REPORT ON M. PASTEUR'S RESEARCHES ON RABIES AND THE TREATMENT OF HYDRO- PHOBIA BY PREVENTIVE INOCULATION.

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PART I.

M. PASTEUR'S FIRST EXPERIMENTS.—FACTS CONCERNING THE TRANSMISSION OF HYDROPHOBIA.—*The Incubation-Period shortened by Inoculating the Virus beneath the Dura Mater.*—*All Forms of Hydrophobia proceed from the same Virus.*—*Symptoms are excessively Varied.*—*The Saliva in Rabies may provoke Death from Three Different Causes.*—*The Medulla Oblongata, Spinal Cord, and Encephalon of Rabid Subjects are Virulent.*—*Virulence affected by Decomposition.*—*Different Methods of Inoculation and their Results.*—*The Nature of Rabies Confirmed by the Quantity of Inoculation Fluid Injected.*—*Furious Madness produced by a Smaller Quantity of Virus than Paralytic Madness.*—*Virus does not Travel along the Peripheral Nerves to the Central Nervous System.*—*Initial Period of Madness disappears; the Symptoms reappear after a long Interval, and Result in Death.*—*Dogs Rendered Refractory to Madness by Inoculation.*—*A Summary of Facts.*

M. PASTEUR'S FIRST EXPERIMENTS.—M. Pasteur's first experiments in connection with hydrophobia were made in December, 1880. He collected, by means of a camel's hair brush, some mucus from the mouth of a child aged 9 years, which had died from hydrophobia in M. Lannelongue's wards in the St. Eugénie Hospital. The child had been bitten in the face by a mad dog in a village—Choisy-le-Roi—near Paris. It was ascertained beyond doubt that the dog was mad.

The mucus was removed from the child four hours after death. Two rabbits were inoculated with it; these died thirty-six hours afterwards. Other rabbits were then inoculated from them, some with the saliva, others with the blood. The second series of rabbits died more rapidly after inoculation than the first, the third series more rapidly than the second, and so on, through several successive series of animals.

M. Pasteur and his pupils, MM. Chamberland and Roux, who helped him to carry out his experiments, detected in the blood of the animals experimented on the presence of a micrococcus presenting the aspect of a figure of 8, indicative of a diplococcus. (Subsequent researches have demonstrated that it was a diplococcus present in normal human saliva.) M. Pasteur cultivated this diplococcus in veal-broth, through several successive generations. He observed that, when it was used for inoculating rabbits, death resulted, accompanied by the same symptoms as those which followed the inoculations made with the saliva and blood removed from the first series of rabbits, which had been inoculated from the saliva taken from the child dead from hydrophobia. This microbe, then, appeared "to be the veritable and sole factor in the new malady."¹

M. Pasteur, not being certain whether the condition called into existence by his inoculations was identical with hydrophobia, presented his note to the Academy of Sciences, headed "Sur une Maladie Nouvelle provoquée par la Salive d'un Enfant mort de la Rage" (On a New Disease produced by the Saliva of a Child dead from Hydrophobia). M. Pasteur seriously doubted that the illness caused by his inoculations was hydrophobia, and stated, in his note to the Academy, that the animals experimented on died without passing through an incubation-period; whereas M. Galtier, Professor at the Veterinary School of Lyons,² had observed that rabbits inoculated from rabid dogs did not exhibit symptoms of hydrophobia until from four to forty days after inoculation; neither had M. Galtier observed the lesions at the necropsies of his animals that M. Pasteur had ascertained to exist in his—an emphysematous condition of the areolar tissue of the groins

and axillæ, areas of pulmonary apoplexy, and swelling of the lymphatic glands of the trachea, groin, and axillæ, accompanied by hæmorrhage. The condition of the blood closely resembled that of animals dead from charbon. Blood removed from M. Galtier's rabbits, which died from hydrophobia, did not communicate the malady to other rabbits. Dogs inoculated with the blood or saliva taken from M. Pasteur's rabbits did not exhibit either dumb nor furious madness. M. Pasteur therefore abstained from arriving at any definite conclusions concerning the nature of the affection which proved fatal to the rabbits experimented on, and concluded his note by saying that he should continue his experiments.

