ | ++++ | +++ | 7.4 | 15 | 3900 | 23000 | 2 |
| | RR | - | - | ++++ | ++++ | +++ | 7.0 | 30 | 1300 | 1000 | none |
| | LF | Caked | - | ++++ | ++++ | ++ | 6.6 | 30 | * | 4000 | none |
| | LR | Hard | Clots | ++++ | ++++ | ++++ | 6.4 | + | 200 | * | * |
| 11 | RF | - | - | - | ++++ | - | 6.4 | 15 | 1200 | 1000 | none |
| | RR | - | - | - | +++ | + | 6.5 | 15 | 100 | 1000 | 3 |
| | LF | - | Clots | ++++ | ++++ | ++++ | 6.6 | 15 | 2600 | 3000 | 1 |
| | LR | - | - | - | ++ | - | 6.4 | 15 | none | none | none |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 12 | RF | - | - | - | ++++ | - | 6.3 | 15 | none | 2000 | 3 |
| | RR | Light | - | - | ++++ | - | 6.3 | 15 | 100 | none | 1 |
| | LF | - | - | - | ++++ | ++ | 6.2 | 15 | 500 | 4000 | 2 |
| | LR | - | - | - | ++++ | - | 6.4 | 15 | 100 | none | none |
| 13 | RF | - | - | - | ++ | - | 6.4 | 30 | 200 | 5000 | none |
| | RR | - | - | - | ++ | + | 6.4 | 30 | 1200 | 1000 | 1 |
| | LF | - | - | - | +++ | - | 6.6 | 30 | 1100 | 4000 | none |
| | LR | - | - | - | ++++ | - | 6.6 | 30 | 100 | none | 1 |
| 14 | RF | - | - | ++++ | ++++ | ++++ | 7.0 | | 5100 | 11000 | 1 |
| | RR | - | Clots | ++++ | ++++ | ++++ | 6.8 | | 1900 | 3000 | 2 |
| | LF | - | - | ++++ | ++++ | ++++ | 6.8 | | 1100 | none | 1 |
| | LR | - | - | ++++ | ++++ | ++++ | 6.8 | | 2700 | 3000 | none |
| 15 | RF | - | - | - | + | - | 6.3 | 15 | 5800 | 34000 | 7 |
| | RR | Weak | - | - | + | - | 6.2 | 15 | 1100 | 2000 | none |
| | LF | - | - | - | ++ | - | 6.2 | 15 | 300 | 5000 | none |
| | LR | - | - | - | ++ | - | 6.3 | 15 | 900 | 7000 | none |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 16 | RF | - | - | - | - | - | 6.4 | 30 | 1200 | 2000 | none |
| | RR | - | - | - | - | - | 6.3 | 60 | 600 | 1000 | none |
| | LF | - | - | - | - | - | 6.4 | 60 | 2600 | 9000 | none |
| | LR | - | - | - | - | - | 6.4 | 60 | 100 | none | none |
| 17 | RF | - | - | - | - | - | 6.4 | 30 | 300 | 7000 | none |
| | RR | - | - | - | - | + | 6.4 | 30 | 1000 | 1200 | none |
| | LF | - | - | - | - | - | 6.4 | 30 | 2400 | 2000 | none |
| | LR | - | - | - | - | - | 6.4 | 30 | 800 | 5000 | none |
| 18 | RF | - | - | ++ | - | - | 6.4 | 15 | 200 | 3000 | none |
| | RR | Weak | - | ++ | - | - | 6.4 | 30 | none | none | none |
| | LF | - | - | ++ | - | - | 6.4 | 15 | none | 2000 | none |
| | LR | - | - | ++ | - | - | 6.4 | 15 | none | none | none |
| 19 | RF | - | - | ++ | - | + | 6.2 | 30 | 600 | 2000 | none |
| | RR | - | - | ++ | - | - | 6.2 | 30 | none | 1000 | 1 |
| | LF | - | - | ++ | - | - | 6.2 | 15 | 400 | none | none |
| | LR | - | - | ++ | - | - | 6.2 | 15 | 300 | none | none |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 20 | RF | - | - | ++ | - | + | 6.4 | + | 22400 | 17000 | 2 |
| | RR | - | - | ++ | - | + | 6.2 | 30 | none | none | none |
| | LF | - | - | ++ | - | - | 6.4 | 30 | 6500 | 27000 | 3 |
| | LR | Shrunken | - | +++ | - | + | 6.4 | 30 | none | none | none |
| 21 | RF | - | - | ++ | +++ | - | 6.4 | 30 | 300 | 4000 | none |
| | RR | Indurated | - | +++ | +++ | +++ | 6.6 | 30 | 100 | none | none |
| | LF | - | - | +++ | ++++ | + | 6.6 | 30 | 2700 | 4000 | 4 |
| | LR | Indurated | - | ++ | ++ | + | 6.4 | 60 | none | none | none |
| 22 | RF | - | - | + | ++ | - | 6.5 | 15 | 100 | none | none |
| | RR | - | - | + | ++ | - | 6.5 | 15 | none | none | none |
| | LF | - | - | + | ++ | - | 6.5 | 15 | 100 | 1000 | none |
| | LR | - | - | + | ++ | ++ | 6.5 | 15 | 1100 | 1000 | 1 |
| 23 | RF | Indurated | - | ++ | ++ | ++ | 6.6 | 15 | 100 | 1000 | none |
| | RR | Indurated | - | ++ | ++ | + | 6.6 | 15 | none | 1000 | none |
| | LF | - | - | + | ++ | + | 6.4 | 15 | 100 | 2000 | none |
| | LR | - | - | + | ++ | + | 6.6 | 15 | 1000 | 1000 | none |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 24 | RF | - | - | ++ | + | - | 6.5 | 15 | 1300 | 6000 | none |
| | RR | - | - | ++ | + | ++ | 6.4 | 15 | 300 | 2000 | none |
| | LF | - | - | ++ | + | + | 6.4 | 15 | none | none | none |
| | LR | - | - | ++ | ++++ | - | 6.6 | 15 | 300 | 3000 | none |
| 25 | RF | - | - | ++ | ++ | + | 6.4 | 30 | 100 | none | none |
| | RR | - | - | ++ | +++ | + | 6.4 | 30 | none | none | none |
| | LF | - | - | ++ | ++ | + | 6.5 | 15 | 200 | 3000 | none |
| | LR | - | - | ++ | +++ | + | 6.4 | 15 | 300 | none | none |
| 26 | RF | - | - | ++ | ++++ | ++ | 6.6 | | 6700 | 2000 | 4 |
| | RR | - | - | ++ | ++++ | ++ | 6.6 | | 1500 | 10000 | 4 |
| | LF | - | - | ++ | ++++ | +++ | 6.6 | | 1500 | 6000 | 5 |
| | LR | - | - | ++ | +++ | ++ | 6.4 | | 400 | 3000 | none |
| 27 | RF | - | - | - | +++ | + | 6.2 | 30 | 100 | 1000 | none |
| | RR | - | - | - | ++ | + | 6.2 | 30 | 12700 | 8000 | 2 |
| | LF | - | - | - | +++ | ++++ | 6.2 | 30 | 300 | none | none |
| | LR | - | - | - | ++ | + | 6.2 | 30 | 1000 | none | none |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 28 | RF | - | - | - | ++++ | + | 6.2 | 15 | 1000 | none | none |
| | RR | - | - | - | ++++ | - | 6.2 | 15 | none | 2000 | 1 |
| | LF | - | - | - | ++++ | + | 6.2 | 15 | 300 | 1000 | 10 |
| | LR | - | - | - | ++++ | - | 6.3 | 15 | 300 | 1000 | none |
