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DEPARTMENT OF THE INTERIOR
BUREAU OF GOVERNMENT LABORATORIES

BIOLOGICAL LABORATORY

I. DOES LATENT OR DORMANT PLAGUE EXIST
WHERE THE DISEASE IS ENDEMIC

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DEPARTMENT OF THE INTERIOR,
BUREAU OF GOVERNMENT LABORATORIES,
OFFICE OF THE SUPERINTENDENT OF LABORATORIES,
Manila, P. I., September 30, 1904.

SIR: I have the honor to transmit herewith, for publication in one bulletin of the Bureau of Government Laboratories, the following: I. Does Latent or Dormant Plague Exist Where the Disease is Endemic? II. Broncho-Pneumonia of Cattle: Its Association with *B. bovisepticus*. III. Pinto (Paño Blanco). IV. Notes on Analysis of the Water from the Manila Water Supply. V. Framboesia: Its Occurrence in Natives of the Philippine Islands.

I am, very respectfully,

PAUL C. FREER,
Superintendent of Government Laboratories.

HON. DEAN C. WORCESTER,
Secretary of the Interior, Manila, P. I.

DEPARTMENT OF THE INTERIOR,
BUREAU OF GOVERNMENT LABORATORIES,
BIOLOGICAL LABORATORY, OFFICE OF THE DIRECTOR,
Manila, P. I., July 15, 1904.

SIR: I have the honor to transmit herewith and to recommend for publication a paper entitled "Does Latent or Dormant Plague Exist Where the Disease is Endemic?" by Dr. Maximilian Herzog, Pathologist Biological Laboratory, and Mr. Chas. B. Hare, Assistant Bacteriologist.

Very respectfully,

RICHARD P. STRONG,
Director Biological Laboratory.

DR. PAUL C. FREER,
Superintendent Government Laboratories, Manila, P. I.

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PART I.

DOES LATENT OR DORMANT PLAGUE EXIST WHERE THE DISEASE IS ENDEMIC?

By MAXIMILIAN HERZOG, M. D., *Pathologist Biological Laboratory*, and
CHAS. B. HARE, *Assistant Bacteriologist*.

On August 21, 1903, Mr. Henry A. Blake, (1) governor of Hongkong, addressed a communication to the secretary of state for the colonies, entitled "Bubonic Plague in Hongkong: Memorandum by His Excellency the Governor, on the Result of the Treatment of Patients in Their own Houses and in Local Hospitals during the Epidemic of 1903." The writer of the memorandum makes some very startling assertions as to the danger of the spread of plague by animals of the most varied kind, and also comes to the amazing conclusion that there existed in Hongkong during the period of time intervening between June 23 and July 10, 1903, between 8,000 and 9,000 or more individuals among the native population in which plague bacilli were present in the circulating blood in spite of the absence of all clinical symptoms of the disease. The governor calls this condition "latent plague" and considers it a potent factor in the spread of the infection, and a factor which can not of course be reached by the ordinary methods employed to limit the spread of and possibly to suppress plague.

Fully to understand the statements of the governor of Hongkong, it will be well to quote a few paragraphs from his memorandum, which read as follows:

We have from Professor Simpson's report evidence that pigs, calves, sheep, monkeys, geese, ducks, turkeys, hens, pigeons, and rats are susceptible to plague, which may be contracted by food or by inoculation direct,

or by means of suctorial insects. To this list the examination mentioned above adds bugs, spiders, flies, and cockroaches. I may add that quails kept in the market for sale were also found to be infected. In paragraph 22, page 100, Professor Simpson points out that domestic animals suffer from chronic plague, and surmises that this is probably one of the bridges by which the interval of the attacks in man is connected. I have for a considerable time been of the opinion that man himself is subject to chronic plague, which may either pass away after a considerable time or continue dormant over the winter months, regaining activity with the annual movement of spring, when the curve of the epidemic is almost constant. This opinion was strengthened by the fact that in August, 1899, the body of a Chinese lift man at Queen's buildings who was accidentally killed when attempting to enter the lift while in motion was found to contain plague bacilli. A similar result followed the examination of a man who on the 4th of March, 1901, was killed at Tal Koo Sugar Works by a bag of sugar falling on his head from a height of 20 feet; while on the 2d of April, 1903, in the body of the chief steward of a ship lying in the dock, found floating with a large wound on the head, were also found plague bacilli. Early in June several men from H. M. S. *Ocean* were sent to the naval hospital, suffering from pneumonia; on examination of their blood seven were found to be suffering from mild cases of plague. In like manner two officers of the Sherwood Foresters who developed feverish symptoms were, on having their blood examined, found to be similarly affected. In the "Boletin Oficial" of Macao, containing the report on the plague epidemic, 1895, Dr. Gomes de Silva, the medical officer who published the report in 1895, stated that during the height of the epidemic he had discovered plague bacilli in his own excreta.

(21) In June I directed Inspector Gidley to obtain as many specimens of blood as possible, on slides secured from the Government bacteriologist. He obtained 110 specimens from men, women, and children taken at random. These slides were sent to Dr. Hunter for examination, who reported that in five slides he found plague bacilli, and in seven slides bacilli were present in considerable numbers, some of which showed bipolar staining. They were not sufficiently distinctive, however, to be regarded as *B. pestis*. These slides were obtained between the 23d of June and 10th of July. Since they were obtained there were but three cases of plague in the district, from none of which a specimen of blood was taken.

(22) I am not unmindful of the fact that these reports were the result of microscopic examination only. But the examination was the same as that on which a great many of the cases treated in the Kennedy Town Hospital were sent to that institution where their cases ran the usual course of plague invasion.

(23) Now, putting aside the seven doubtful slides, it will be seen that of those people examined at random 4.54 per cent were found to be infected with plague though to all appearances perfectly healthy. If we exclude all the well-to-do, and take the working coolie population alone, they

probably number 180,000, and, assuming the same average amount of infection, there are among that class alone 8,172 persons at present infected in Hongkong. If even a quarter of that average be accepted for the 105,000 inhabitants of the superior class the number of infected will be increased to 9,634. In Appendix G¹ will be found the number of rats examined in each month of the present year with the proportion of the infected rats. I am afraid that the incidents mentioned in paragraph 5 weakens deduction as regards Hongkong. But, from whatever source the rats were procured, the proportion of infection in June was 9 per cent or 4.46 per cent more than the percentage of the slides examined, or, if doubtful cases mentioned by Dr. Hunter be included, 1 per cent less, while in January the proportion falls to 0.8 per cent. This being so, with the complete circle of vermin, insects, food, rats, domestic animals, and man all infected in possibly similar—possibly different—proportion, it appears to me unsound to concentrate attention upon the rat as the principal means of bridging over the dormant season.

It appears that Governor Blake, after writing the above, felt the great danger of coming forward with so sweeping an assertion, and in the introduction to his memorandum he himself makes an appeal for a more through scientific investigation of the hypothesis of the existence of latent or dormant plague among the natives of countries where this disease is endemic. He says:

My hypothesis in paragraph 23 may not bear the light of scientific investigation, and, as the hypothesis of a layman, may not carry much weight, but I venture to submit that it is worthy of scientific inquiry, for while a timely glass of water may prevent a great conflagration, and plague at its first introduction may be stamped out by immediate segregation and thorough disinfection, its endemicity once established this is no longer practicable, and, if the hypothesis of dormant or chronic plague in man be ultimately proved to be correct, it is difficult to see how quarantine for even ten days can prevent its annual recurrence, or how any practicable examination of departing passengers can prevent its export from the plague center and possible dissemination elsewhere if suitable conditions for its propagation be present. What the remedy or what the necessary precaution, I leave it for scientific men to determine, but if my hypothesis results in a wider radius of investigation the experiment will not have been useless.

THE RESULT OF BLOOD EXAMINATIONS IN CASES OF PLAGUE.

It is, of course, obvious to any one versed in examinations of this nature that a diagnosis of plague can not be made by a microscopic examination of the blood. Such an examination may possibly be

¹ Omitted in this bulletin.

resorted to in urgent situations, when a rapid clinical diagnosis is desired, but to base far-reaching deductions upon such a microscopic examination is certainly not permissible. What is necessary in order to determine beyond doubt the presence of plague bacilli in the circulating blood is the examination of the latter by cultural methods. We have undertaken a series of such examinations in order to determine whether or not there exists such a thing as latent or dormant plague, as suggested by the governor of Hong-kong. Before giving the details of these experiments it will be well to make a survey of the work that has been done with reference to the presence of plague bacilli in the blood in undoubted cases of this disease.

The German Plague Commission (2), in its report published in 1899, page 265, made blood examinations in the case of 141 plague patients, including 17 who were in the period of convalescence varying between the seventeenth and twenty-fifth day after the disease. These examinations were made in the following manner: A drop of blood was obtained from the finger, with the usual aseptic precautions, and inoculated into agar tubes, while at the same time a cover-glass preparation was also made. It was found that in a number of cases where the culture method furnished positive results, the mere microscopic examination failed to demonstrate the presence of the bacilli. Of 124 patients whose blood was obtained during the climax of the disease, in 81 even repeated examinations did not demonstrate the presence of plague bacilli, in 10 the bacilli were encountered only once, while in 33 they were repeatedly found in the blood. Of 81 patients the examinations of whose blood were always negative, 52 recovered and 29 died. Of 10 cases, in which there was a positive result only once, the other examinations being negative, 8 died and 2 recovered. It is interesting to note that in one the bacilli could be detected two and three days before death, while twelve hours before the crisis and at the post-mortem examination it was impossible to find them. Seventeen convalescent patients invariably failed to show any bacilli in the circulating blood.

Zobolotny (3), in his researches on plague, says that the bacilli are found in large numbers in the blood of animals sick with the disease, but that in the case of human beings the bacteria are much less abundant and sometimes can not be found at all.

Musehold (4), quoting the work of Albrecht and Gohn, reports that the latter examined 122 cases of undoubted plague. In 55 the bacilli were found by the culture method in the circulating blood during life. Four of these 55 cases recovered, and in 2 of them the bacilli were present in large numbers in the circulating blood. In the case of the 51 patients who died the bacilli were found in considerable numbers in the blood on the day of death as well as on the previous one.

Cairns (5) studied the blood of patients during an epidemic of plague occurring in Glasgow in 1900. He gives the results of examinations made *inter vitam* on cases which subsequently terminated fatally. Four of these may be cited in connection with our investigation. In three fluid drawn from the buboes developed pure cultures of *Bacillus pestis*, and only in one of the four fatal cases was it possible to obtain cultures of the bacilli from the blood during life. The other three cases gave negative results. In one of these, seven daily consecutive examinations were made, as well as one shortly before death, all of them proving negative.

Calvert (6) studied two epidemics of plague in Manila in 1900 and 1901. He found that the clinical examination of the blood for *Bacillus pestis* gave unreliable results, it being impossible to place any weight on the negative findings. He made his examinations by taking the blood from the ears of the patients at intervals of four hours, and examining it in smears as well as by the culture method. This plan was followed until the death or recovery took place. Thirty-six cases, 4 of which recovered, were examined in this manner, most of them being followed to autopsy, when the plague organism was demonstrated by culture and even by animal inoculation.