It is unnecessary here to reproduce the objections which M. Colin, of Alfort, made to the contents of M. Pasteur's note; or the communications of MM. Lannelongue and Raynaud concerning the transmission of hydrophobia to rabbits, after inoculating them with fragments of salivary glands, and with the saliva removed from the same child which had furnished M. Pasteur with his original inoculating matter; inasmuch as all these questions are decided in a letter from M. Pasteur to the Academy of Medicine,³ in which he states that the new disease (*nouvelle maladie*) he had produced in rabbits, and consequently that induced by M. Lannelongue, were not hydrophobia, but another pathological condition, due to the presence of a micro-organism, found in the buccal cavity of human beings. The following week, M. Vulpian confirmed M. Pasteur's statement, in a communication describing his own experiments. He had killed rabbits by injecting saliva from healthy people, and had found in the blood of these rabbits the same micro-organism as that described by M. Pasteur, who had used, in his control experiments, saliva taken from children with broncho-pneumonia. M. Pasteur's letter, and M. Vulpian's communications, close what may be called the preliminary period of M. Pasteur's experiments on hydrophobia. These experiments can be divided into three distinct stages—the first, or preliminary stage, which we have just described; the second, in which the different methods of transmitting hydrophobia are studied; the third, and last, in which the different methods of inoculating animals, and the human subject, are described.

In our report, we propose following this order, which seems to us the most logical, and also essential in making a clear exposition of the facts on which is based the method of inoculation for hydrophobia.

FACTS CONCERNING THE TRANSMISSION OF HYDROPHOBIA.—On May 30th, 1881, M. Pasteur stated at the Academy of Sciences, that, as the blood of rabid animals failed to communicate hydrophobia, he and his assistants, MM. Chamberland, Roux, and Thuillier, had determined to test the truth of the view expressed by M. Duboué in 1879, in a note presented to the Academy of Medicine by M. Bouley. The conclusions arrived at by the author appear in the *Bulletin de l'Académie*, 1879, page 865. M. Duboué's opinions were purely theoretical. He supposed that the nervous system, and especially the medulla oblongata, were important factors in the development of rabies. This supposition was not confirmed by the experiments of M. Galtier, of Lyons,⁴ who made more than ten inoculations with fragments of cerebellum and spinal cord removed from mad dogs, all of which were unsuccessful.

M. Pasteur, however, had obtained more successful results, and had, by inoculating animals with medulla oblongata, and frontal portions of the cerebral hemispheres, and the cerebro-spinal fluid, removed from animals dead from hydrophobia, succeeded in transmitting the disease. The incubation-period varied from eight to forty days. These experiments demonstrated that the nervous system, as well as the saliva, contained the virulent element. This was a most important fact arrived at, inasmuch as the saliva always contains more or less micro-organisms capable of complicating and masking the action of the virus of rabies by causing death to arrive more rapidly. Thus, with rabid saliva, experiments are less demonstrative than those made with portions of the central nervous system, which can always be obtained, by observing certain precautions, free from external contamination.

In this same note to the Academy of Sciences, M. Pasteur announced another important discovery. If, instead of inoculating the rabid matter by subcutaneous injections, it were, after trephining the cranium, placed on the cerebral surface, the uncertainty which attached to former methods was thus avoided. Hydrophobia was invariably manifested; not one inoculation failed. Sometimes dumb

¹ *Bulletin de l'Académie de Médecine*, 1881, page 380.

² Galtier, *Bulletin de l'Académie de Médecine*, 25 Janvier, 1881.

³ Note on Hydrophobia by M. Pasteur and his assistants, MM. Chamberland, Roux, and Thuillier, *Comptes Rendus de l'Académie des Sciences*, tome xcii, p. 1259.

¹ *Comptes Rendus de l'Académie des Sciences*, 1881, tome xcii, p. 102.

² *Ibid.*, 1879, tome xci, p. 2438.

madness was developed, sometimes furious, that is to say, both forms of hydrophobia.