| 29 | RF | - | - | ++++ | ++++ | ++ | 6.6 | | 1300 | 5000 | none |
| | RR | - | - | ++ | ++++ | + | 6.4 | | 3000 | 5000 | 3 |
| | LF | - | - | +++ | ++++ | ++ | 6.6 | | 5000 | 7000 | none |
| | LR | - | - | ++ | ++++ | + | 6.2 | | 100 | none | none |
| 30 | RF | - | - | - | ++++ | - | 6.2 | 30 | 1600 | 3000 | none |
| | RR | Atrophied | Flakes | ++++ | ++++ | +++ | 6.4 | 15 | 36600 | 40000 | 7 |
| | LF | - | - | ++++ | ++++ | ++ | 6.2 | 15 | 300 | none | 3 |
| | LR | - | - | - | +++ | - | 6.2 | 15 | 600 | 2000 | none |
| 31 | RF | - | - | - | ++++ | - | 6.2 | 45 | 1400 | 1000 | none |
| | RR | - | - | - | ++++ | + | 6.0 | 45 | 300 | 3000 | none |
| | LF | - | - | - | ++++ | + | 6.2 | 45 | 100 | 1000 | none |
| | LR | - | - | - | ++++ | - | 6.2 | 45 | none | 2000 | none |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 32 | RF | - | - | - | ++++ | + | 6.0 | 15 | 17800 | 15000 | 13 |
| | RR | - | - | - | ++ | - | 6.0 | 15 | 3300 | 103000 | 1 |
| | LF | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry |
| | LR | - | - | - | ++++ | - | 6.0 | 15 | * | 28000 | 5 |
| 33 | RF | - | - | - | ++ | - | 6.2 | 30 | 2400 | 31000 | 25 |
| | RR | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry |
| | LF | - | - | - | ++++ | - | 6.2 | 30 | 3900 | 5000 | none |
| | LR | - | - | +++ | ++++ | +++ | 6.4 | + | 49600 | 152000 | 14 |
| 34 | RF | - | Bloody | +++ | ++++ | ++++ | | | * | * | * |
| | RR | - | Bloody | +++ | ++++ | ++++ | | | * | * | * |
| | LF | Caked | Bloody | ++++ | | ++++ | | | * | * | * |
| | LR | Caked | Bloody | ++++ | | ++++ | | | * | * | * |
| 35 | RF | - | - | - | ++++ | ++ | 6.4 | 15 | 5000 | 8000 | 6 |
| | RR | Light | - | - | ++++ | + | 6.6 | 15 | none | none | none |
| | LF | - | - | - | ++++ | ++ | 6.6 | + | 3200 | 5000 | 5 |
| | LR | - | - | - | ++++ | + | 6.4 | 15 | 100 | none | none |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 36 | RF | - | - | - | ++++ | ++ | 6.4 | | 100 | none | none |
| | RR | - | - | - | ++++ | ++ | 6.6 | | 100 | 1000 | none |
| | LF | - | - | - | ++++ | ++ | 6.6 | | 400 | 5000 | 2 |
| | LR | - | - | - | ++++ | + | 6.4 | | 4200 | 3000 | 1 |
| 37 | RF | - | - | - | ++++ | + | 6.5 | 15 | 200 | none | 1 |
| | RR | - | - | - | ++++ | + | 6.5 | 15 | 600 | 2000 | none |
| | LF | Light | - | - | +++ | + | 6.5 | 15 | 3700 | 9000 | none |
| | LR | - | - | - | ++++ | + | 6.5 | 15 | 800 | 2000 | none |
| 38 | RF | - | - | - | ++++ | + | 6.8 | | none | none | none |
| | RR | - | - | - | ++++ | + | 6.6 | | 2700 | 5000 | none |
| | LF | - | - | - | ++++ | + | 6.6 | | 700 | none | 1 |
| | LR | - | - | - | ++++ | + | 6.4 | | 500 | none | none |
| 39 | RF | - | - | - | ++++ | ++ | 6.6 | 15 | 900 | 5000 | none |
| | RR | - | - | - | +++ | + | 6.4 | 15 | none | none | none |
| | LF | - | - | - | ++++ | + | 6.4 | 15 | 200 | none | none |
| | LR | - | - | - | +++ | + | 6.4 | 15 | 400 | 1000 | none |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 40 | RF | - | - | ++ | ++++ | ++ | 6.6 | 30 | * | * | 13 |
| | RR | - | - | ++ | ++++ | + | 6.6 | 30 | 17000 | 38000 | 9 |
| | LF | - | - | ++ | ++++ | + | 6.6 | 30 | 15000 | 51000 | 10 |
| | LR | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry | Dry |
| 41 | RF | - | - | ++ | ++++ | + | 6.4 | | 100 | none | 5 |
| | RR | Weak | - | +++ | ++++ | +++ | 6.6 | | 3000 | 7000 | 1 |
| | LF | - | - | ++ | ++++ | + | 6.4 | | 70000 | 145000 | 23 |
| | LR | - | - | ++ | ++++ | + | 6.4 | | 9500 | 3000 | none |
| 42 | RF | - | - | - | +++ | + | 6.2 | 60 | 100 | 1000 | 1 |
| | RR | - | - | - | +++ | - | 6.2 | 60 | none | none | 1 |
| | LF | - | - | - | ++++ | - | 6.4 | 30 | 400 | 6000 | none |
| | LR | - | - | - | ++++ | + | 6.3 | 30 | 300 | none | 2 |
| 43 | RF | Hard | Watery | ++++ | ++++ | ++++ | 6.8 | 15 | 200 | none | 20 |
| | RR | - | - | ++ | ++ | ++ | 6.6 | 15 | 500 | 1000 | none |
| | LF | - | - | ++ | ++ | ++ | 6.4 | 15 | none | 1000 | 1 |
| | LR | - | - | ++ | ++ | ++ | 6.4 | 15 | none | 1000 | 1 |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| 44 | RF | Hard | - | - | +++ | + | 6.4 | 15 | 600 | none | none |
| | RR | Hard | - | - | ++ | + | 6.4 | 15 | 100 | none | 1 |
| | LF | Hard | - | - | ++ | ++ | 6.4 | 15 | 200 | 1000 | 1 |
| | LR | Hard | - | - | +++ | +++ | 6.4 | 15 | none | none | none |
| 45 | RF | - | - | ++++ | +++ | +++ | 6.7 | 15 | none | none | none |
| | RR | Lumpy | - | ++ | ++++ | ++++ | 6.4 | + | 500 | none | * |
| | LF | Lumpy | - | ++++ | ++++ | +++ | 7.4 | + | 2600 | 2000 | 9 |
| | LR | Lumpy | - | ++++ | ++++ | +++ | 6.8 | 30 | 300 | none | 2 |
| 46 | RF | - | - | + | ++++ | ++ | 6.6 | 15 | 600 | 1000 | 2 |
| | RR | - | - | + | +++ | + | 6.5 | 15 | 100 | none | * |
| | LF | - | - | + | ++ | - | 6.6 | 15 | 3000 | 1000 | none |
| | LR | - | - | + | +++ | - | 6.6 | 30 | none | none | 2 |
| 47 | RF | - | - | ++++ | | ++ | 6.6 | 60 | 8600 | 20000 | 18 |
| | RR | - | - | ++ | | ++ | 6.4 | 30 | 1300 | 1000 | none |
| | LF | - | - | ++++ | | ++ | 6.4 | 60 | 3000 | 75000 | * |
| | LR | - | - | ++ | | ++++ | 6.4 | 30 | 200 | 3000 | 2 |

| | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| 48 | RF | Hard | Lumpy | ++++ | ++++ | - | 7.2 | + | 100 | none | none |
| | RR | - | - | +++ | +++ | - | 6.5 | 15 | 400 | 2000 | 1 |
| | LF | - | - | +++ | +++ | + | 6.5 | 15 | * | * | none |
| | LR | - | - | +++ | ++++ | ++ | 6.5 | 60 | 500 | none | none |
| 49 | RF | Light | - | ++ | ++++ | + | 6.8 | 60 | 5000 | 8000 | 23 |
| | RR | - | - | ++ | ++++ | + | 6.7 | 30 | 1500 | 4000 | 17 |
| | LF | Light | - | +++ | ++++ | ++ | 7.1 | + | 6300 | 39000 | 11 |
| | LR | - | - | ++ | ++++ | - | 6.8 | 60 | 100 | 1000 | * |
| 50 | RF | - | - | ++ | ++++ | - | 6.6 | 30 | 10200 | 34000 | 25 |
| | RR | - | - | ++++ | ++++ | + | 6.8 | 60 | 5900 | 22000 | 12 |
| | LF | - | - | ++ | ++++ | - | 6.6 | 60 | 13600 | 52000 | 10 |
| | LR | - | - | ++ | ++++ | + | 6.6 | 60 | 18200 | 69000 | 20 |
| 51 | RF | - | - | ++ | ++ | + | 6.4 | 15 | none | none | none |
| | RR | - | - | - | - | - | 6.3 | 15 | 100 | 1000 | none |
| | LF | - | - | - | - | - | 6.4 | 15 | none | 1000 | none |
| | LR | - | - | - | - | - | 6.3 | 15 | none | none | none |