This author gives the following table of positive findings of the bacilli in the blood:

	Per cent.
24 hours before death in 31 cases.....	100
48 hours before death in 7 cases.....	22
72 hours before death in 5 cases.....	16.12
96 hours before death in 2 cases.....	6.45
120 hours before death in 1 case.....	3.22

On looking over the table it appears that the plague bacilli could be found in all fatal cases twenty-four hours before death, but that forty-eight hours before death the percentage of positive findings was much smaller. In searching for the bacillus in the blood of plague patients who finally succumb to the disease, the chances of finding the microbe five days before the fatal termination are only one out of thirty. All of this shows that even in fatal cases the plague bacilli are not found in the peripheral blood at an early date.

Jennings (7), in his *Manual of Plague*, page 112, says that it is extremely rare to find plague bacilli in the blood in large numbers except immediately before death. Their absence, therefore, in the early stages of an attack is frequent, and must not be regarded as a negative diagnosis of plague.

Terni (8), who studied the plague in Rio de Janeiro, in an excellent article on the disease makes the following statement:

"The examination of the blood is by no means reliable. It is astonishing that Galeotti places any value in blood examinations in the diagnosis of early plague."

Terni found that in many cases which were diagnosed as plague septicemia, examinations of the blood, both microscopic and by culture method, were negative. This was true even at the post-mortem examination, because the bacilli were exclusively localized in the lymph channels. Even in the most profound cases, in individuals particularly predisposed to the disease, the bacilli were found in the blood only in moderate numbers. Their presence could never be compared with what has been found to be true in connection with other septicemic microbes, such as anthrax or diplococci. A multiplication of the plague bacilli is found only in exceptional cases in the circulating blood during the agonal stage.

One of us has been studying for some time the morbid anatomy and histo-pathology of a number of cases of plague. These studies appear fully to confirm the statement of Terni to the effect that plague, as a rule, is not to be looked upon as a true septicemia, but, on the contrary, as an infection particularly confined to the lymphatic system. Even in cases where sections from the lymph glands contain innumerable bacilli, the lumina of the blood vessels are generally free from such microbes. In fact, in the study of sections from all of the internal organs when plague bacilli are seen they are always found in the lymph channels or lymph spaces and not in the blood vessels.

Powell (9) has recently reported the result of 3,400 blood examinations of febrile diseases in Bombay. Most of these cases were malaria, but 117 of them were plague. In only 15 of the latter were the bacilli easily seen in blood smears. With reference to the finding of the bacilli in the blood in cases of plague, the author says: "As regards the recognition of the plague bacilli in the finger blood, for some years I was very sceptical about the reports of certain medical men, and until within the past eighteen months had been unable to detect the bacilli except on cultures. At the beginning of this year there was a particular type of septicæmic plague, in which the bacilli were found in every case. Such cases in my experience always died. One case seemed to be convalescent and had a normal temperature for three days, but suddenly died. Plague bacilli were found eleven days before death."

The *Indian Plague Commission*, speaking of the bacteriologic diagnosis of plague by microscopic examination of suspected material, makes the following statements:

(156) In the case of blood derived from a patient who is suspected of suffering from plague, the detection of bacteria possessing the morphological characters of the plague bacillus will (especially if these are present in large numbers and when it is determined that these become decolorized by Gram's process) be conclusive evidence that the patient is suffering

from plague. The mere finding of a few isolated bacteria arranged together as diplococcal forms can not, especially when Gram's test has not been applied, be accepted as establishing the diagnosis of plague. We have in view here in particular certain suspected cases which occurred in Calcutta in 1896, in which it was claimed by Professor Simpson that the diagnosis of plague was confirmed by his bacteriological examinations. We would note with regard to these first that in some of the suspected patients only a few isolated diplococcal forms were found after long searching through a number of films prepared from the blood. Again, in view of some of the figures reproduced in the record of Dr. Simpson's evidence, particularly Figs. I, VI, C, and F, it is important to note that it does not seem to have been determined whether bacteria became decolorized when treated by Gram's process, and, lastly the fact must not be lost sight of that saprophytic diplococci of various kinds are widely distributed in nature, and that there is always a possibility of microscopical preparations containing as contaminations a small number of such diplococcal forms.

In looking over these statements (all that we can find in the literature at our disposal), one is certainly impressed with the fact that the finding of plague bacilli in confirmed cases of the disease, except very shortly before death or in the rarer cases of plague septicæmia, is the exception and not the rule. Indeed, we should be mindful of the fact that plague, as a rule, is not a septicæmia but a bacterial infection localized in the lymphatic system. It is, therefore, from a purely theoretical standpoint, highly improbable that there should exist a dormant, latent clinical form of the disease in which the patients harbor the bacilli in the circulating blood.

Our investigations to determine whether such is the case or not were made on a number of native Filipinos and Chinese of this city. We attempted to get material which, if latent plague exists at all, would give us some evidence to this effect, therefore we selected natives from houses in which plague cases had occurred. We examined a number of the inmates of Bilibid Prison, particularly such as were under the most unfavorable hygienic conditions, namely, insane prisoners and those of the third class who were most crowded in their quarters. Since we could not always get natives of this description, we selected also a number which were not under particularly unfavorable hygienic conditions, such as native police officers and native members of the Constabulary.

While plague has never at any time been as widespread here as in Hongkong, a sufficient number of cases have occurred to make

it clear that the disease is endemic, though fortunately not markedly epidemic.

According to the monthly sanitary reports of the Board of Health plague has prevailed in Manila since 1900 to the following extent:

Prevalence of plague in Manila, 1900 to 1904.

1900.

Month.	Chinese.	Filipinos.	Americans and other Caucasians.	Total number of cases.	Total number of deaths.
January.....	3	15		18	11
February.....	36	12		48	35
March.....	52	12		64	48
April.....	43	11		54	44
May.....	13	7	2	22	18
June.....	14	5		19	11
July.....	5	8		13	7
August.....	8	9	1	18	11
September.....	6			6	9
October.....	5	2		7	5
November.....	1			1	
December.....		1		1	
Total.....	186	82	3	271	199

1901.

January.....	4	3		7	5
February.....	15	11	1	27	20
March.....	49	14		63	51
April.....	73	38		111	91
May.....	97	40		137	124
June.....	24	30	1	55	54
July.....	18	20	1	39	38
August.....	12	16	1	29	26
September.....	7	4		11	12
October.....					
November.....					
December.....	1	4	1	6	6
Total.....	300	180	5	485	427

1902.

January.....					
February.....	1			1	1
March.....		1		1	1
April.....					
May.....					
June.....	1			1	1
July.....					
August.....	1			1	1
September.....	1			1	1
October.....		2		2	2
November.....	1			1	1
December.....		2		2	2
Total.....	5	5		10	10

Prevalence of plague in Manila, 1900 to 1904—Continued.

1903.

Month.	Chinese.	Filipinos.	Americans and other Caucasians.	Total number of cases.	Total number of deaths.
January-----		1		1	1
February-----	7	10		17	15
March-----	18	15		33	33
April-----	35	15	2	52	49
May-----	16	9	2	27	23
June-----	9	23		32	25
July-----	3	11		14	9
August-----	10	1		11	9
September-----	3	1		4	4
October-----	3			3	2
November-----		2		2	2
December-----		2		2	2
Total-----	104	90	4	198	174

1904.

January-----	4	6		10	7
February-----		6	1	7	6
March-----	3	12		15	14
April-----	8	7		15	15
May-----	9	8		17	16
Total for five months-----	24	39	1	64	58

SUMMARY.

Reported in—	Plague cases reported.	Plague cases fatal.
1900-----	271	199
1901-----	485	427
1902-----	10	10
1903-----	198	174
1904 (Jan. 1 to May 31)-----	64	58

It appears from these statistics that plague had completely died out during four of the months of 1902, since during this period not a single case came under observation. However, since August, 1902, until the time of writing the present report there has not been a month entirely free from plague, though the figures have generally been low, the maximum during this period being reached in April, 1903, when 52 cases of the disease were reported.

The object in giving these figures in connection with our report is to show that plague has been sufficiently prevalent here for several years, so that blood examinations should furnish evidence

of latent plague provided that such a form of the disease exists at all.

The figures of plague morbidity for Hongkong during the same years are as follows:

	Cases.
1900.....	1,086
1901.....	1,637
1902.....	540
1903.....	1,135

BLOOD EXAMINATIONS OF 245 APPARENTLY HEALTHY NATIVE FILIPINOS AND CHINESE.

The method employed to ascertain whether apparently healthy Filipinos or Chinese of Manila have any plague bacilli in their blood was as follows:

The bend of the elbow, generally of the left arm, was very thoroughly cleansed first with strong alcohol and then with a strong solution of mercury bichloride, and finally with alcohol and sterile distilled water. A rubber bandage was then placed around the arm above the elbow and 1 cubic centimeter of blood was drawn from one of the veins by the aid of a sterile hypodermic syringe. The blood so obtained was added to 50 cubic centimeters of bouillon in a flask. The bouillon used was prepared as usual, and, when neutral to litmus, 0.5 gram of bicarbonate of soda was added to each 1,000 cubic centimeters of the bouillon, making it slightly, but decidedly, alkaline. This forms a very excellent culture medium for plague bacilli. As a control experiment in some of the cases about twice the amount of blood was drawn from the vein, and the 2 or 3 cubic centimeters so obtained was distributed in two flasks. One of the latter was then immediately inoculated with a very small amount of material from a young plague culture. This was, of course, done to see whether plague bacilli, if present, would develop in the bouillon in the presence of a small amount of freshly drawn blood. It may be said that the control flasks always developed a typical plague growth, so that evidently nothing in the arrangement of the experiment prevented development of the plague bacilli if any were present. The culture flasks to which blood had been added were kept either in a dark chest at room temperature or in an incubator which was fairly constant at 35° C. The media were inspected daily and when a growth developed it was examined in stained preparations and by culture on agar.

The native Filipinos whose blood was examined included 32 laborers from the Serum Institute and the morgue. The native servants of the latter, where all the plague post-mortem examinations of the city are made, are, of course, particularly exposed to infection, and would be especially prone to show latent plague provided that such a condition exists. These 32 cases were kindly examined for us by Dr. E. H. Ruediger, bacteriologist in the Serum Institute, whose technique differed from the method generally used only in that he employed a 5-per-cent carbolic acid solution to sterilize the elbow. Dr. Ruediger also examined all of his 32 flasks by stained cover-slip preparations and by culture methods whether they showed any change in appearance or not.

The blood examinations in all of the 245 cases were made between March 4 and May 20, 1904—i. e., during a period when from thirty-five to forty cases of plague were reported in Manila.

The following is a summary of the examinations:

On March 4 there were examined 5 native Filipinos from a house in Santa Cruz in which an ambulatory case of plague terminating in embolism of the pulmonary artery had occurred. Specimens were taken from 10 native Filipino police officers on April 6 and 7. Nine of these men lived in infected districts—i. e., those in which cases of plague had recently occurred; 1 came from a noninfected district. Ninety native Filipinos, members of the Philippines Constabulary, were examined between April 13 and 30. These men live in barracks, but are often free to visit their families and friends. During the first week in May 32 native Filipinos, laborers at the Serum Institute and the morgue, were the next subjects investigated. In the former place the various vaccines and sera, including plague vaccine and serum, are prepared, and in the latter are performed all the necropsies on plague cases. Fifty-eight native Filipinos, prisoners in Bilibid Prison, were taken on May 12 and 13. In this place about 4,500 men are confined. Last year a number of the inmates died from pneumonic plague, but during this one no case has occurred there, although one of the prisoners died of pulmonary plague four days after his discharge. Of these 58 men 16 were insane prisoners, all in advanced stages of degenerative mental disease, and the remainder were the so-called third-class prisoners, who lived under the most unfavorable conditions to be found in the prison.