The Incubation-Period shortened by Inoculating the Virus beneath the Dura Mater.—The incubation-period, after inoculation by subcutaneous injection, is excessively variable; sometimes it is very long; inoculation after trephining is followed by a much shorter incubation-period. An interval of three weeks is the maximum of time that elapses between the introduction of the rabid virus under the dura mater, and the appearance of the first symptoms of hydrophobia. It is needless to dwell on the importance of this note, which demonstrates the virulence of the nervous system, and furnishes an accurate method of inoculation for hydrophobia, which, by shortening the incubation-period, saves much valuable time for the experimenter, and relieves him from anxiety. Rather more than a year later, M. Pasteur and his assistants completed the note of which we have given the substance above, and described fresh facts, which may be enumerated as follows.⁶

1. *All Forms of Hydrophobia proceed from the same Virus.*—Dumb madness and furious madness, indeed, all forms of hydrophobia, proceed from the same virus.

2. *Symptoms are excessively Varied.*—The symptoms of hydrophobia vary. Each separate case may be said to have its special symptoms. There are grounds for supposing that this non-uniformity of the symptoms depends on the regions of the central nervous system in which the virus localises and develops.

3. *The Saliva in Rabies may provoke Death from Three Different Causes.*—In saliva, the virus of hydrophobia is combined with different kinds of microbes. This saliva, used for inoculation, may cause death from three different morbid agents, from the microbe presenting the form of figure 8 (which killed the rabbits M. Pasteur used for those experiments, which he fully described in the note, entitled, "Sur une maladie nouvelle, provoquée par la salive d'un enfant mort de la Rage," to which we have referred at the beginning of this report), from the formation of a considerable quantity of pus, and from rabies.

4. *The Medulla Oblongata, Spinal Cord, and Encephalon of Rabid Subjects are Virulent.*—The medulla oblongata, encephalon, and spinal cord found in a person or animal dead from rabies are virulent. The medulla oblongata is always virulent; the spinal cord is also always virulent in parts, and frequently throughout its entire length; the encephalon may be virulent, sometimes only in certain parts, sometimes in its entirety.

5. *Virulence affected by Decomposition.*—So long as the nervous system does not undergo decomposition, its virulent properties remain intact. M. Pasteur kept, during three weeks, the brain of an animal dead from hydrophobia; it was kept submitted to the influence of a temperature of 12° C. (53.6° Fahr.), and it was still virulent.

6. *Different Methods of Inoculation and their Results.*—In order to inoculate rabies with certainty and rapidity, one of two methods may be adopted; inoculating the cerebral surface immediately beneath the arachnoid membrane, after trephining the cranium; or venous injection of the virus. These methods always result in the appearance of hydrophobia, and greatly shorten the incubation-period, which, on an average, lasts from six to ten days.⁷

7. Hydrophobia, communicated by venous injection of the virulent matter, is frequently different from the form of hydrophobia, resulting from the bite of a mad dog, or from cerebral inoculation; this last method generally produces furious madness, and that of venous injection, dumb madness. In virtue of the region of its localisation, it might be called medullary hydrophobia; paralysis frequently occurs, the characteristic bark of rabies is more or less rare, the animal is seldom furious, and frequently tormented by most violent itching. The symptoms exhibited by animals, inoculated by venous injections, indicate that the spinal cord is first attacked, that is to say, that the virus is localised and developed there.

8. If death do not follow inoculation of rabid saliva or a venous injection of virulent blood, the animal is not, on that account, exempt from the danger of contracting hydrophobia, on a further inoculation, after trephining, or a venous injection of virulent matter. These facts are directly opposed to the statements made by M. Galtier, on August 1st, 1882, at the Academy of Sciences.

9. Hydrophobia is never spontaneously arrested when the acute

period is reached; it may be at the period of initial symptoms, but only then. Sometimes hydrophobia is arrested at this period, and, after a long interval has elapsed, the malady reappears, and death ensues.

M. Pasteur also stated in the above note that he had then at his laboratory four dogs, all refractory to the influence of the virus of hydrophobia. These dogs were inoculated at the same time as other dogs; but they had escaped the disease, whilst the others had contracted it, and died. In one of the four dogs refractory to hydrophobia, the first symptoms of the disease spontaneously disappeared, and did not reappear. The animal was then inoculated twice, after trephining had been effected, and yet remained free from hydrophobia. The three other dogs did not exhibit any symptoms of hydrophobia, but were nevertheless refractory to the influence of the virus. M. Pasteur hesitated to decide between two probabilities: one, that the dogs were naturally refractory to the virus of hydrophobia, or that they had acquired immunity from it by the slight attack they had contracted.