* Bacteria too numerous to count.

Abbreviations indicate right front, right rear, left front, and left rear quarters, respectively.

TABLE II

COMPARATIVE CHART OF RENNET AND BROM CRESOL PURPLE

| Cow No. | Quarter | Rennet | Brom Cresol Purple | Cow No. | Quarter | Rennet | Brom Cresol Purple |
|---|---|---|---|---|---|---|---|
| 1 | RF | 15 | - | 5 | RF | 60 | +++ |
| | RR | + | ++++ | | RR | 30 | - |
| | LF | 30 | - | | LF | 30 | - |
| | LR | 30 | - | | LR | + | - |
| 2 | RF | 15 | - | 6 | RF | 30 | +++ |
| | RR | 60 | +++ | | RR | + | ++++ |
| | LF | 15 | - | | LF | 30 | ++ |
| | LR | 30 | ++ | | LR | + | ++ |
| 3 | RF | 30 | - | 7 | RF | 30 | - |
| | RR | 60 | - | | RR | 30 | - |
| | LF | + | - | | LF | 30 | - |
| | LR | + | - | | LR | 15 | - |
| 4 | RF | 15 | + | 8 | RF | 30 | +++ |
| | RR | 30 | + | | RR | 30 | ++ |
| | LF | 30 | + | | LF | 30 | ++ |
| | LR | 15 | - | | LR | 30 | ++ |

| | | | |
|---|---|---|---|
| 9 | RF | + | ++++ |
| | RR | Dry | Dry |
| | LF | 15 | ++ |
| | LR | 15 | ++ |
| 10 | RF | 15 | ++ |
| | RR | 30 | ++ |
| | LF | 30 | ++ |
| | LR | Dry | Dry |
| 11 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |
| 12 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |
| 13 | RF | 30 | - |
| | RR | 30 | - |
| | LF | 30 | - |
| | LR | 30 | - |
| 15 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | ++ |
| 16 | RF | 30 | ++ |
| | RR | 60 | ++ |
| | LF | 60 | ++ |
| | LR | 60 | ++ |
| 17 | RF | 30 | - |
| | RR | 30 | ++ |
| | LF | 30 | - |
| | LR | 30 | - |
| 18 | RF | 15 | - |
| | RR | 30 | +++ |
| | LF | 15 | ++ |
| | LR | 15 | - |
| 19 | RF | 30 | +++ |
| | RR | 30 | + |
| | LF | 15 | +++ |
| | LR | 15 | ++++ |

