Investigators are divided into two well defined groups with reference to the interpretation of the relation of certain streptococci cultivated from poliomyelitic nervous tissues to epidemic poliomyelitis. One group affirms that the streptococci bear a causal relation to poliomyelitis and are even related biologically to the globoid bodies of Flexner and Noguchi, while another group denies that they possess any essential etiologic importance and views them merely as secondary invaders.

The question at issue is an important one in every way, because upon its true answer will depend the prophylactic measures adopted to prevent epidemics of poliomyelitis and the direction which effort will take in perfecting an efficient agent for specific therapy.

Until recently the effort made to treat cases of poliomyelitis specifically has been with the blood serum of convalescent and recovered cases of the disease. This procedure is based upon several kinds of conclusive experimental data. However, too few observations are available to decide whether the method gives unmistakable thera-

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Therapeutic results in human cases of poliomyelitis, although the indications are favorable. The employment of the serum derived from recovered cases of poliomyelitis followed not only on account of the detection of its neutralizing property in vitro for the poliomyelitic virus, but also because of the failure to induce antibody formation in a variety of domestic animals, including the horse, by the injection of the nervous tissues of monkey or man carrying the virus. Incidentally, it may be stated that only imperfect success in developing antibodies in rabbits and monkeys has attended the repeated injection of cultures of the globoid bodies.

A far greater measure of success has been claimed for the streptococci in producing antibodies for the virus of poliomyelitis. Rosenow and Nuzum and Willy assert that animals immunized with the streptococci cultivated from poliomyelitic cases exhibit various antagonisms to the virus. Monkeys inoculated with streptococci are said to be protected from subsequent infection with the poliomyelitic virus; the blood of the protected monkeys is stated to be neutralizing in vitro for the virus; and finally, horses immunized with the virus are said to yield a serum which possesses neutralizing, protective, and therapeutic properties, even when applied to man.

It is this last statement which calls for painstaking control. It is obvious that to obtain a decision from the treatment of human cases of poliomyelitis would require a large and varied series of observations extending over a long period of time and embracing epidemics of considerable magnitude and various degrees of severity. Moreover, the observations would have to be carefully controlled by comparison with an equal number of cases, occurring simultaneously, untreated with the antiserum, under approximately identical conditions and equally accurately studied. To secure these data might require several years, as has been the case notably with the serum treatment of diphtheria and epidemic meningitis. The question arises,
therefore, whether a method is not available by means of which a
probable decision may be reached more expeditiously and with
greater certainty. It is because of our belief that a decisive experi-
mental method is at hand that the series of experiments to be reported
were performed.

EXPERIMENTAL.

The injection intracerebrally into monkeys of minute quantities of
an active virus of poliomyelitis is followed by paralysis and, as a rule,
by death of the animal. The injection of far greater quantities of the
same virus into the blood stream produces no symptoms. If, how-
ever, as Flexner and Amoss\textsuperscript{10,11} have shown, the meninges and choroid
plexus are chemically inflamed by a simultaneous or previous in-
jection of sterile horse serum, monkey serum, or even isotonic saline
solution, the virus is enabled to pass from the blood into the nervous
tissues and thus to induce the characteristic changes which lead to
paralysis and even to death. The same authors found only one sub-
stance which, when injected intraspinally, prevented the localization
of the virus in the nervous organs after intravenous injection, and
that is the serum derived from monkeys which have survived a
poliomyelitic infection. Moreover, this serum is capable of setting
aside the effects of the chemical inflammation incited by horse serum
or other foreign fluids injected intraspinally. In other words, when
the immune convalescent serum is introduced into the meninges in
animals previously injected intraspinally with horse serum or other
fluids mentioned, followed by an intravenous injection of the virus,
no paralysis or other evidence of infection results. This experiment
gives such decisive and unequivocal results that it seems particularly
adapted to determine the therapeutic value of a serum or other product
reputed to be effective in the treatment of poliomyelitis in man.