In this same note, M. Pasteur suggests the probability of arriving at a method of inoculation which might be applied to human subjects.

More than a year later on—February 25th, 1884—M. Pasteur stated, at a meeting of the Academy of Sciences, that he felt justified in asserting that there are not any dogs which were constitutionally refractory to hydrophobia; but he believed that he had found a method which rendered them so. He abstained from giving fuller details, or making positive affirmations, until his experiments were carried on further. It might be possible that the incubation-period in these animals would be very long.

These experiments suggest the possibility of inoculation becoming a prophylactic measure for hydrophobia. His researches, nevertheless, were not solely directed by this idea; we find in the same note that M. Pasteur was engaged in ascertaining the different methods of transmitting hydrophobia,⁸ also the differential characteristics of this disease, special to each animal species, and the region in which the virus becomes localised.

Whilst studying the comparative virulence of the spinal cord—especially at the lumbar enlargement, and that of the medulla oblongata, of animals inoculated by venous injections, and killed when the first symptoms of hydrophobia set in, M. Pasteur ascertained that the cord may have virulent properties, but not the medulla oblongata. The hypothesis that M. Pasteur formulated a year previously, which assumed that the form of hydrophobia resulting from venous inoculation might be called medullary hydrophobia, because its principal symptom is paralysis, was thus verified.

M. Pasteur, in the further course of his researches concerning the localisations and development of the virus of hydrophobia, observed that not only was it present in the central nervous system, and in the salivary glands, but also in the peripheral nerves. Animals inoculated with fragments of the pneumogastric nerve, and also of the sciatic, were seized with hydrophobia; this fact explains that intense excitability which in human subjects is manifested by that remarkable symptom in hydrophobia known as *ærophobia*; the entire peripheral nervous system is affected.

M. Pasteur also ascertained that the saliva and the salivary glands, the parotid, maxillary, and sublingual, were as virulent in dogs seized with hydrophobia developed from venous or cerebral inoculation, as in those attacked by spontaneous hydrophobia (*rage dite spontanée*). Another fact also determined by M. Pasteur's researches is, that the virulence of the central nervous system remains intact during several weeks, if decomposition be prevented; likewise pure virus, kept in glass tubes, hermetically closed, retains its virulence during an entire month in summer weather.

M. Pasteur observed that the virus of hydrophobia is not always present in the cerebro-spinal fluid. In the preceding communication to the one from which we are now quoting, he stated that inoculation by venous or subcutaneous injection produces principally the paralytic form of rabies; the animal, especially if it be a dog, is not furious, and never barks in that particular way known as the barking of a mad dog, neither has it a propensity to bite. Furthermore, furious madness is easily provoked by venous and hypodermic injections, if very small doses of the virus are used.

The Nature of Rabies Confirmed by the Quantity of Inoculation-Fluid Injected.—Portions of rabid medulla oblongata were mixed with sterilised broth, and injected into the veins of three dogs, in the following quantities. In the first dog inoculated, half a cubic centimetre of this mixture was injected into its veins; in the second 1-100th of this quantity, or, in other words, 5 milligrammes; in the third, 1-200th, or 2.5 milligrammes.

⁸ Nouvelle communication sur la rage avec la collaboration de MM. Chamberland et Roux.—*Comptes Rendus de l'Académie des Sciences*, 25 Février, 1884, p. 457. (M. Thuillier, M. Pasteur's third assistant, died at Alexandria in September, 1883.)

⁶ Nouveaux faits pour servir à la connaissance de la Rage.—*Comptes Rendus de l'Académie des Sciences*, tome xcv, 1882, p. 1187.

⁷ We do not describe the precautions necessary to be taken in order to avoid the accidents that can attend venous injections; we imagine our readers are acquainted with them; should they wish to refresh their memory, they will find them minutely described in Cyon's, Burdon Sanderson's, Foster's, and Bruntton's *Laboratory Handbooks*. Neither do we consider it necessary to describe trephining, as this operation is described in all manuals of surgery.