On May 16 to 20 there were examined 50 Chinese, small shopkeepers, clerks, and coolies, either from houses in which plague cases (in one instance two such cases) had occurred or from those adjoining.

RESULTS OF THE EXAMINATION.

Most of the flasks to which 1 cubic centimeter of blood had been added remained sterile; although a few developed growths which,

however, were clearly contaminations from the air, such as common molds or similar forms of life. One of the cases taken by Dr. Ruediger developed *Staphylococcus pyogenes aureus*; and those of two other natives developed a bacillus which, when examined in a stained cover-glass preparation, might possibly be mistaken for *Bacillus pestis*. One of these organisms, however, in culture looked very different from the bacillus of plague and also retained Gram's stain. This bacillus developed in a flask to which had been added blood from one of the insane prisoners. The other growth occurred in a flask containing blood from a member of the Constabulary. This organism also greatly resembled morphologically the plague bacillus, but it kept Gram's stain, and when rubbed in large amounts on the shaved abdomen of a guinea pig failed to produce disease. In short, not in a single instance out of 245 examinations of persons of whom a large percentage had certainly been greatly exposed to plague infection did we find any evidence of the existence of plague bacilli in the blood.

CONCLUDING REMARKS.

From our investigations conducted on 245 native Filipinos and Chinese it may be safely concluded that a condition of latent or dormant plague does not exist in Manila, and there is hardly any reasonable doubt that it does not exist in Hongkong. There certainly has not been furnished the slightest proof of such a character as to stand the searchlight of exact methods of bacterial investigation to indicate that there is such a thing as latent human plague, with the presence of plague bacilli in the circulating blood, in the absence of clinical symptoms of the disease.

The governor of Hongkong himself, in his memorandum, clearly sets forth some of the circumstances which unite to make it practically impossible to completely eradicate the disease in Victoria City.

In Hongkong (1) (memorandum, par. 3) it is the custom of the Chinese—if they can possibly do so—to dump human corpses dead from plague into the street, in order that they may not be found in the houses and thereby subject the inmates to quarantine, disinfection, etc. In spite of measures to prevent this procedure, the number of corpses dead from the plague and so disposed of has, during the ten years preceding 1898, increased from 25.1 per cent to 32.7 per cent.

The Chinese in Hongkong, according to the governor's memorandum, offer passive resistance to the catching of rats in their houses, being afraid that plague bacilli might be found in the rodents, as this would lead to measures of disinfection or to the repair of their houses. The Chinese rat catchers are said to be dishonest; they fail properly to label the rats, so that infected houses escape detection, and they import rats from the outside of Hongkong and label them at random. In general they are very unreliable in their work and are actuated solely by the desire to secure from anywhere the largest number of rats in order to obtain the premium offered for each.

To those who know how Chinese houses are constructed [says the memorandum, in par. 6], it will be apparent that effective fumigation is practically unattainable. While, even if the spraying process, scrubbing and disinfection of clothing reached externally everything in the room, it would not kill vermin lying deep in the joints and cracks of the tables, chairs, and settees, or beds. Nor would it reach the vermin with which the heads of the poorer classes of coolies are infested. But apart from this, what took place in many cases when a case of plague was detected was that before the constable could arrive to take charge of the house, goods liable to injury by disinfection were removed by the door, or, if too late for this, were taken on to the roof, always easily accessible, and deposited in some neighboring house.

In W. J. Simpson's report (11) "On the Causes and Continuance of the Plague in Hongkong, etc.," which is quoted in the memorandum of the governor, we find the following statements as to the sanitary conditions in the Chinese quarters in the city of Victoria.

When a case of plague has once occurred in a house, there is a great tendency in subsequent years for the same house, or that adjoining, or that on the opposite side, or that close by, to be attacked with plague. When plotted out on a map, the distribution of plague appears to be closely connected with previous infection of the house or of a defined locality, the infection having been retained in an unrecognized form in the interval. The houses which suffer principally are, speaking generally, the most insanitary and the oldest. It has already been mentioned, how closely packed the buildings are in the older portions of the town, narrow streets and high houses being the leading features, by which the admission of sunlight and fresh air is considerably obstructed. Narrow streets and high houses, however, are not peculiar to Hongkong; they are to be found in other towns, with their injurious effects on health, but in Hongkong there is moreover in the Chinese quarters a defect in the construction of the houses which intensifies the obstruction of light. The rooms are long and narrow, with a window at each end, the front

window looking into a wide and covered veranda, and the back window into a small open space at the back, which forms a sort of wall between the two houses. The lower floors of many of the houses are remarkable for their darkness, and this in a region not far from the Tropics; they are also frequently damp.

Since the epidemic of 1894 many of the lower floors of the worst kind have been changed into storerooms to contain the goods and merchandise for which Hongkong is in entrepôt. These storerooms as a rule are infested with rats, which at times find their way up to the rooms on the higher floors. The basements are generally rat ridden, both floors and walls, and from the walls being often hollow it is easy for rats to reach the upper floors.

The admission of sunlight into the dwelling rooms of Chinese tenement houses is still further obstructed by the subdivision into several cabins or compartments, sometimes numbering up to six, which every room is subjected to. Each cabin is let out to a separate tenant and not infrequently accommodate a separate family. These compartments or cubicles are windowless rooms, and are often so dark that it is impossible for any one, coming directly from the light outside and drawing or opening the door of the cubicle, to see at once whether it is occupied * * *. Some attempts have been made to improve this state of things by limiting the height of the subdividing walls to six feet. The condition which obtained before this improvement has made it somewhat difficult to realize, for what I am describing is that which now exists. Fresh air and sunlight never get into the cubicles except perhaps the compartment at each end of the room opposite the window. The subdivision of a single room into a number of rooms called cubicles is an ingenious device for crowding together a large number of people into a small space and securing a correspondingly large rental, but it is an arrangement which engenders disease and favors its spread. There is no doubt whatever that every such windowless cubicle is unfit for human habitation and should not be permitted * * *. Probably another cause for the continuance of the plague, besides the insanitary condition of the houses referred to, is the very inadequate number of latrines and urinals with which Hongkong is provided. The number of public latrines appears to be twenty-nine belonging to the Government, and seventeen to private owners. The total number of seats is 1,202. Most of them have urinals attached, and in addition there are three small public urinals in the town. Seeing that all the men and boys go to the public latrines, there are no sanitary appliances in the houses except earthen pots, which are used exclusively by the women and children. The total inadequacy of the latrine accommodation provided is obvious. It is not one seat to one hundred of the male population. On the Kowloon side of the colony the latrine and urinal accommodation is still more deficient. Large blocks of houses have been built, and not a single latrine or urinal provided by the builder of the block. It is impossible under these conditions that the ground should escape being sewage polluted * * *. The existing latrines are far from being models

of what they should be. They are in fact insanitary in structure and deficient in light and ventilation.

Quite recently Surgeon-General Evatt, P. M. Q., His Majesty's troops, Hongkong, has been quoted by the daily papers in an interview in which he speaks in the strongest terms of the insanitary conditions, which he holds responsible for the continued prevalence of plague in Hongkong. He calls Victoria City the plague-distributing center of the world, a standing menace to the human race. So we have the most convincing evidence, both official and unofficial, as to the vicious sanitary conditions in Hongkong. These furnish the soil in which plague thrives, from which it can not be completely eradicated, and from which it breaks forth again and again in menacing epidemic form.

From our own investigation carried on in Manila, we have certainly good reasons to deny the existence of latent or dormant plague in our city. The statements of those who have looked into the conditions in Hongkong likewise appear unfavorable to the theory of the governor, and they offer an explanation more in accord with what we do know positively as to the nature of plague infection, and as to the spread of infectious diseases in general.

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PART II.

BRONCHO-PNEUMONIA OF CATTLE: ITS ASSOCIATION WITH *B. BOVISEPTICUS*.

By PAUL G. WOOLLEY, M. D., *Director of the Serum Laboratory*, and
WALTER SORRELL, D. V. S., *Veterinarian Serum Laboratory*.

The reason we care to dwell upon these cases is not that the above-mentioned pulmonary affection is uncommon or not well known, but because of its relation, here at least, to hemorrhagic septicæmia.

It is agreed by all authors that the causes of catarrhal pneumonia, in animals as well as in man, are not always the same. However, in general the chief ones are also those of acute bronchitis. Following this it happens, either because of swelling of the mucous membrane, or by aspiration of bronchial secretion, that the small subdivisions of the bronchi become plugged, and a condition of atelectasis arises in that part of the lung supplied by the affected bronchioles. Then because of the *locus minoris resistentiæ* furnished by the atelectatic part, and because of the usual presence of infectious material in the bronchial secretions and in the aspirated material, inflammatory processes occur, which may be limited or which may spread by continuity to the neighboring tissue. The changes following atelectasis may either be a gradual atrophy of the affected parts of the lung, or following infection, bronchiectasis, abscess formation, pleurisy, gangrene, etc., resolution, or calcification. The disease occurs especially in young animals and in older ones whose physical condition is below par. In young animals it may be epizootic.

The symptoms are said to vary. Schneidemühl says that the

first signs point to acute bronchitis, upon which ensue pulmonary symptoms, such as increased respiratory frequency and cough.

The course of the disease according to Diekerhoff and Schneide-mühl is neither typical nor regular. Sometimes it progresses very rapidly. In other cases it may last for weeks or months.

In the more chronic ones a purulent pneumonia with pleuritis and pulmonary gangrene may ensue and death follow. It is in this last group, that of chronic cases, that ours belong.

These occurred in the herd of the Government Serum Laboratory. The calves were imported for use in the preparation of vaccine virus. The cattle were used in the preparation of antirinderpest serum. All of the calves originally came from China, and upon arriving in Manila had been treated with prophylactic doses of antirinderpest serum and kept under careful observation for several weeks before they were used for the intended purpose. At the time of vaccination there was no indication of disease in any member of the herd, their appetites were good, they were in excellent physical condition, and their temperature curves were normal. (Inasmuch as tuberculosis has never been observed in cattle here the vaccine calves are not slaughtered after collecting the virus, but are kept on hand until disposition can be made of them in some other way. As a rule, the physical condition of the animals improves after taking the vaccine. When this is not the case slaughter is resorted to when there is no particular reason, as in the present instances, of allowing the animals to live.)

The steer mentioned below was also imported from China for use in the preparation of antirinderpest serum, but its condition was never satisfactory for this purpose.

The histories follow:

CASE I.