| | | | |
|---|---|---|---|
| 20 | RF | + | +++ |
| | RR | 30 | ++ |
| | LF | 30 | ++ |
| | LR | 30 | ++ |

| | | | |
|---|---|---|---|
| 25 | RF | 30 | ++ |
| | RR | 30 | ++ |
| | LF | 15 | ++ |
| | LR | 15 | ++ |

| | | | |
|---|---|---|---|
| 21 | RF | 30 | ++ |
| | RR | 30 | ++ |
| | LF | 30 | ++ |
| | LR | 60 | ++ |

| | | | |
|---|---|---|---|
| 27 | RF | 30 | ++ |
| | RR | 30 | ++ |
| | LF | 30 | ++ |
| | LR | 30 | ++ |

| | | | |
|---|---|---|---|
| 22 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 28 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 23 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 30 | RF | 30 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 24 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 31 | RF | 45 | ++ |
| | RR | 45 | ++ |
| | LF | 45 | +++ |
| | LR | 45 | ++++ |

| | | | |
|---|---|---|---|
| 32 | RF | 15 | - |
| | RR | 15 | - |
| | LF | Dry | Dry |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 40 | RF | 30 | + |
| | RR | 30 | + |
| | LF | 30 | + |
| | LR | Dry | Dry |

| | | | |
|---|---|---|---|
| 33 | RF | 30 | - |
| | RR | Dry | Dry |
| | LF | 30 | - |
| | LR | + | ++++ |

| | | | |
|---|---|---|---|
| 42 | RF | 60 | - |
| | RR | 60 | - |
| | LF | 30 | - |
| | LR | 30 | - |

| | | | |
|---|---|---|---|
| 35 | RF | 15 | + |
| | RR | 15 | - |
| | LF | + | ++++ |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 43 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 37 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

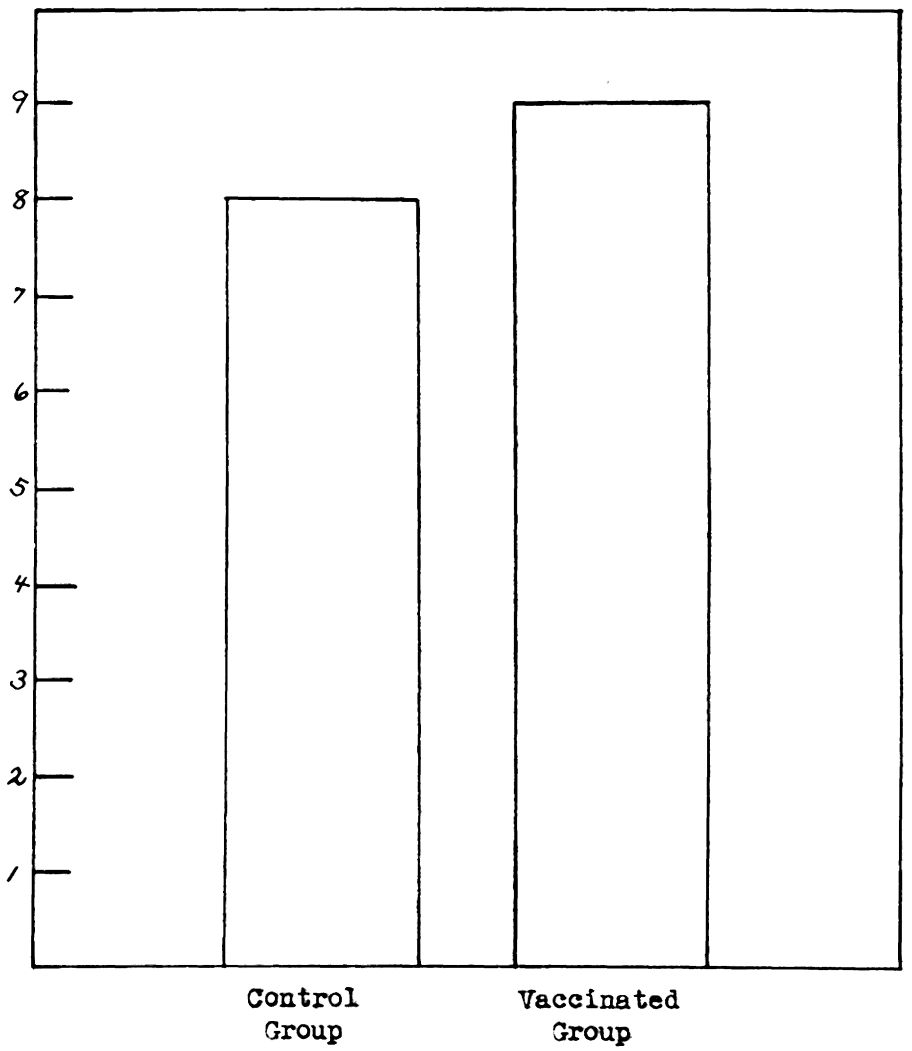
| | | | |
|---|---|---|---|
| 44 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 39 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

| | | | |
|---|---|---|---|
| 45 | RF | 15 | - |
| | RR | + | ++++ |
| | LF | + | ++++ |
| | LR | 30 | - |

| | | | |
|---|---|---|---|
| 46 | RF | 15 | - |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 30 | - |
| 47 | RF | 60 | - |
| | RR | 30 | - |
| | LF | 60 | - |
| | LR | 30 | - |
| 48 | RF | + | ++++ |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 60 | - |
| 49 | RF | 60 | - |
| | RR | 30 | - |
| | LF | + | +++ |
| | LR | 60 | - |
| 50 | RF | 30 | - |
| | RR | 60 | - |
| | LF | 60 | - |
| | LR | 60 | - |
| 51 | RF | 15 | ++ |
| | RR | 15 | - |
| | LF | 15 | - |
| | LR | 15 | - |

