A CONTRIBUTION TO THE EPIDEMIOLOGY OF POLIOMYELITIS.*

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As compared with the period prior to 1909, when the first experimental transmission of epidemic poliomyelitis to the lower animals was accomplished, our present knowledge of the pathology of the disease may be said to be comprehensive.[1] We now possess information in many ways accurate and full regarding the causative microörganism, its portal of entry into and paths of exit from the body, the period of its persistence in the tissues, the places of its location among the organs, the manner in which its presence brings about the characteristic lesions and symptoms of the infection, and certain important immunity reactions which it displays. The data upon which this knowledge is based is being extended by experiment; but up to the present the experimental studies have not yielded results that illuminate particularly the epidemiology of the affection. The observations recorded in this paper are believed to bear upon that aspect of the disease.

PATHOGENICITY OF THE VIRUS.

Hitherto by the virus of poliomyelitis there has been understood an emulsion or filtrate prepared from infected tissues, originally of human origin, capable of transmitting poliomyelitis to monkeys. As long as the virus had not been cultivated artificially or rendered visible no other criteria than successful inoculation served to identify it. In spite of the cultivation of the organism outside the body the above definition holds in practice, for the reason that cul-

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[1] Landsteiner, K., and Popper, E., *Ztschr. f. Immunitätsforsch., Orig.*, 1909, ii, 377. Flexner, S., and Lewis, P. A., *Jour. Am. Med. Assn.*, 1909, liii, 1639.

tivation is still difficult and in inexperienced hands uncertain, and because also less account can be taken of the artificially cultivated virus in considering pathogenicity as its virulence is readily lost.[2]

Somewhat conflicting results have been recorded as to the power of infected human tissues (usually parts of the central nervous organs) to convey paralysis to monkeys. The number of successful implantations has varied from one half or less of the specimens tested[3] to all those inoculated.[4] No conclusion regarding the virulence of the original materials can be drawn from these figures which often are based upon a few tests in which wide differences in method play a part. One exception only to this generalization may have to be made. Of all countries visited recently by epidemic poliomyelitis Sweden seems to have suffered most severely. Elsewhere than in Sweden it has been found difficult to communicate experimental poliomyelitis to monkeys with filtered washings of the nasopharyngeal mucous membranes from acute cases.[5] The deduction seems obvious, therefore, that the Swedish virus, as it exists in nature, is more active for monkeys than the strains occurring in other countries. And yet the conclusion cannot be accepted unreservedly since even small differences in the methods employed for preparing the washings may account for the discrepancy. Thus far in Sweden the virus, which is believed to be carried by healthy persons, has not been transmitted to monkeys so as to set up the typical experimental paralysis, in spite of many trials, while it has been inoculated successfully in the United States.[6] There is nothing inherently improbable in the supposition that the virus of immediate human origin in one place should be more active because better adapted for monkeys than elsewhere at the same period. But in becoming adapted to the monkey the virus undergoes profound modifications; therefore caution is needed before the view is

[2] Flexner, S., and Noguchi, H., *Jour. Exper. Med.*, 1913, xviii, 461.

[3] Landsteiner, K., in Kolle-Wassermann, Handbuch der pathogenen Mikroorganismen, 2d edition, Jena, 1913, viii, 427.

[4] Flexner, S., and Clark, P. F., *Jour. Am. Med. Assn.*, 1911, lvii, 1685.

[5] Kling, C., Wernstedt, W., and Petterson, A., *Ztschr. f. Immunitätsforsch., Orig.*, 1911–12, xii, 316, 657; 1912, xiv, 303.

[6] Flexner, S., Clark, P. F., and Fraser, F. R., *Jour. Am. Med. Assn.*, 1913, lx, 201.

adopted that such already modified strains exist in nature. And yet it appears that in Sweden and probably in still other countries the virus fluctuates considerably in its effects upon human beings, as indicated by the varying prevalence, severity, and mortality of the epidemics of poliomyelitis.