Calf No. 437 was received at the laboratory on January 12, 1904, and received 100 cubic centimeters of antirinderpest serum at once. It remained apparently well with a normal temperature until January 24, when the latter rose to 40.7° C. The next day the temperature was 40.8° in the afternoon, and the following day it reached 41.2°. From this point it gradually declined to normal on January 30. On February 1, 5 cubic centimeters of virulent rinderpest blood was given to the animal, and on the third day after, it was vaccinated. Following this operation, there was what at first appeared to be the usual temperature of vaccinia, which reached 40.6° on the fifth day after vaccination. But this temperature instead of declining persisted with slight variations between 39° C.

and 41° C. until March 1, when it became normal and stayed so until death, March 24, 1904. On several occasions the blood of the animal was examined for trypanosomas with negative results.

During the course of the disease, the animal gradually lost weight in spite of a reasonably good appetite, and as it progressed the appetite became impaired and the coat rough and starchy. There was no cough, nor in fact any other notable symptoms than the gradual wasting.

The autopsy was made about twelve hours after death. The only appreciable lesions present were in the lungs. The other organs were in an apparently healthy condition, with the exception of the prescapular lymph glands, which were enlarged and edematous.

The apices of the lungs were chiefly affected. These portions of the organ were almost completely solidified. The surface of the hepatized portions were mottled with yellowish-white areas which stood out distinctly upon a reddish-purple ground. In palpating, the finger was able to detect that these lighter areas were firmer than the darker colored portions of the lungs, which had an edematous consistency. Over several of the nodules there was a thin, fibrinous membrane under which the pleura was congested and roughened. Upon section, the affected portions of the organs cut with increased resistance, and from the incisions a frothy serum oozed. The general color of the surface was dark red, mottled by the grayish yellow of the sometimes almost caseous nodules. Sections of the latter had all the macroscopic characteristics of the gray hepatized tissue of pneumonia, being granular and dry. In an occasional one the process had gone beyond the simple hepatization and the center had softened, producing a rather creamy material.

From several of such tubercles, cultures were made upon agar and in bouillon, and pieces of the tissue were placed in Zenker's solution, and absolute alcohol, for further study. Smears made from the pulmonary nodules showed a few small ovoid bacilli, which stained with the ordinary anilin dyes and were decolorized both by Gram's and Gabbett's methods.

Cultures of organisms were obtained which had all the characteristics of the bacillus described by one of us as the cause of an epidemic of hemorrhagic septicæmia among the carabaos of the Government corrals during the past year. The chief characteristics of this organism were its polar staining, rounded ends, nonmotility, and occasional encapsulation. It grew invisibly on potato, did not produce gas in either solid or liquid glucose media, did not coagulate or acidify milk, did not form spores or liquefy gelatin, but did reduce nitrates and also gave the indol reaction. It was pathogenic for guinea pigs, causing death within twenty-four hours after intraperitoneal injection of 1 cubic centimeter. It was the *Bacillus bovisepiticus* of Kruse.

The tissues preserved for sectioning were embedded in celloidin, and sections from these were stained with hæmatoxylin and eosin, by Gram's method and by the carbol-fuchsin-acid one for tubercle bacilli. The stained sections showed a generally edematous condition of the parenchyma. The trabeculæ were somewhat thickened and the fibrous tissue of the affected portion of the lungs was generally increased. The epithelial linings of the

bronchi were convoluted and hyperplastic, the cells being in many places three or four layers thick, and in the dilated lumina there were considerable accumulations of epithelial cells, polymorphonuclear leucocytes and a minimal amount of fibrin.

The air spaces of the affected lobes which were not involved in the consolidation were filled with a granular material with which there were occasional desquamated cells and leucocytes. The peri-bronchial tissue was the seat of well marked round cell infiltrations and the peri-bronchial connective tissue was considerably increased.

Sections of the small nodules of consolidation showed that the chief change in these was a coagulation necrosis. The centers of these areas were crowded with leucocytes and granular, cellular detritus enmeshed in fibrin. The walls of the air cells were perceptible as faintly pink-stained bands in which no nuclei or only shadows could be seen. As the periphery of these areas was approached, the lines became more distinct, and about the latter was a zone of congestion. There were no giant cells seen in any sections, and no "acidfast" bacilli could be found. There were, however, a number of very short bacilli between the cellular contents of the abscesses.

CASE II.

Calf No. 423 was received from Hongkong on January 6, 1904, and given a preliminary prophylactic dose of 100 cubic centimeters of anti-rinderpest serum. It was vaccinated on January 13, after which its temperature rose to 40° C. where it remained with slight remissions until January 19, reaching normal on the 20th. On January 21 it received 10 cubic centimeters of virulent rinderpest blood, and following this the temperature again rose and varied between 39.2° and 40.6° for the next eight days. On February 1 it received 50 cubic centimeters of virulent blood, and following this there was again a rise, which, however, was transient. From this time on the temperature remained within normal limits. Death occurred April 30, somewhat more than three months after the arrival of the animal at the laboratory.

During the last month or five weeks of its life a steady and gradual decline was evident. The animal lost weight in spite of a constantly fair appetite. Upon several occasions the blood was examined for trypanosomas, each time in vain. No cause could be found for the wasting. There was no cough, in fact no other symptoms than the gradual emaciation, and increasing weakness.

The day before death the animal was unable to stand but was lying in its stall in the hospital shed, eating the grass before it. It died the following night and was found in the morning in its sleeping position.

Upon opening the thorax a considerable amount of clear serofibrinous fluid gushed out. The peritoneal cavity contained no fluid and was apparently normal. The subpleural and mediastinal tissues were edematous and gelatinous in appearance. The prescapular glands and periglandular tissues were also edematous. The liver appeared pale but otherwise normal. There were a few petechiæ in the capsule of the spleen, which was of normal size and consistence.

The cecum contained a number of oesophagostomas and fasciolas.¹ There were no gastro-intestinal hemorrhages or ulcerations.

The lungs were the seat of a patchy hepatization, which involved about one-half of the entire pulmonary tissue, chiefly the apices and anterior lobes. In the hepatized areas, and perceptible to the eye and finger, upon the surface of the lungs, were small nodules. These were small and paler than the adjacent tissue, which had an almost purple color. Section of these organs showed that the fibrous tissue was generally increased in the affected portions, the trabeculæ being quite prominent. The surfaces of cut sections showed mottled gray and red, the gray being most apparent in the centers of the lobules. Certain of these gray areas were quite dry and almost caseous, and were as large as a navy bean.

From these nodules cultures of a nonmotile polar-staining bacillus which did not stain by Gram's method and did not form spores were obtained on agar. The colonies in agar varied in size from 0.5 to 1 millimeter in diameter, were thick and opaque at their centers, were not chromogenic, had smooth, regular edges and were somewhat sticky and mucilaginous. On agar slants after twenty-four hours the growth was colonial, but with a tendency to confluence. The water of condensation was clouded.

On glycerin-agar the growth was more luxuriant, quite thin but almost opaque, and so moist that it was inclined to run down the surface of the slant.

Milk became just perceptibly acid after seventy-two hours, but without coagulation or reduction of the litmus. The growth on potato was invisible. In glucose broth and agar no gas was formed. In Dunham's peptone, to a liter of which 5 cubic centimeters of a 5-per cent solution of potassium nitrate had been added, a brilliant cholera red reaction could be obtained after twenty-four hours.² After seventy-two hours a pellicle appeared in the ordinary bouillon; the medium was diffusely clouded and a flocculo-gelatinous sediment was thrown down.

Of a twenty-four-hour-old broth culture, 1 cubic centimeter was injected into the peritoneum of a healthy guinea pig. The animal survived this treatment.

Cellodin sections cut from material fixed in Zenker's solution, and stained with hematoxylin and eosin, showed changes that correspond in all general respects with those described in the previous case.

CASE III.

Calf No. 415 was received at the laboratory on December 29, 1903, and given the usual prophylactic dose of 100 cubic centimeters of antirinderpest serum. The temperature remained normal until after vaccination, which

¹ Specimens of these parasites were sent to Chas. Wardell Stiles for identification.

² This method was suggested to us by Dr. W. B. Wherry. For some time we have all been troubled by the inconsistency of the peptone used in making Dunham's solution. Dr. Wherry, however, found that a constant reaction could be obtained by adding traces of nitrates to that fluid.

was done eight days later. Following this, on the third day, the temperature rose abruptly to 41.6° C. and then gradually fell to normal within the next week. On January 20, 1904, it was given a subcutaneous injection of 10 cubic centimeters of virulent rinderpest blood, following which there was a reaction of 1.4° C., after which the temperature fell to normal and continued so to the time of death on May 14, 1904, almost five months after the animal had been received. At no time were trypanosomes found in the blood. The clinical history in the case was not unlike that in the previous one; the salient points being gradual loss of weight in spite of retention of appetite, normal temperature, no cough, roughened and starchy coat, and loss of strength.

At autopsy the chief lesions, and in fact the only macroscopic ones, were in the lungs. There was not as much edema as in the previous case, and there was a less general pulmonary involvement. But here, as in the other ones, the apices and anterior margin of the lungs were partially solidified, mottled with red and gray, and filled with small, firm nodules, some miliary and some the size of a large bean. Some of these on section proved to contain a semifluid purulent material, while others were dry and gray.

Cultures were made as before from these nodular lesions and in this case two organisms were isolated, one *B. pyocyaneus*, the other a polar staining bacillus agreeing in general with the organism previously described, but varying from it in some cultural characteristics.

Morphologically, it was identical with the organisms from the previous cases. Culturally, variations were most marked in broth and on agar. On the latter, the growth was generally composed of colonies, but these were somewhat larger than the ones described for *B. bovissepticus*. The growth was much more luxuriant and whiter and there was a decided tendency to confluence on the part of the colonies.

In bouillon there was at first a diffuse cloudiness with no sediment and no pellicle. Later, a gelatinous sediment was deposited, a very delicate pellicle was developed, and the body of the medium became clear. There was also a deposition of floccules on the sides of the tubes.

The cholera-red reaction was obtained after twenty-four hours. Milk was unchanged, and there was an invisible growth on potato.

Intraperitoneal injection of 1 cubic centimeter of this organism killed a healthy guinea pig in less than twenty-four hours. At autopsy an acute hemorrhagic peritonitis was discovered, and the organism recovered.

CASE IV.

Calf No. 464 was received at the laboratory on January 12, 1904, and was injected with 100 cubic centimeters of antirinderpest serum. The following day its temperature was 40.1°, and on the succeeding it was normal and remained so for the next ten days. On January 20 it was vaccinated with vaccine virus, and following this its temperature rose, reaching a maximum, 41.1°, on the day after the vaccine was collected. Two days later it was again normal. On February 1, 1904, 10 cubic centimeters of virulent rinderpest blood was injected subcutaneously, and

following this the animal's temperature became quite irregular, reaching 40.5°, and with daily remissions of 1 to 2 degrees; but after twelve days the maximum had fallen to about 39.2° C. and with smaller remissions. This continued until death occurred, May 9, 1904, about four months after its arrival in Manila. Up to this time it was able to eat, but it gradually became emaciated and very weak.

At autopsy the lungs were found to be affected in a manner similar to the condition of the others described above. In this instance, however, the apices were alone affected over an area in each of about the size of the palm of a hand. These were purplish-red, edematous, and contained small areas of solidification, which appeared grayish or gray in sections. There was a fibrinous exudate over the pleural surface of some. The other organs showed no change other than mild parenchymatous degeneration. The body was anemic.¹

Cultures were made from the nodules in the lungs. On the original plate cultures two types of colonies were noted: One very small, similar to the typical colonies of *B. bovisepiticus*, the other larger and with more tendency to spread, and upon slant cultures to coalesce. Both of these were studied.