TABLE III




COMPARATIVE RESULTS OF VACCINATION

TABLE IV

FINAL GROUPING OF COWS ACCORDING TO PHYSICAL EXAMINATION

| Cow No. | Group | Cow No. | Group | Cow No. | Group |
|---|---|---|---|---|---|
| 1 | 4 | 18 | 4 | 35 | N |
| 2 | 2 | 19 | 1 | 36 | N |
| 3 | 3 | 20 | N | 37 | 2 |
| 4 | 3 | 21 | 4 | 38 | 3 |
| 5 | N | 22 | N | 39 | N |
| 6 | 4 | 23 | 3 | 40 | 4 |
| 7 | N | 24 | 1 | 41 | 4 |
| 8 | 3 | 25 | N | 42 | 2 |
| 9 | 4 | 26 | N | 43 | 3 |
| 10 | 4 | 27 | N | 44 | N |
| 11 | 2 | 28 | N | 45 | 4 |
| 12 | N | 29 | N | 46 | N |
| 13 | N | 30 | 1 | 47 | N |
| 14 | 4 | 31 | N | 48 | 4 |
| 15 | 2 | 32 | 1 | 49 | N |
| 16 | 1 | 33 | 4 | 50 | 1 |
| 17 | 2 | 34 | 4 | 51 | N |

N indicates normal.
Figures 1, 2, 3, and 4, correspond to Groups I, II, III, and IV.

# VI

## DESCRIPTION OF PLATES

### Plate I

A cow showing angular shaped mammary gland caused by mastitis.

### Plate II

A close-up view of cow showing enlargement of front quarter, caused by mastitis.

### Plate III

An agar plate culture of Streptococci.

### Plate IV

An agar slant culture of Streptococci.

### Plate V

A cross section of a mammary gland showing: 1. Normal area. 2. Beginning necrosis. 3. Necrosis with sloughing of cistern. 4. Enlargement and necrosis of teat canal.

### Plate VI

Kidney from a cow which died from acute mastitis, showing petechial hemorrhages.