By successive passages of human strains of the virus through monkeys a high degree of virulence may be attained for this species. How great the changes are that take place can be inferred only, since in the inoculation of filtered extracts of the nervous and other organs we cannot actually measure the number of micro-organisms introduced. But where the original materials may prove infective only when in a state of an emulsion of which several cubic centimeters may be required, ultimately 0.1 to 0.001 of a cubic centimeter of Berkefeld filtrate may suffice to cause paralysis. The rise in virulence thus indicated is not determined merely by the smaller effective dose but also by the circumstance that while at the beginning of the adaptation the proportion of monkeys developing paralysis is smaller and the number of recoveries after paralysis larger than at a later period, once the adaptation has been accomplished all the animals inoculated tend to become paralyzed and to succumb to the disease.[7]

After it had acquired a state of high virulence the virus retained maximal activity for monkeys over a long period of time. But whether this maximum becomes a fixed quality was not known. This point is one of fundamental importance as regards, among other things, the question of the causes responsible for the rise and fall of epidemic waves of the disease in nature. We have carried a particular strain of the poliomyelitic virus (M A) through a series of monkeys beginning in the autumn of 1909[8] and ending in the autumn of 1913. At the outset the virus conformed in activity to the description given: at first infection was irregular and recovery after paralysis not uncommon. After several passages adaptation of the virus was secured and infection followed more regularly and then constantly upon inoculation for which small doses of the filtrate

[7] Flexner, S., Huxley Lecture, *Lancet*, 1912, ii, 1271, and *Science*, 1912, xxxvi, 685.

[8] Flexner, S., and Lewis, P. A., *Jour. Am. Med. Assn.*, 1909, liii, 1639.

sufficed. Recovery at first rarely and later never occurred.[9] For precautionary reasons the inoculations were usually made in duplicate or multiple series. At the same time portions of the spinal cord and brain from infected animals were regularly set aside in 50 per cent. glycerin in the refrigerator, so that if an inoculated monkey succumbed to an intercurrent disease,—dysentery, pneumonia, or tuberculosis,—as occasionally happens, the strain would not be lost.

The M A strain retained maximal virulence during a period of about three years, from the winter of 1909 to the winter of 1912, when a change became apparent. The power to infect began to fluctuate, while among the paralyzed animals recovery began to take place. Once begun the infective power diminished more and more although not regularly. Even at this time several successive inoculations might be succeeded by infection and death. But gradually and somewhat irregularly the virulence deteriorated more and more until the state of the virus resembled in its infecting power that of the original human material from which it was derived. At first the filtrates and only later the emulsions acted irregularly and unreliably. The infective power was not, however, completely abolished, for paralysis could still be induced in occasional animals by means of large doses of filtrates or emulsions, but by no means certainly, irrespective of the size of the dose administered.

During the long period of maximal infectivity the virus seemed to have acquired established or fixed virulence. It now became obvious that fixation of pathogenic effect in the sense in which this term is applied to the vaccine or rabic virus did not occur. Moreover, the return of the maximal virus to a state similar to that of the original human source is apparent only at the present time since thus far no tendency to rise again has been noted in the deteriorated strain. This is true of all the collateral specimens of the M A strain carried through the several series, the deterioration not being confined to a single specimen of the strain. Whether after a resting period a second enhancement can be accomplished by a new series of passages cannot of course be predicted. But

[9] Flexner, S., and Lewis, P. A., *Jour. Exper. Med.*, 1910, xii, 227.

that the deterioration began at a time considerably previous to its discovery is shown unmistakably by our attempts to secure a vigorous strain by returning to glycerinated specimens set aside during the still active period of the virus. After a varying number of passages fluctuation again set in and a similar irregularity in pathogenic effects appeared necessitating abandonment of the series. Long glycerination itself preserves the virus in the state of activity possessed at the time it was set aside. By resorting to a specimen (K) originally also adapted in 1909[10] and passed through an occasional monkey at long intervals, a highly virulent strain was again secured for experimental purposes. If a comparison of the two strains M A and K, adapted at about the same time, is permissible the conclusion to be drawn from their relative present states of activity is to the effect that frequent and long continued passage through monkeys finally brings about a depression of virulence, while preservation in a state of latency for a period equally great exerts no depressing action.

When the M A virus became highly potent the inoculation series were continued in two general ways. What was termed the regular series was designed merely to secure a constant passage virus. For this purpose Macacus monkeys were systematically inoculated, usually intracerebrally, with 1 or 2 c.c. of a Berkefeld filtrate prepared from a 5 per cent. suspension of the fresh medulla and spinal cord of a recently paralyzed animal. The disease thus caused ran the typical course of experimental poliomyelitis. The rule was to etherize the paralyzed animals within this series. Coincidentally with the regular series other series of inoculations were conducted in the course of the study of special aspects of the problem of poliomyelitis. The filtrate for this second group of experiments was usually given in smaller doses. The active virus caused paralysis in quantities of filtrate varying from 0.1 to 0.01 c.c. or less; hence the doses employed ranged from 0.1 to 0.5 c.c., depending on the purpose of the injection. The incubation period of the series receiving the smaller doses was somewhat greater than that of the regular series, but the final result was the same. In some instances intrasciatic[11] and combined intrasciatic and intraperitoneal injections[12] were employed. An active virus causes infection by intraneural injection almost as constantly as by intracerebral inoculation. Falling off in virulence is expressed in (a) failure to cause paralysis, (b) mild infection followed by recovery, and (c) by atypical symptoms and clinical course, followed by either recovery or delayed paralysis and death. The typical course is characteristic. Following an

[10] Flexner, S., and Lewis, P. A., *Jour. Am. Med. Assn.*, 1909, liii, 1913.