The only cultural differences in these two strains were noted in agar, upon which one showed a greater inclination to spread and coalesce, and in broth in which one (464¹) caused a uniform clouding with a gelatinous sediment, while the other (464²) clouded the media less diffusely and formed flocculi, which were present not only in the body of the fluid but also on the sides and bottom of the tube. Both gave a brilliant indol reaction. Both were pathogenic for guinea pigs.

Sections for material fixed in Zenker's solution and imbedded in celloidin showed the same histologic picture as that described in above cases.

CASE V.

Calf No. 491 was received at the Serum Laboratory on February 24, 1904, and was given a prophylactic dose of 100 cubic centimeters of anti-rinderpest serum. A transient rise of temperature to 41.2° C. and a gradual return, during seven days, to normal followed this. On March 5, 1904, 5 cubic centimeters of virulent rinderpest blood was injected subcutaneously, upon which a scarcely perceptible rise of temperature lasting but twenty-four hours was noted. Four days later the animal was vaccinated and again the temperature rose, remained between 39° and 40° C. during the inoculation disease, and then gradually fell to normal. On March 25, 1904, a second injection, this time 50 cubic centimeters, of virulent blood was made, and this gave rise to a slight increase in temperature, which lasted but forty-eight hours.

During the following month the temperature remained above normal,

¹In the small intestine there were some round worms, which were preserved and sent to Chas. Wardell Stiles for identification. In his report he says that they are in all probability a new species of *Hæmonchus*.

varying from 39° to 40° C., and only dropped immediately before death. The animal died on May 5, 1904, a little over two months after its arrival at the laboratory.

At autopsy, done about eighteen hours after death, the upper halves and the anterior lobes of the lungs were found to be of mottled purplish-red color, and almost completely consolidated. In the areas of consolidation were a few nodules, generally of a pyramidal shape with their bases in the pleura.

The pleural surfaces of these were covered with a thin fibrino-purulent exudate, which upon removal showed the yellowish color of the nodule. On section the nodules appeared to be abscesses, surrounded by consolidated pulmonary tissue, and containing a thick yellowish pus.

Throughout the consolidated portions of the lungs there were smaller, almost miliary, firm areas. In general these were in the centers of lobules about which the trabaculæ surrounding them were increased in size and quite prominent.

The other organs showed no marked changes.

Cultures from the abscesses upon glycerin-agar and in broth showed no growth.

Histological examination of sections showed approximately the same arrangement in and about the nodules as in the previous cases.

CASE VI.

Calf No. 453 was received at the Serum Laboratory on January 12, 1904, and given 100 cubic centimeters of antirinderpest serum. The next day its temperature was 40.2° C., after which it fell to normal, remained so for three days, and then gradually rose to 40.8°, afterwards again gradually falling, the course covering a period of one week. From this time until death the temperature remained normal, except for a transient rise after vaccination and one following injection of 50 cubic centimeters of virulent rinderpest blood. It died on April 27, 1904, about three and one-half months after coming to the laboratory.

At autopsy the pulmonary lesions were similar to those found in No. 491, though less extensive, being almost limited to the apices and the extreme exterior borders.

Plate cultures from the nodules after twenty-four hours at 37° showed three slightly variant types of colonies. Transplants were made from each type and studied as B. 453,¹ 453,² and 453.³

After carefully comparing their appearances in and on various media, no other difference could be seen than that of size. B. 453¹ was a trifle larger, generally, than the other two. All produced typical colonies—i. e., the ones which were larger, more opaque, and which showed a slight tendency to coalescence.

These various differences between these strains will be discussed later. Suffice it to say now, that, in general the organisms in their cultural and morphologic characteristics were identical with *B. bovisepiticus*.

Histologic examination showed a condition that agreed with that seen in previous cases.

CASE VII.

Steer No. 333 was received at the Serum Laboratory on January 6, 1904, and was given 100 cubic centimeters of antirinderpest serum. Two days later it showed some indisposition, and upon examination the temperature was found to be normal, the pulse somewhat accelerated, respirations rapid and shallow, coat staring, appetite diminished, and rumination performed with indifference. Later the temperature became irregular with an evening rise which often reached 40° C. About ten days later, after the first examination, the animal developed a shallow, painful cough, which became more marked as the disease progressed. Tuberculosis, although up to that time unknown in these Islands, was suspected and the steer was killed sixty-one days after coming to the laboratory.

At autopsy a very widespread pathologic condition of the lungs was found affecting almost the entire extent of both organs. This consisted in a chronic bronchitis with bronchiectasis upon which a broncho-pneumonia had been engrafted. The bronchial walls were injected and covered with a sticky mucopurulent material. There were several bronchiectatic cavities, one the size of a goose egg, the others smaller, which had smooth fibroid walls within which were greenish-yellow muco-caseous masses of very foul-smelling material. The parenchyma of the lungs was studded with small areas of consolidation, some of which had undergone softening. The lungs were generally edematous. There were no marked changes elsewhere save congestion and cloudy swelling.

Cultures made from the small nodules at the time of autopsy showed no growths.

Tissues were preserved in Zenker's solution and embedded in colloidin. Sections from these materials were stained with hematoxylin and eosin, thionin, genetian violet (Gram and Weigert methods), and by the ones used to demonstrate tubercle bacilli.

The lungs on section showed an intense peri-bronchial infiltration, which was composed for the most part of cells of an epithelioid character with abundant protoplasm and vesicular nuclei. There was an increase in the connective tissue of these accumulations, which were also more or less regularly enclosed by fibrous capsules, making it appear as though the epithelioid accumulations had occurred between the mucous membranes and the surrounding muscular layers. Outside of the encapsulating layers were occasional lymphoid accumulations.

The mucous membrane of the bronchi was hypertrophied and corrugated—the cells being two or three layers thick and many of them vacuolated. The lumen was filled with mucous material in which numerous leucocytes were embedded.

In several places the mucous membrane on one side was hypertrophic, while on the other it was flattened and atrophied. Projecting into the lumen from the hypertrophied side were masses composed of small round cells and stroma, about whose internal edges were a row of large cells of peculiar appearance. One of these was an enormous cell whose protoplasm was very large in amount and whose nucleus was about 3 to 4

times the size of a poly-morphonuclear leucyte, and vesicular. The other ones were not as large, but were of the same type, except one large giant cell from whose position, and the arrangement, it is plain that it is formed by the coalescence of epithelioid (endothelial) cells. The remainder of the lumen was filled with cells similar to those described.

That the structures of the growth were granulomata might be seen in this same section, for the mixture of round epithelioid cells projecting into one large bronchus was well supplied with mature and immature blood vessels. There was no giant cell formation in these epithelioid or round-celled nodules. The only place, apparently, where this was found was on the lumen side of the projecting growths, and even here the occurrence was an exception.

The trabeculae of the lungs were increased in volume. The perivascular connective tissue was increased and the parenchyma was injected.

It certainly seemed that in some places there was a tendency to syncytial formation in the mucous membranes of the bronchi, especially in those in which the peripheral growth had encroached largely upon the lumens.

In other sections the profuse fibroid overgrowth was most well marked. In such bronchi, so nearly obliterated that nothing but remnants of mucous membranes were to be seen embedded, masses of round and vesicular cells occurred. Giant cells could be seen in these, but here, too, these were nearer the peripheral margin of the nodules, and they seemed in a certain degree to have some relation to the smaller bronchioles upon which the cellular accumulations had encroached.

In other sections nodules could be seen whose centers evidenced commencing rhexis.

The liver showed a remarkable degree of fibroid change, evidently originating about the bile ducts. The tissue, besides its formative elements, containing many cells with vesicular nuclei. New gall-duct formations were also noticed.

No organisms were seen in the sections that resembled glanders bacilli, and there were no "acidfast" organisms discovered.

CASES VIII AND IX.

Calves Nos. 486 and 405.—Neither in their clinical symptoms nor in their general condition did these animals show any marked symptoms of disease. Their temperature kept within normal limits except after virulent blood injections and after vaccinations. They had no cough. They ate up to the day of death.

In the case of No. 468, which became much weaker than did No. 405, appetite was present and the animal ate its fodder eight days after it had been unable to rise to its feet.

The lesions were comparable to those described in the other calves, but in both cases they were limited to the apices of the lungs. From the lesions of each animal an organism was obtained in cultures which was similar to the ones already described—viz, to *B. bovisepiticus*.

The histologic examination showed approximately similar changes, although the processes were less advanced.

That these cases arose in the way Diekerhoff and Schneidemühl describe, there can be but little doubt. In certain animals, whose physical condition warranted slaughtering, we have noticed small areas of atelectasis, which might or might not have been congenital, a point which we have been unable to determine. In certain others we have found both acute and subacute bronchial changes, with no macroscopic parenchymatous changes. Whenever two such conditions coincided in the same animal the ideal opportunity would be afforded for the production of more serious pathologic complex, and we imagine that this is exactly what has happened in certain of these cattle.

It is an interesting feature of these cases that the bacilli of hemorrhagic septicæmia should be associated with so large a proportion.

Just what the duration of the disease has been in these animals can not be determined accurately. In the steer it was undoubtedly at least fifty days, and in the others it was probably thirty to ninety.

Infection has undoubtedly taken place in each case following the primary bronchitis, still later incipient pneumonic changes setting in.

Since the organisms present in the lesions proved to be the bacillus of hemorrhagic septicæmia, we may conclude that this is as common an inhabitant of the respiratory tract of animals here as it is in the United States.

Finally it may be said that from the above-mentioned facts that these cases are not examples of chronic infection with *B. bovissepticus*, but simply of an implanted infection in the course of other pathologic conditions, or perhaps in certain cases of a terminal infection. It seems probable, from our experience, that when primary infection with this organism occurs the disease runs a somewhat more acute course.

Another interesting fact was brought out in the bacteriologic investigation of these cases—namely, that the cultural characteristics of the *B. bovissepticus* are not constant. Even with organisms of approximately equal virulence the growths on and in media vary considerably (these are perhaps most marked in bouillon as the accompanying table will show), and with races of unequal virulence the differences are still greater.

No.	Pellicle.	Broth sediment.	Fluid.	Side of tube.	Gelatin liquefaction.	Agar.
464 ^a	Santy	Flocculo-viscid	Diffusely cloudy	Precipitate	0	Large colonies; tendency to confluence.
464 ^b	do	do	Cloudy floccul.	do	0	Fine dewdrop colonies; no confluence.
423	Whitish	do	Diffusely clouded	do	0	Moderately luxuriant, confluent growth.
415	Santy	do	Clear	Precipitate	0	Confluent growth.
453 ^a	Whitish	Viscid	Diffusely clouded	do	0	Very delicate growth of fine colonies; no confluence.
453 ^b	do	do	do	do	0	Delicate growth of small colonies; no confluence.
453 ^c	do	do	do	do	0	Do.
437	Santy	do	Clear	Precipitate	0	Small colonies; no confluence.
7	do	do	do	do	0	Very small colonies; no confluence.