Plate I

Plate II

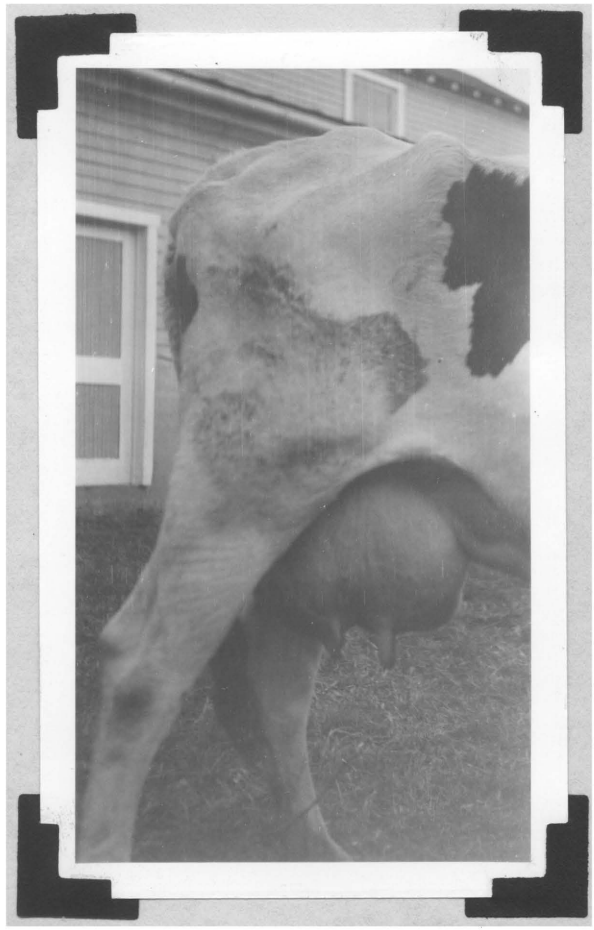


Plate III

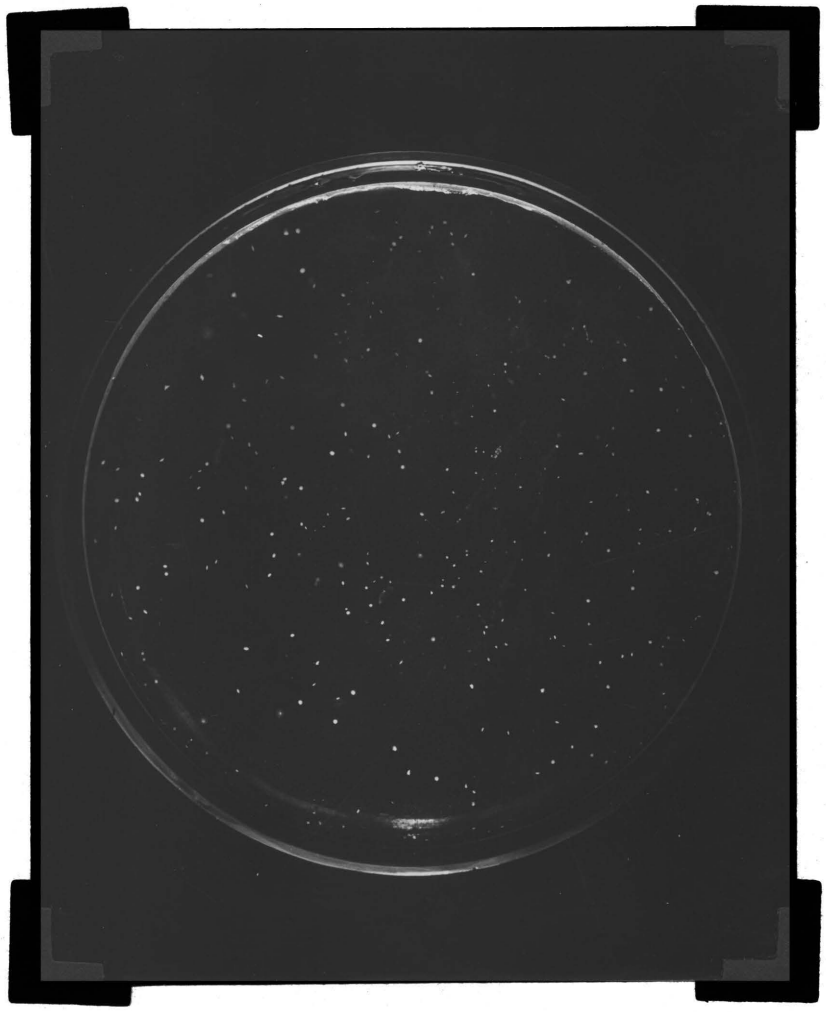


Plate IV