On the tenth day, the dog first inoculated began to lose his appetite; he became paralytic on the eighteenth day, and died on the twentieth; he had neither barked nor bitten. The second dog had a suspicious appearance on the thirty-eighth day; on the thirty-ninth, he barked the characteristic bark of a mad dog; the next morning, he was found dead. The third dog had never shown any symptoms of rabies.

In another experiment, made on another series of three dogs, the inoculations were made as follows. One cubic centimetre of rabid medulla was mixed with sterilised broth, and injected into the veins of a dog; one-twentieth of this quantity (or fifty milligrammes) was injected into a second dog; and, in a third, twenty-five milligrammes.

The first dog presented an incubation period of seven days; the second, one of twenty days; the third, one of twenty-five days. The first two dogs inoculated died from paralytic rabies; the third, from furious madness, with barking and biting propensities.

Furious Madness produced by a Smaller Quantity of Virus than Paralytic Madness.—Thus, these experiments indicate that furious, barking, biting hydrophobia, or the paralytic form, can be produced according to the will of the experimenter. The first form, the most terrible as well as the most dangerous, is produced by a smaller quantity of virus absorbed in the animal organism than that producing the paralytic form. This fact probably explains the more frequent occurrence of furious madness among dogs, which results from the bite of a mad dog, when only a small quantity of virus is lodged in the wound by the dog's teeth.

M. Pasteur has also established another important fact: inoculating a quantity of virus insufficient to produce the symptoms of madness (*phénomènes rabiques*) does not confer immunity from rabies; subsequent inoculation is followed by the appearance of the disease.

Virus does not Travel along the Peripheral Nerves to the Central Nervous System.—The certainty that attends venous injections of virus completely destroys the hypothesis maintained by some, that rabid virus travels from the peripheral nervous system to the central, observing the course of the nerves. This hypothesis entirely disregards anatomical and physiological data; a virus cannot, evidently, observe the same behaviour as a sensation, and travel like one. It is thus more logical to suppose that it is absorbed by the circulatory system.

M. Pasteur, in order to demonstrate that this hypothesis is erroneous, inoculated a rabbit in the auricular vein. He anticipated the possible objection that the virus, after having been carried through the entire circulatory system, returned to the wound, and was there absorbed by the nerves, by severing the ear with the thermo-cautery just below the inoculation-point; nevertheless, the animal contracted hydrophobia; therefore, the supposition that the virus travels along the nerves is negatived.

Initial Period of Madness disappears; the Symptoms reappear after a long Interval, and result in Death.—In this same communication, M. Pasteur says that he has again observed, in dogs and rabbits, instances in which the first symptoms of madness appear, disappear, and, after a long interval, return, and result in death. This rarely happens with dogs and rabbits, but among fowls these phenomena are more frequent. Rabies in fowls has special characteristics; the animal is sleepy, loses appetite, becomes paralytic, peripheral anaemia is manifested by a discoloration of its comb, and it dies without having presented any violent symptoms.

When the virus has been passed through a long series of animals of the same species, it acquires a sort of certainty in the periodicity of the symptoms provoked, and in the influence it exercises over a given species; this influence varies, according to each species. The virulence of virus varies according to the animals in which it has been cultivated, even though the different cultivations have a common origin. As an example of this, M. Pasteur said that he had in his possession a virus which communicated hydrophobia to rabbits in seven or eight days; the length of the incubation-period could be calculated, almost exactly, within a few hours' difference; he had also another virus, which communicated hydrophobia to guinea-pigs in five or six days, and always with the same certainty. But this certainty is only obtained after the virus has been passed through a considerable number of animals; during the early passages, the incubation-period is subject to considerable varieties.

M. Pasteur believes that the virulence of the virus is in inverse proportion to the incubation-period. A virus communicating hydrophobia in twenty days, is not half so virulent as a virus that gives it in ten. This rule must not be considered absolute. If the proportions of virus inoculated be much less or much greater at one time than at another, the several results will vary. In young animals, the period of incubation is shorter than among adults.