No.	Milk.	Potato growth.	Glucose bouillon.	Peptone indol.	Gram's stain.	Motility.	Pathogenesis.	Remarks.
464 ^a	No change	Invisible	No gas	Positive	0	0	Guinea pig	Medium size; indol reaction brilliant.
464 ^b	do	do	do	do	0	0	do	Small indol reaction; brilliant.
423	Very faint acid	do	do	do	0	0	do	Largest of nine races.
415	No change	do	do	do	0	0	do	Medium size; indol reaction brilliant.
453 ^a	do	do	do	do	0	0	do	Small.
453 ^b	do	do	do	do	0	0	do	Very small.
453 ^c	do	do	do	do	0	0	do	Do.
437	do	do	do	do	0	0	do	Do.
7	do	do	do	do	0	0	do	Do.

It may be well to note here that, unless otherwise stated, the reaction of the media used is 1 per cent acid to phenolphthalein, and it was brought to this degree not by neutralizing with alkali and then adding hydrochloric acid, but by adding simply enough alkali to produce the right degree of acidity.

An excellent discussion of the cultural characteristics of the members of the bacilli of hemorrhagic septicemia will be found in Moore's book on "The Pathology of Infectious Diseases of Animals."

A fuller discussion of the variations of *B. bovisepiticus*, especially as regards pathogenicity, must be left for further study. From the few facts at the present time available we are not willing to draw conclusions.

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PART III.

REPORT ON PINTO (PAÑO BLANCO).

By PAUL G. WOOLLEY, M. D., *Director of the Serum Laboratory.*

Under the terms paño blanco, pinta, pinto, caraté, mal pintado, mal de los pintos, mal del pinto, peint, cute, cativi, quirica, pannus, carateus, and the spotted disease of Central America, is included a group dermatomycoses, characterized by peculiar nonpigmented patches on the skin, in the scales from which hyphæ or spores or both, of a mold-like fungus are found, which resemble in some cases *Penicilium*, in others *Aspergillus*, in still others *Monilia*.

Heretofore this epiphytic disorder has been reported from Mexico and Central and South America; another disease resembling it in some respects has been observed by Legrain in North Africa, and by Sandwith in Egypt, but, so far as I know, no previous report has come from the Philippine Islands.

The case which I wish to record is not the only one which I have seen in Manila, but it is the only one from which I have been able to obtain specimens for examination. All of the affected persons whom I have noticed have shown only the white variety, of which the following case is an example.

The history of the case is as follows: The patient was a Filipino laundryman, 15 years old, and in good health. There was no similar disease in any of his immediate family.

Upon inspection it was noticed that there were pinkish white patches, irregular in size and shape, on the ankles, dorsa of the feet, shins, knees, elbows, hands, wrists, and one on the right shoulder. This last-mentioned lesion the boy says was the one first noticed. The largest ones were over the external malleoli of the

ankles. These, the boy says, appeared after the one on the shoulder. The patches on the knees and elbows occurred later. None of these patches were of the same shape or size, nor were they definitely defined, but they shaded from their clear white centers to the normal brown of the skin. Neither were the lines of extension regular, so that the outlines of the patches were irregular and crenated. About the larger areas were smaller ones, some barely visible and of a faint pinkish white or very light-brown color.

On palpation it was evident that the skin over the larger patches was slightly rougher than the normal and that it felt somewhat thicker. The palpating finger could detect no abnormal variation in the covering of the smaller spots. There was but a minimum amount of scaling, and there was some itching.

The rate of extension had been extremely slow, for in three years the largest patch had a diameter of but 7 by 5 centimeters.

When asked regarding the cause, the boy said that the first spot came from carrying laundry baskets on his shoulder, and that the other ones followed traumata of one kind or another. There were no lesions on the palms of the hands or soles of the feet.

From one of the larger lesions on the ankle scrapings were made and examined in a solution of caustic potash (25 per cent). Among the epithelial cells branching, segmented hyphæ were seen forming a coarse mesh work. The mycelium was somewhat finer than that of *Trichophyton*; it was in general evenly refragent, but in places beaded or granular. The spores were darker in color than the rest of the organism and less refractile. An occasional fructification was found in the smears, and in these the arrangement of the spores was like that of *Penicilium*. When treated with dilute fuchsin the spores were stained a very deep red. The hyphæ showed an inner segmented arrangement with a continuous inclosing capsule.

There was nothing in any of the preparations to suggest the description of *Gastambide*. The mycelial filaments were usually long, branched, and terminated in a bunch of spores. The description given by Montoya y Flores seems to apply more accurately to the fungus of this case.

There can be no doubt of the nature of the disease. The clear white spots with almost normal looking skin can be confused with no other skin affection with which I am acquainted. Diseases caused by trichophytons are extremely common in Manila and are

known generally as "dhobie itch," which is so common in the natives that in thirty cases of skin disease taken at random in Bilibid Prison twenty-four showed trichophyton filaments in caustic potash preparations.

It is possible that a brown pinta might be confused with pityriasis versicolor should the small patches occur on the face where it is said that the latter may occur. However, in the present case the clear white color of the irregular patches, the presence of sensation and of itching, together with microscopic findings, are enough to assure a correct diagnosis.

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PART IV.

NOTES ON ANALYSIS OF THE WATER FROM THE MANILA WATER SUPPLY.

By CHARLES L. BLISS, B. S., *Chemist.*

In December, 1903, it was suggested by the Superintendent of Government Laboratories and the Director of the Biological Laboratory that a systematic investigation, both from a chemical and bacteriological standpoint, be made of the water supplied the city of Manila. After careful consideration it was decided that this investigation should consist of the usual sanitary chemical analysis and a count of bacteria, these to be made at regular intervals of one week and to extend over a period of several months. Later it was decided to examine the water for the presence of amebas and also to extend the investigation so as to include samples taken from the course of the watershed.

There were several reasons why such examinations were desirable:

(1) Up to that time no chemical analysis of the city water had been made at the laboratory, and frequent requests for statements in regard to the water had to be denied.

(2) It was supposed the water would vary considerably from time to time—not only from one season to another, but even from day to day. It seemed very probable that during the rainy season especially it would be subject to great changes within the course of a few hours, and that the results would be markedly different from the ones obtained during the dry, hot months preceding. This seemed all the more probable if any conclusions can be reached from the appearance of the water on different days. While ordi-

narily it is quite clear, a few hours after a rain it shows turbidity, which is very marked after a heavy storm. The watershed contains a quantity of suspended matter in the form of a fine silt, most of which gradually deposits when it is allowed to stand for a short time. This would naturally lead to the supposition that a variation could also be expected in the soluble constituents and in the number of bacteria.

(3) Although numerous bacteriological examinations have been made of the city water since the organization of the Biological Laboratory, these had not previously been made at regular intervals, but only at random as opportunity offered; moreover, they were undertaken for different purposes. At times merely a count of bacteria was needed; at others, a search for certain specific organisms was undertaken, so that no continuous record was made of the number of organisms found in the water.

(4) Amebas had been frequently found in the water in past times,¹ but it was not definitely known whether or not they were always present. With the prevalence of amebic dysentery in the city it was especially desirable to know whether these organisms were constantly present in the water.

The investigation was begun on December 14, 1903, and continued during seven months. It extended over the greater portion of the cooler weather, the hot season, and the beginning of the rainy season. At first weekly examinations were made; but after three months, there having been but little variation, examinations were made every alternate week. All samples were taken from a tap in the laboratory except on the following days: February 23, from El Deposito; February 29, from the Mariquina River at Santolan, and March 7, four samples from the Mariquina River (the watershed) at different points.

The taking of the samples (with the exception of those from El Deposito and Mariquina River), the methods of manipulation, the apparatus and chemicals were as nearly uniform as possible throughout the entire period, Monday of each week being selected as the day for doing this work. The last sample was analyzed on

¹Dr. Strong called attention to the presence of amebas in the water and to its unfitness for drinking purposes in his annual report as Director of the Biological Laboratory in 1902 and 1903. See Report of the Superintendent of Government Laboratories for 1902 and 1903.

July 15, for the reason that there had been an excessively heavy rainfall on the four days preceding, during twenty-seven hours of which time $17\frac{1}{2}$ inches of rain had fallen, the city and outlying districts having been completely under water from the evening of the 12th till a day or so later. A few weeks earlier an examination made eight or nine days after a rather severe typhoon showed results very nearly the same as those which had been obtained throughout the series; it was therefore thought best to take a sample very soon after the flood, rather than wait until the following week. It might be remarked that even in this instance there was but very little variation from the usual results.

Two samples were taken from one of the laboratory taps each Monday morning, after the water had been allowed to run for at least one-half hour; one was retained for chemical analysis, the other, with the usual precautions to prevent contamination, was sent to the Biological Laboratory.¹ Both examinations were begun immediately. The sample for chemical analysis was collected in a 3-liter, glass-stoppered bottle, which was first rinsed thoroughly, filled and emptied two or three times, and then filled to the neck; the stopper was well rinsed and immediately inserted. This bottle was used for no other purpose throughout the series. The determinations of nitrites, nitrates, ammonia, and oxygen consumed were started at once so as to obtain results representing the true condition of the water before any changes due to oxidation, reduction, or bacterial action could vitiate it, all necessary precautions to prevent contamination by laboratory fumes being taken. As the results in chlorine and hardness would not be affected by any changes which might take place in the water, and as the residues would be but very little if at all altered, these determinations were deferred until opportunity to make them was at hand; but in every instance they were undertaken before the end of the week. In order to represent the water as it actually came from the pipes, all analyses were made with the unfiltered liquid.

The chemical analysis consisted of the determination of total residue; fixed residue; loss on ignition; nitrogen in the forms of

¹ The counting of the bacteria was done by Mr. Clegg, of the Biological Laboratory, and the determination of the presence of amebas was also done by Mr. Clegg in conjunction with Dr. Musgrave, who has compiled his results in Bulletin No. 18, Bureau of Government Laboratories, Biological Laboratory.

nitrites, nitrates, free and albuminoid ammonia; oxygen consumed; chlorine; also hardness. As some of the methods of manipulation were different from those usually employed, a brief description is given.

Total residue was determined in a platinum dish easily holding 100 cubic centimeters, by evaporation on the water bath, and heating for thirty minutes at 95° after drying. The dish, after the weighing of the total residue, was heated uniformly to low redness for three or four minutes. It was then placed in the desiccator and weighed as soon as cold. There was practically no change in the appearance in any sample on heating; at most only a very slight darkening and but little odor were perceptible, indicating the presence of only small amounts of organic matter. The loss on ignition was therefore due largely to decomposition of carbonate, as the heat was not sufficient to volatilize any chlorides.

Nitrites.—The reagents were prepared as follows:

(a) Eight grams of naphthylamine hydrochloride were dissolved in water, 8 cubic centimeters of concentrated hydrochloric acid added, and the solution diluted to one liter.

(b) Sulphanilic acid, a saturated solution in water containing 5 per cent concentrated hydrochloric acid. The test was made by placing 50 cubic centimeters of the water in a Nessler tube, adding 1 cubic centimeter of each of the above solutions, and mixing well by gently shaking. After thirty minutes the color was noted. In no instance did the depth of color indicate more than a very faint trace of nitrites, and very frequently none developed. Therefore a quantitative estimation could not be made.