[11] Flexner, S., and Lewis, P. A., *Jour. Am. Med. Assn.*, 1909, liii, 1913.

[12] Leiner, C., and von Wiesner, R., *Wien. klin. Wchnschr.*, 1909, xxii, 1698; 1910, xxiii, 91.

incubation period of from five to seven days there succeed excitement, general tremor, weakness, and then paralysis of muscles affecting first the extremities, next the trunk, and lastly the respiration. Exceptionally the muscles of respiration are affected early and convulsions and death occur before paralysis of the extremities is noted. When the course is atypical either reinoculation or microscopical examination of the spinal cord may be required to establish the infection.

The fact of the loss of power of the M A strain of the virus is brought out in table I. Other instances showing this loss have not been included. These were examples of border-line inoculation with smaller doses of virus; but they serve to exclude still further any effect of resistance as such as being a determining factor among the monkeys.

TABLE I.

Illustrative Examples of Weakened Virus.

Date of inoculation.	Materials inoculated.	Clinical result.	Remarks.
Aug. 24, 1912	Emulsion of spinal cord	Typical paralysis	Death on 6th day.
Aug. 26, 1912	Filtrate of preceding emulsion	No effect	
Sept. 1, 1912	Emulsion	Atypical paralysis	Death on 8th day.
Sept. 23, 1912	Filtrate	Atypical paralysis	Recovery; immune.
Oct. 7, 1912	Emulsion	Atypical paralysis	Death on 11th day.
Dec. 9, 1912	Filtrate intrasciatic	No effect	Two animals used.
Dec. 18, 1912	Filtrate	Atypical paralysis	Recovered.
Jan. 21, 1913	Emulsion intrasciatic	No effect	Two animals used.
Feb. 8, 1913	Filtrate 0.1, 0.2, 0.3 c.c. intracerebral	Paralysis not strictly typical	Three animals used.
Apr. 17, 1913	Filtrate	Typical paralysis or no effect	Of 4 animals 2 escaped paralysis.
July 2, 1913	Emulsion	Slight ataxia; no progress	Recovery.
July 9, 1913	Emulsion	Atypical paralysis	Recovery.
July 10, 1913	Emulsion	Partial paralysis	Recovery with residual paralysis.
July 19, 1913	Filtrate	Typical paralysis; prostrate	Recovery with residual paralysis.
July 21, 1913	Filtrate	Typical paralysis; prostrate	Recovery with residual paralysis.
Aug. 26, 1913	Filtrate	Ataxic; partial paralysis	Recovery with residual paralysis.
Sept. 9, 1913	Emulsion	No paralysis	Four other animals inoculated with filtrate from this emulsion without effect.
Sept. 10, 1913	Filtrate	Partial paralysis; rapid partial recovery	Recovery with residual paralysis.
Oct. 23, 1913	Emulsion	Partial paralysis	Recovery with residual paralysis.

BEARING ON EPIDEMIOLOGY.

We do not possess a generally acceptable theory to account for the epidemic waves of disease. What is required is an adequate explanation of the initial rise, persistence, and the final fall of the wave as represented by the varying number of the affected. That mere presence of the microbic causes of disease does not suffice to produce epidemics has long been known. It is just the discrepancy between the occurrence of the microbic causes in sporadic cases of potentially epidemic diseases and the absence of true epidemics that has led to the formulation of the hypothesis of concomitant causes of von Pettenkofer and of Nägeli. While the one supposes a necessary ripening of the microbic agent in the earth as a prerequisite, the other invokes the coöperation of a second although unknown but subsidiary microörganism.[13] The subject has not been rendered essentially more comprehensible by the discovery of the healthy and chronic carriers of infectious microörganisms, or by the more ready detection of so called abortive cases of infection. Indeed, these discoveries only add to the perplexity since they prove that potentially infective microörganisms capable of starting epidemics are more frequently present in our surroundings than has hitherto been supposed.