Dogs rendered Refractory to Madness by Inoculation.—M. Pasteur

again stated, in this communication, that the dogs, which he had described in his preceding communication, as refractory to madness, he did not believe to be so by nature; but that he believed he had arrived at a method by which he rendered dogs exempt from the influence of the virus of hydrophobia. These experiments required to be repeated several times before arriving at positive results. In case there should happen a delay in the appearance of the phenomena of rabies, M. Pasteur abstained from making positive statements, and would only say that he possessed twenty-three dogs, all refractory to madness; and rendered so by inoculations of virus, varying in strength. This communication terminates what we, in this report, have termed *Researches on the Transmissibility of Hydrophobia*.

Summary of Facts.—M. Pasteur's experiments, up to this stage, may be summarised as follows. 1. Not only saliva and salivary glands of animals seized with rabies are virulent, or, in other words, capable of transmitting rabies, but also the peripheral and central nervous systems. 2. This virulence is maintained during several days by observing a temperature varying from 0° to 12° Cent. (from 32° to 53.6° Fahr.), and avoiding decomposition. 3. There is more certainty attached to inoculation for rabies with nervous substance than with saliva or salivary glands, because rabid virus can be obtained from the nervous system, free from foreign micro-organisms. 4. Inoculating the cerebral surface, after trephining, also inoculating by venous injection, is attended by more certain results than introducing the virus in the subcutaneous areolar tissue. 5. The form of rabies developed by inoculating the cerebral surface is generally that of furious madness (*rage furieuse*). Furious madness, paralytic madness, or dumb madness, can be produced by venous injections of smaller or larger quantities of virus. 6. An animal is not rendered exempt from hydrophobia after inoculation with too small a quantity of virus to produce the disease; inoculation with a stronger dose will produce the malady. Rabid virus obtained by successive inoculations through a series of generations, in the same animal species, is attended with great certainty in the regularity and nature of its effects, always providing that the concomitant conditions are identical; the incubation-period becomes considerably shortened, and the virus reaches its maximum of virulence.

[To be continued.]

HUDDESFIELD.—Dr. J. S. Cameron suggests the important inquiry whether we are not apt to pay too great attention to the group of "zymotic diseases." He admits that undoubtedly this is the group of most preventable diseases; but he observes that, even of these, the diseases that have been most fatal in Huddersfield during the past few years have been the most preventable of all—namely, measles and whooping-cough. "In both cases, the deaths are due almost entirely to affections of the lungs, supervening upon, or perhaps more strictly part of, the original disease, but in a large number of cases curable. Unfortunately, however, mothers are ignorant of the dangers of the disease, and equally ignorant as to the means of treatment." Dr. Cameron recommends the issue of a handbill on the management of young infants. During 1884, the diseases of child-life, again, were exceptionally prevalent. Whooping-cough, which was very fatal towards the close of 1883, continued to prevail during the first half of the following year, and caused one-sixteenth of all the deaths in the borough during that period. In the third quarter of the year, diarrhoea played the part taken during the two earlier quarters by whooping-cough, and produced 9.5 per cent. of the mortality from all causes. Dr. Cameron deals at some length with the relations of meteorological conditions to the prevalence of autumnal diarrhoea, and he seems to incline to the view that a high temperature and an absence of rain are two of the most important conditions affecting our death-rate from this disease. He attaches much importance to the persistent and regular flushing, in a dry season, by means of hose-pipes, of the drains in the yards and alleys; and he considers that, by thus artificially imitating one of the functions of the rain, the fatality from zymotic diarrhoea was very greatly limited in Huddersfield in the dry season of 1884. "I feel pretty confident," he says, "that, could we afford the water and the labour, we might by such constant flushings diminish the infantile mortality from autumnal diarrhoea in the thickly peopled parts of the town by at least three-fourths." The rate of mortality from all causes was, during 1883, 21.38; and during 1884, 19.54 per 1,000. The zymotic death-rate was, in 1883, 1.68 per 1,000; and in 1884, 1.77. No death from small-pox has occurred in the borough since 1880.

MEDICAL MAGISTRATES.—Dr. F. Bazard has been appointed a magistrate for the borough of Northampton. Mr. R. W. Watkins, of Towcester, has been appointed a justice of the peace for Northamptonshire.