Nitrates.—The aluminum reduction method was employed. After a few hours, when the reaction was completed, the ammonia formed, together with the free ammonia originally present in the water was estimated directly by the Nessler process; the free ammonia (determined in another portion of the sample) was deducted, the remainder being the ammonia formed from the nitrates. In the present series the determination of nitrates was begun within a few minutes after the sample was received, and the Nesslerizing was done on the following day. If nitrites are present in appreciable amount an allowance should be made for them.

A little more than 50 cubic centimeters of the sample were placed

in a 250 cubic centimeter glass-stoppered bottle; 2 cubic centimeters of sodium hydroxide of 33 per cent strength and 2 grams of aluminum filings were added, and the loosely stoppered bottle allowed to stand at room temperature until the next day. The solution was then filtered, with all precaution, into a tube, filling it to the 50 cubic centimeter mark. This solution was then Nesslerized, the necessary correction for free ammonia being made.

Free ammonia.—A round-bottom flask of 1 liter capacity with short neck was connected with a condenser 1 meter in length, the Nessler tube being slipped over the other end. On the day the sample was received the distilling flask was rinsed with distilled water; about 200 cubic centimeters of a solution of distilled water containing 1 gram of sodium carbonate were then added, and the greater part of the water distilled off, until the apparatus was free from ammonia. After cooling, 500 cubic centimeters of the sample were added to the residue, and the distillation continued at such a rate that a Nessler tube was filled to the 50 cubic centimeter mark in about ten minutes. Three tubes of 50 cubic centimeters each were collected and Nesslerized. As a matter of fact one tube would usually have been sufficient, at most two; for the third one never showed more than a slight trace of ammonia, and often none at all.

Albuminoid ammonia.—Fifty cubic centimeters of alkaline permanganate (8 grams of permanganate, 200 grams of potassium hydroxide, and 1,100 cubic centimeters of water, evaporated to 1 liter) were now added to the contents of the flask and distillation was continued, four tubes of 50 cubic centimeters each being collected. The distilling apparatus was used for no other purpose throughout the series, and it was always well protected from fumes.

Nesslerizing.—The Nessler solution was prepared according to the usual method and the standard solution of ammonium chloride contained (0.03812) gram of pure ammonium chloride in one liter. One cubic centimeter represents 0.00001 gram of nitrogen.

The distilled water in the laboratory was found to be free from even the slightest perceptible trace of ammonia; it was tested each time. A number of Nessler tubes were thoroughly rinsed with this ammonia-free water, then filled to the 50 cubic centimeter mark; portions of the standard ammonium chloride solution were measured in from a normal capillary pipette reading accurately

in hundredths of a cubic centimeter. The amounts used were 0.2, 0.4, 0.6, 0.8, 1, 1.2, 1.5, 2, and 2.5 cubic centimeters. Two cubic centimeters of the Nessler reagent were added to each, and also to the tubes containing the ammonia (free, albuminoid, and that from the nitrates). After allowing thirty minutes for development of the color, the comparisons were made. The results were expressed in terms of nitrogen per million.

Oxygen consumed.—The solution of pure potassium permanganate contained exactly 0.3953 grams in 1 liter. One cubic centimeter represented 0.0001 gram available oxygen, and the solution of pure ammonium oxalate was of exactly equivalent strength. These two solutions were kept in a dark closet and were standardized from time to time.

One hundred cubic centimeters of the water were measured into a 200 cubic centimeter Erlenmeyer flask from a pipette; 2 cubic centimeters of pure concentrated sulphuric acid were added, and then 10 cubic centimeters of the permanganate solution; the flask was then suspended in a boiling-water bath for thirty minutes. It was then taken out and 10 cubic centimeters of the oxalate solution were immediately added. The solution became colorless within a minute or so, and was then titrated with the permanganate till a faint pink color appeared, which remained permanent for a few moments. This determination was made in duplicate, two flasks being carried through at the same time. In no instance did the two titrations vary more than 0.05 cubic centimeters, and usually they were identical. For convenience, the titrations were made with a very narrow pipette graduated in 0.05 cubic centimeters. Results were expressed as parts of oxygen consumed per million.

Chlorine.—A solution of pure, recrystallized and dried silver nitrate was made containing 4.7940 grams in 1 liter. One cubic centimeter represented 0.001 gram of chlorine. A 5-per cent solution of potassium chromate freed from chlorides by precipitating with silver nitrate was used. The chlorine determinations were made in duplicate. Two portions of 250 cubic centimeters each were measured into casseroles of about 300 cubic centimeters capacity, carefully evaporated to about 50 cubic centimeters, 1 cubic centimeter of the chromate solution added, and the mixture titrated with the silver solution. By using one as a comparison

the first change in color from a pure to a reddish yellow was noted very sharply, and the two results never varied by more than 0.05 cubic centimeter; usually they were identical. A pipette similar to the one employed in the titration with permanganate was used. Results were expressed in terms of chlorine per million.

Hardness.—A solution of pure calcium chloride was diluted so that 1 cubic centimeter contained 0.0002218 grams, or represented the equivalent of 0.0002 gram of calcium carbonate. Fifty cubic centimeters of this solution should require exactly 14.25 cubic centimeters of standard diluted soap solution in order to form a lather which covered the surface of the liquid and persisted for five minutes. The soap solution was standardized each time and the proper value obtained, 50 cubic centimeters of the above calcium chloride being used for this purpose. This was compared with 50 cubic centimeters of the sample treated in the same manner, and the degree of hardness determined by reference to published tables. In the two or three instances, when the results varied from those usually obtained, and in all cases where any doubt as to the exact end-point existed the determinations were repeated.

Analyses of the Manila water supply.

[Results are given in parts per million.]

Laboratory No.	Location.	Date.	Total residue.	Fixed residue.	Loss on ignition.	Nitrites.	Nitrates.	Free ammonia.
1739	-----	Dec. 14, 1903	220	190	30	-----	0.150	0.0079
1745	-----	Dec. 21, 1903	181	148	33	-----	.271	.0049
1756	-----	Dec. 28, 1903	188	152	36	-----	.100	.000
1780	-----	Jan. 4, 1904	188	142	46	0	.112	.008
1781	-----	Jan. 11, 1904	191	160	31	0	.080	.002
1803	-----	Jan. 18, 1904	179	148	31	0	.110	.006
1821	-----	Jan. 25, 1904	176	152	24	0	.198	.002
1840	-----	Feb. 1, 1904	178	152	26	0	Trace.	Trace.
1880	-----	Feb. 8, 1904	162	142	20	(1)	.100	Trace.
1922	-----	Feb. 15, 1904	168	152	16	0	.138	.002
1944	Deposito	Feb. 23, 1904	168	145	23	0	.274	.006
1996	Santolan	Feb. 29, 1904	164	138	26	0	.292	.028
2025	Mariquina River	Mar. 7, 1904	160	134	26	0	.124	.036
2026	do	do	153	127	26	0	.139	.021
2027	do	do	174	147	27	Trace.	.112	.028
2028	do	do	173	145	28	0	.136	.024
2069	-----	Mar. 14, 1904	167	138	29	(2)	.220	Trace.
2157	-----	Mar. 28, 1904	173	147	26	(2)	.220	Trace.
2225	-----	Apr. 11, 1904	174	138	36	Trace.	.158	.002
2308	-----	Apr. 25, 1904	165	136	29	Trace.	.200	Trace.
2352	-----	May 9, 1904	180	145	35	(1)	.158	.004
2385	-----	May 23, 1904	169	135	34	(1)	.120	Trace.
2434	-----	June 6, 1904	196	150	46	(2)	.360	Trace.
2464	-----	June 20, 1904	174	138	36	0	-----	Trace.
2487	-----	July 5, 1904	178	150	28	0	.360	Trace.
2511	-----	July 15, 1904	191	159	32	0	.220	Trace.

Analyses of the Manila water supply—Continued.

[Results are given in parts per million.]

Laboratory No.	Location.	Albumin- amonia.	Oxy- gen.	Chlo- rine.	Hard- ness.	Bacte- ria.	Ame- bas.	Remarks.
1739	-----	0.078	1.90	2.13	85.7	400	—	Heavy rain during few days preceding; water very turbid.
1745	-----	.073	1.85	2.23	85.7	550	—	Do.
1756	-----	.031	.875	2.60	90.0	600	—	Water distinctly turbid.
1780	-----	.052	.86	2.40	94.3	460	—	Water slightly turbid.
1781	-----	.034	.85	3.00	101.5	250	—	Do.
1803	-----	.038	.65	3.04	94.3	200	—	Do.
1821	-----	.044	.65	3.00	87.1	200	—	Do.
1840	-----	.048	.90	3.00	95.0	150	—	Water almost perfectly clear.
1880	-----	.048	.90	2.60	104.0	125	+	Do.
1922	-----	.044	.95	3.20	95.0	150	+	Do.
1944	Deposito	.052	.95	3.30	109.0	120	(4)	
1996	Santolan	.086	1.25	3.40	97.0	112	0	
2025	Mariquina River.	.100	1.60	3.60	97.0	-----	+	All contained deposit, apparently organic matter and silt. Determinations of residue were made with filtered water. Each gave slight dark color and odor on ignition.
2026	do	.080	1.50	3.20	93.2	208	+	Do.
2027	do	.062	1.07	3.60	93.2	105	+	Do.
2028	do	.060	1.35	3.60	94.8	267	+	Do.
2069	-----	.048	1.00	3.88	95.0	120	+	
2157	-----	.052	.82	4.14	101.8	120	0	
2225	-----	.062	1.07	3.80	97.8	108	+	
2303	-----	.048	1.20	4.40	95.0	125	+	
2352	-----	.064	1.20	4.20	98.0	175	+	
2385	-----	.042	1.05	4.40	92.0	130	+	
2434	-----	.074	1.60	4.20	91.0	206	+	Heavy rains just previous; water turbid; slight sediment. Slightly dark color and odor on ignition.
2464	-----	.040	-----	36.0	83.0	135	+	No rain immediately preceding.
2487	-----	.050	1.70	2.80	71.4	450	+	Very turbid. Heavy rains.
2511	-----	.068	2.20	3.16	58.8	(5)	+	Very turbid. (Flood.)

¹ Very faint trace.² Faint trace.³ Distinct trace.⁴ Not sufficient sample. A sample taken following week, bacteria, 100; amebas, 0.⁵ Rapid growth over surface prevented count.

The counts of bacteria and determinations of the presence of amebas were made by Dr. W. E. Musgrave and Mr. M. T. Clegg, both of the Biological Laboratory.

On examining the above table the following maxima and minima appear during the period covered by the report :

	Minimum.	Maximum.
Total residue	153	220
Fixed residue	127	190
Loss on ignition	16	46
Nitrites (N)	0	Trace.
Nitrates (N)	Trace.	.36
Free ammonia (N)	0	.036
Albuminoid ammonia (N)	.031	.100
Oxygen consumed (O)	.65	2.20
Chlorine (Cl)	2.13	4.40
Hardness	58.8	109

As will be seen despite the great variation in the weather conditions the differences in analytical results were not very great. At times the chlorine ran as high as 4.40 indicating some contamination, but these maxima were only transitory. However, a water may show a very high degree of purity, so far as this can be determined by chemical analysis, and yet be unfit for drinking purposes because of either the large number of micro-organisms which it contains or because of their nature; so that it may be possible to convey the etiological factor of typhoid fever, cholera, dysentery, etc., by a drinking water which chemically would be pronounced unobjectionable. The chemical analysis may indicate a probable pollution with sewage or with other matter which may be suspicious or dangerous at all times, and it may condemn such a water. However, in the case of the Manila supply the long series of analyses gave such results that no one would be justified, even at the worst, in stating from a chemical standpoint that this water was either injurious or deleterious to the public health. For this reason, as has been repeatedly pointed out by others, a bacteriological examination is essential.