Perhaps a factor which has not up to the present been sufficiently considered is that of variations among the microörganisms themselves that may be directly responsible for the production of epidemics. That microörganisms, along with all living things, tend to vary in their biological properties, has long been known, but it is only recently that these variations have been recognized as constituting mutations.[14] The variations thus far studied relate chiefly to (a) colony formation and fermentation effects, and (b) serum and drug reactions of fastness, while variations affecting the quality of virulence have been little considered in this respect.

The number of examples known in which mutation in the quality of virulence has occurred is already considerable. Thus per-

[13] Gotschlich, E., in Rubner, M., von Gruber, M., and Ficker, M., Handbuch der Hygiene, Leipzig, 1913, iii, pt. 1, 206.

[14] Müller, R., *Ztschr. f. indukt. Abstammungs- u. Vererbungslehre,* 1912, viii, 305.

manent alterations of this character have been produced in vaccine virus and the fixed rabic virus. Not a few pathogenic bacteria may be changed profoundly in virulence by animal passages, oftenest with the effect of intensification but not infrequently with the contrary effect. Even in artificial cultures enhancement as well as diminution of virulence has been noted and particularly in the case of plague bacilli and meningococci.[15] The modifications in virulence appear at one time quickly or even suddenly and at another develop gradually. There is no doubt that under many natural conditions the passage of infectious microörganisms rapidly from animal to animal or person to person leads to great enhancement of virulence as, to mention one example only, in the pneumonic form of the plague. There exists experimental foundation for the belief that during the rise the microbic causes are more virulent than during the fall of epidemics. The questions to be answered are whether it is this variation, perhaps mutation, among the potential microbic causes of epidemics that is responsible for the waves mentioned, and if so what the nature of the agency is that brings about the mutational changes. It is because the data bearing on the cycle of pathogenicity of the M A virus seem to offer answers to these important questions that they have been set down here in detail.

At the outset the virus of human poliomyelitis possesses relatively weak pathogenic action for monkeys. By means of a few passages the infective power rises and soon a maximum is reached which endures for several years. Ultimately, the infective power falls off and soon becomes greatly diminished, so that finally the power is no greater than at the outset. This succession of phenomena dependent on changes in virulence finds a counterpart in the phenomena noted during the rise, persistence, and then fall of numbers of cases, among man and animals, that constitute epidemics of disease. Moreover, the fluctuations in virulence upon which the phenomena in the first instance depend are the product, as far as can now be determined, of causes acting upon the M A virus from within, that is, the result is due to internal rather than to external effects. In the

[15] Gotschlich, E., in Kolle-Wassermann, Handbuch der pathogenen Mikroorganismen, 2d edition, Jena, 1911, i, 166, 167.

course of the long propagation of the M A virus through monkeys the species was constant and the methods employed for inoculation remained uniform. These causes, whatever their nature, operate to produce a cycle of activity indicated by rise, fixation and fall in infecting power. And this is the cycle, apparently, that many epidemics pass through in the course of their appearance and disappearance.

There exists another fact inherent in all epidemics which lies, however, outside the present consideration, namely, the varying number of susceptible persons who fall victims to the prevailing disease. Our observations bear upon the conditions which make epidemics possible rather than those which determine their actual extent.

In the light of this presentation the part played by sporadic and abortive cases and of the microbe carriers of potentially epidemic diseases becomes more comprehensible. We may consider this class of infected persons or animals as carrying specific microörganisms lacking high virulence for their respective kind. And we may begin to see how the conversion, through favoring causes, of microörganisms of low into others of high virulence, may be the signal for the appearance of epidemics, not necessarily confined to one place but, possibly, arising almost simultaneously in separated and even remote places when the conditions are similar; just as, on the other hand, the immediate transportation of already elevated microörganisms from a place in which an epidemic is already prevailing to new places may start similar severe outbreaks there.

SUMMARY.

A strain of the poliomyelitic virus was propagated in monkeys for four years, during which time it displayed three distinct phases of virulence. The several phases covered different periods of time. At the outset the virulence was low, but by animal passages it quickly rose to a maximum; this maximum was maintained for about three years, when, without known changes in the external conditions, a diminution set in and increased until at the expiration of a few months the degree of virulence about equalled that present

at the beginning of the passages in monkeys. The cycle of changes in virulence is correlated with the wave-like fluctuation in epidemics of disease which also consist of a rise, temporary maximum, and fall in the number of cases prevailing. And an explanation of epidemics of disease is inferred in variations or mutations among the microörganismal causes of disease affecting chiefly the quality of their virulence.