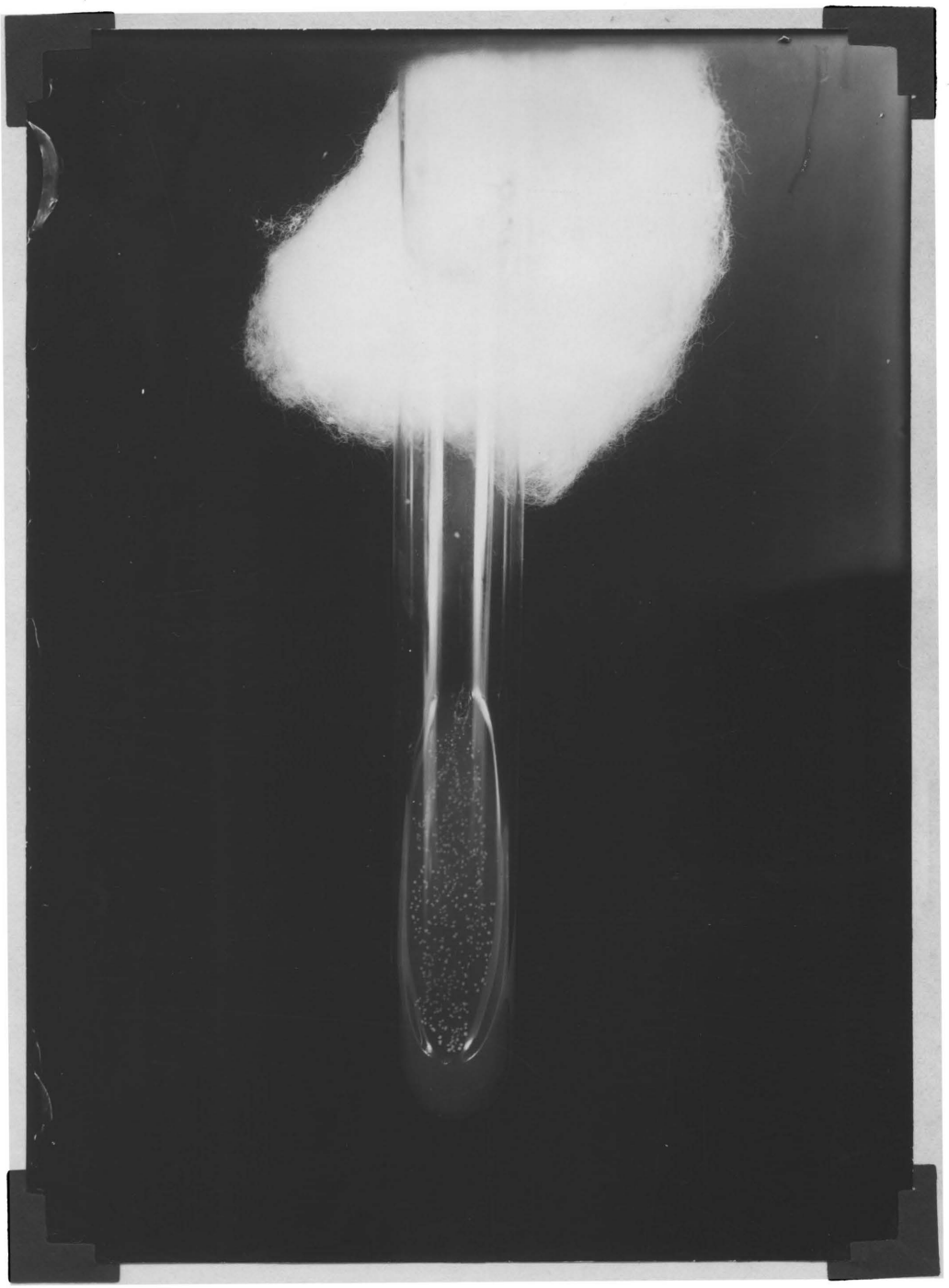


Plate V

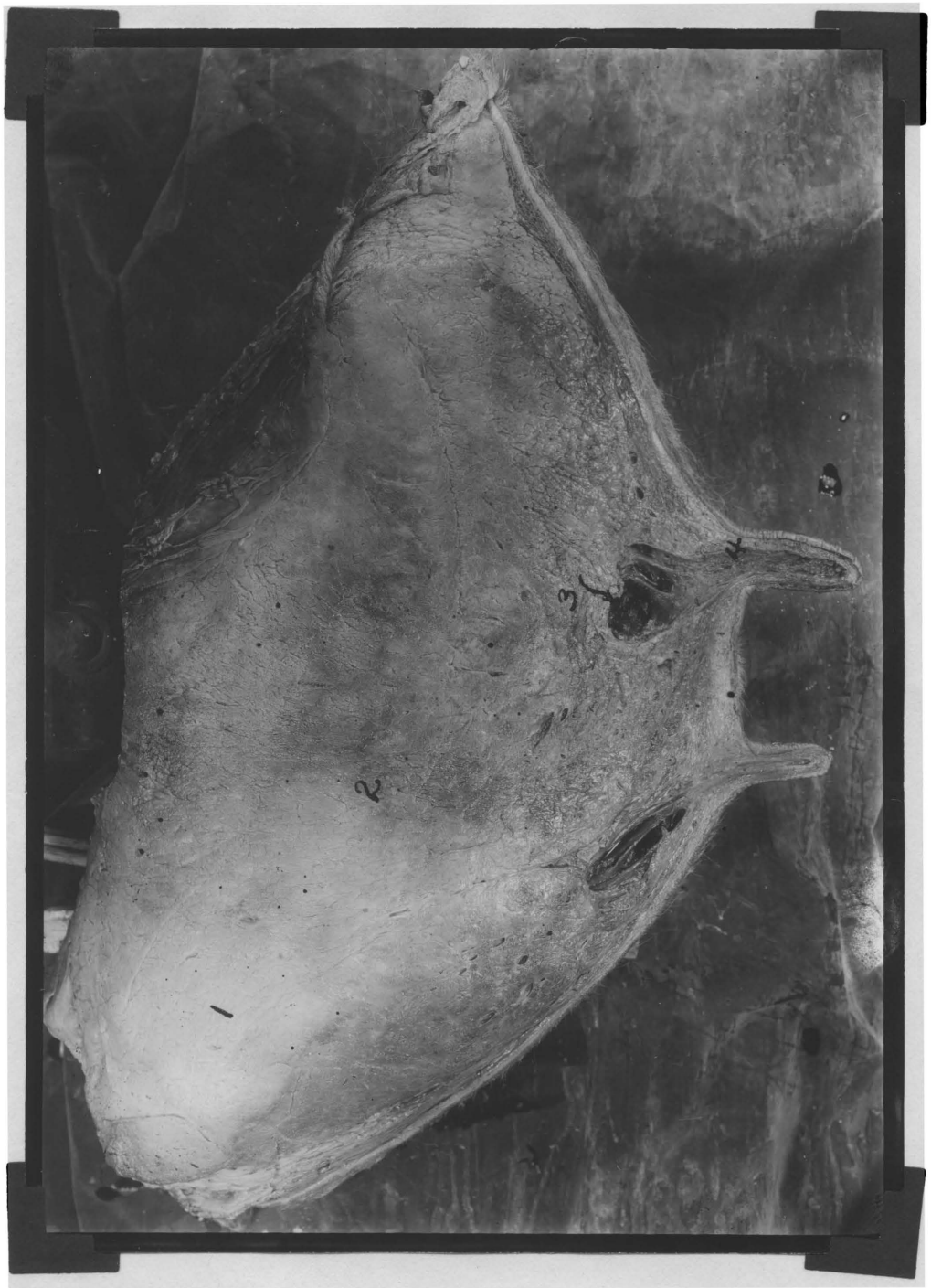


Plate VI

# VII

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