However, in glancing over the number of bacteria it will be seen that the maximum, on December 28, was 600, and this large number was quite unusual, the average being below 250 organisms to 1 cubic centimeter and in many cases even below 150. Bacteriologically, therefore, the water may not be regarded as very suspicious, especially since the general series of determinations did not demonstrate pathogenic organisms to be present, and indeed typhoid fever is of but rare occurrence in Manila. During the cholera epidemic it does not seem likely that any of the cases could have been referred to direct infection from the Manila water supply.

A factor however, which, apart from the chemical and bacteriological analysis, is the most important, is shown in the last column, where it is demonstrated that amebas, whether pathogenic or not, are almost constantly present in the water supply, and Dr. Musgrave and Mr. Clegg, of the Biological Laboratory, in Bulletin No. 18, "Amebas: Their Cultivation and Etiologic Significance," have shown that amebic dysentery can be sometimes produced in monkeys by cultures made from the water supply. Neither a chemical analysis nor a bacteria count will demonstrate the presence of these dangerous factors of disease, and consequently, in the Tropics at least, if we wish to obtain a fair idea of the condition of the water, a determination of the presence or absence of amebas is necessary.

The ordinary sanitary analysis does not include an examination for substances which in themselves are injurious. The quantities of ammonia, nitrate, chlorine, etc., found in an ordinary water are harmless—they are simply indicative of a possible pollution, but before judgment can be passed in regard to the sanitary analysis, the source of the water, the geological conformation of its surroundings, and so forth, must be taken into consideration. An amount of chlorine, for example, which would be perfectly normal in water from one locality might indicate contamination with sewage in that from another. The total residue in this series was always below 200 parts per million excepting on one occasion which was after a heavy rain.

Very little darkening of the residue on heating was noted in any sample; sometimes there was none, so that in this respect no criticism can be made of the water. The presence of nitrites in measurable quantity is sufficient ground for condemning a water as a rule, for nitrites indicate bacterial action. However, in this water nitrites were frequently absent altogether, and only on one occasion were there more than a trace, this occurring immediately after a heavy rain.

The amount of nitrates was always very low. The highest amount of free ammonia found was 0.008 per million, excepting in five samples taken from the Mariquina River itself where the results varied from 0.0021 to (0.036), so that by the time the water from the river reaches Manila the ammonia has largely become oxidized to nitrate. This is borne out by the fact that the amount of nitrate in those samples which were taken on March 7 was

lower than the usual quantity obtained. Albuminoid ammonia, as a rule, was also quite low. None of these factors, therefore, would indicate a contamination of the river water.

The same may be said of the amount of oxygen consumed. No figure exceeding 2 was found excepting in one instance, and that was three days after the great flood in July. The amount of oxygen consumed seemed to be greater immediately after heavy rains.

The low results obtained in the investigation of chlorine are favorable indications as to the quality of the water. One would expect rather higher chlorine values in the waters of the Philippine Islands owing to the proximity to the sea, but this is not as a rule the case, as analyses of waters from other localities have demonstrated.

After heavy rains more or less insoluble suspended matter, both inorganic and organic, is rinsed into the supply. However the turbidity of the Manila water is generally due to a very fine silt which has its rise in some of the clay beds at the source of the river, and is for this reason harmless. Therefore the analyses made in the Chemical Laboratory show that the water supply of the city of Manila is of a very good quality, but the constant presence of amebas, as demonstrated in the Biological Laboratory, render the water unsafe for drinking purposes unless it is boiled.

PART V.

FRAMBOESIA: ITS OCCURRENCE IN NATIVES OF THE PHILIPPINE ISLANDS.

By PAUL G. WOOLLEY, M. D., *Director of the Serum Laboratory.*

In a recent visit to Benguet Province, in the central part of northern Luzon, I became interested in a peculiar disease which was called "Lépra" by the native Igorrotes, this term having been taught them by the Spaniards. Inasmuch as I was entirely unprepared for a complete study, and because it is extremely difficult to obtain any history or to make complete examinations of these people, the following report will be meager, but it is interesting, since I feel certain that the disease is one closely related to if not identical with frambœsia.

In Baguio, Benguet, I saw, with Dr. Thomas of the Civil Sanitarium, two cases; one, a woman aged about 35 years, and her son, the latter of some 15 years, both of whom presented small raspberry growths upon the face. In the mother, these were situated at the corners of the mouth; in the son, in the nasolabial folds, and they much resembled the growth pictured in the New Sydenham Society's Atlas (Fasc. XIV, Pl. B. fig. 8). Both of these persons showed pigmented scars on the neck and face. At a later time I searched for these two people in their native town of Agno, Benguet, but was unable to find them.

The case from which I procured the tissue which I will describe below, I saw one morning as Mr. Barron—the sanitary inspector of the province—and I were returning from a long trip in the mountains. As we stopped to rest at a little native village, we

noticed that several of the children had peculiar looking, sluggish ulcers on their legs, necks, or bodies, and also showed signs of considerable anemia. We asked concerning this affection and were told that the Spanish called it "Lepra," and that many people had it. This was all we could learn, except that the sores were not painful, though they showed the effects of scratching, and that they eventually healed. One grown person, a woman, was found who had similar lesions on her neck. Whether she had others elsewhere we were unable to discover. One lesion on the neck (see Sydenham Society's Atlas, Fasc. XIV, Pl. LXXXV, upper left-hand corner of lower figure) was a shallow ulcer with a firm grey base, a well-defined, firm margin, a slight yellowish secretion, not surrounded by any appreciable induration, not painful, and situated on the neck just below the angle of the jaw. This was excised and preserved in commercial alcohol. Near it were some pigmented areas, somewhat darker than the normal skin, not appreciably thickened, and possessed of sensation, and which the woman said were at the sites of healed ulcers similar to the ones we saw. (New Syden. Soc. Atlas, Pl. LXXXV.) An infant which the woman carried had similar lesions on its legs, face, and neck. Another child of the same woman, which, however, we did not see, was said to be afflicted in the same manner.

The gross appearance of the lesions would lead one to think of leprosy, tuberculosis, epithelioma, syphilis, or yaws. Leprosy I think may be excluded. There were no cases of outspoken leprosy among the persons of the pueblo in which these persons lived. There were no anesthesias or leucoplakias in any of the cases examined. The histologic examination was negative. Tuberculosis could be excluded since neither the lesions nor the scars had the classic appearance of lupus, nor was the histologic evidence sufficient to support such a diagnosis. Epithelioma could only be excluded by microscopic examination. All in all the cases seemed most like syphilis. None of the people of the pueblo showed outspoken signs of this disease, although, as stated, a complete examination could not be obtained. The inhabitants of this region generally are rigidly moral and rigorous punishment is inflicted upon any who overstep the bounds. But while they live morally clean lives, their physical surroundings are filthy, which may account for the modification of the frambœsial lesion and the predominance of

infection. Treatment, of course, has not been tried, so one can not say what effect mercury or iodides might have, but from the evidence that I have at present I incline to the idea that the cases seen were examples, not of syphilis, but of frambœsia.

The tissue was embedded in celloidin, and the sections which were made later were stained with hæmatoxylin and eosin, Unna's polychrome blue, Gram's stain, and by the tubercle bacillus methods. Microscopic examination showed that the lesion consisted of a marked hyperplastic acanthosis, with a round-celled infiltration of the underlying, and especially of the perivascular, connective tissues. The first impression was that one was dealing with an epithelioma, but closer examination showed this to be a delusion. The acanthus layer of the skin was thickened and prolonged in strands and columns of bizarre shapes. In many places in this hyperplastic epithelium there were larger or smaller islands of connective tissue, each, apparently, representing the path of a blood vessel. In the centers of such areas and about the vessel were collections of small round and plasma cells, but this small-celled infiltration was most marked in the larger strands of the sub-malpighian connective tissue. Here the increase of these formative elements was remarkably prominent, and there was throughout the sections the same perivascular arrangement. Within these areas there were occasional leucocytes; there were also fibroblasts in varying stages of development, and a number of plasma cells were present within the round cell accumulations. At the site of the ulceration the structure of the lesion was modified by the destructive process. Here all the layers were invaded by a multitude of polymorphonuclear leucocytes, the blood vessels were widely dilated, and there was a certain amount of superficial degeneration, but the structure of the lesion in the not degenerated parts was still perceptible. So far as the arrangement of the layers of the skin was concerned there was no distortion. In many parts of the sections a peculiar appearance was seen which gave one the same impression as that produced by the scales of a fish, or by the overlapping of shingles upon the roof of a house. This was apparently due to the fact that certain of the acanthus cells took a more intense stain on one side. In this phenomenon (seen best in polychrome blue specimens), the nucleus did not participate. In none of the sections, and these included the whole of the lesion studied, were there

any giant cells, tubercle of lepra bacilli, and no evidence of cell inclusions was seen.

As for the occurrence of such lesions in frambœsia, and their distribution, little can be said excepting as quotations from authors who have had considerable experience with the disease. Manson says, in discussing the distribution of the yaws, that they may be scattered over the entire body, or the crop may be limited to one or two growths, or they may be confined to a circumscribed region of the skin. Moreover, there may be successive crops evolved, especially when the person affected is debilitated. Morris remarks that the disease in adults is more chronic than in children. When the yaw develops normally it does not ulcerate, but Manson says that the tumors instead of being absorbed, may break down and ulcerate, the ulceration usually being confined to the yaw itself, although it may go deeper and give rise to extensive sores. With the development of the deeper and more extensive forms of ulceration, the typical lesion of frambœsia may disappear for a time, or perhaps permanently. If such is the case, the ulcerations are said not to be infective and to not communicate the disease, although they may persist for years. Nicholls, quoted by Mason, states that ulceration occurs in about 8 per cent of the cases. There is no histologic description of the variety of the lesion which I have encountered to which I can refer, though from Unna's and Charlouis's description of the typical yaw it is but a modification of the latter. These writers speak of the true yaw as a cutaneous plasmoma complicated by epithelial hyperplasia and hyperkeratosis. Except that the lesion described above is not raised it certainly corresponds in many details with Unna's and Charlouis's description.

REFERENCES TO LITERATURE.

- MANSON. Tropical Diseases. London, 1903.
 MORRIS. Diseases of the Skin. Philadelphia, 1898.
 HYDE. Diseases of the Skin. Philadelphia, 1900.
 UNNA. Hautkrankheiten. Berlin, 1894.
 SCHEUBE. Die Krankheiten der warmen Länder. Jena, 1900.

SECTION STAINED WITH HÆMATOXYLIN AND EOSIN. (ZEISS COMP. OC. 6, OBJ. A. A.)

This shows the hyperplasia of the acanthus layer of the skin, the dilated blood vessels, and the perivascular accumulations of small round and plasma cells. Photomicrograph by Martin.

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