The experiments to be described were carried out in the following
manner. An active, fresh poliomyelitic virus was obtained in the
usual manner by inoculating a monkey intracerebrally with a sus-
pension of glycerolated virus. On the 1st day of complete prostra-
tion, the animal was etherized, and the brain and spinal cord were

\textsuperscript{10} Flexner, S., and Amoss, H. L., \textit{J. Exp. Med.}, 1914, xx, 249.
\textsuperscript{11} Flexner, S., and Amoss, H. L., \textit{J. Exp. Med.}, 1917, xxv, 525.
aseptically removed. With the spinal cord and medulla, a 5 per cent suspension in isotonic salt solution was prepared, shaken, and centrifuged, and the clear supernatant fluid injected intravenously into Macacus rhesus monkeys. Two separate sets of experiments were performed. Control animals and animals treated with Rosenow's serum, with normal horse serum, and with convalescent monkey serum, in the same manner, were tested simultaneously. The protocols follow. The outcome is as sharp and hence as decisive as the experimental results could make it.

**Experiment 1.—**Monkey A, control. Nov. 13, 1917. Injected intravenously 50 cc. of the virus prepared as described. The animal remained well.

Monkey B, normal horse serum control. Nov. 12, 1917, 5.10 p.m. Injected intraspinally 2.5 cc. of normal horse serum. Nov. 13, 11.15 a.m. Injected intravenously 50 cc. of virus. 12 noon. Injected intraspinally 2.5 cc. of normal horse serum. Nov. 14. Injected intraspinally 2.5 cc. of normal horse serum. Nov. 15. Repeated intraspinal injection of normal horse serum. Nov. 16. Repeated intraspinal injection of normal horse serum. Protects left leg; ataxic. Nov. 17, a.m. Left arm, right deltoid, and both legs paralyzed; head tremor; ptosis of the left eyelid; almost prostrate. 4.30 p.m. Died.

**Autopsy.**—Macroscopic and microscopic lesions of poliomyelitis. No visible changes in viscera.


**Autopsy.**—The spinal cord and brain were edematous and the gray matter was congested. Microscopic examination of the central nervous system showed marked perivascular infiltration and some neurophagocytosis in the gray matter of the medulla and cervical enlargement characteristic of poliomyelitis. Perivascular infiltration (Figs. 2 and 3), congestion, neurophagocytosis, and meningeal infiltration in lumbar enlargement. Focal infiltration of lymphocytes, cell degeneration, and neurophagocytosis in the posterior root ganglia.

Monkey D, serum of recovered monkeys. Nov. 12, 1917, 4.55 p.m. Intraspinal injection of 3 cc. of mixed serums from several rhesus monkeys which had recovered from experimental poliomyelitis and subsequently received subcutaneous injections of the virus contained in the spinal cord and medulla (reinforced
immune). Nov. 13, 11 a.m. Intravenous injection of 50 cc. of virus suspen-
sion followed by intraspinal injection of 3 cc. of immune serum. Nov. 14, 15,
18, 19, and 20. Intraspinal injections of 3 cc. of immune serum. The clinical
course of this animal was in striking contrast with the preceding. At no time
were any symptoms present; the animal continued apparently normal through-
out the treatment and is well at the present time (Jan. 1, 1918).

This experiment was repeated with precisely the same results. The protocols follow. In the second experiment, the normal horse
serum control was omitted.

Experiment 2.—Monkey E, control. Nov. 26, 1917. Intravenous injection of
50 cc. of the virus. No symptoms appeared, and the animal has remained normal
up to the present time (Jan. 1, 1918).

Monkey F, Rosenow's serum. Nov. 26, 1917, 6 p.m. Intraspinal injection of
2 cc. of activated Rosenow's antipoliomyelitic horse serum. Nov. 27, 11.35 a.m.
Intravenous injection of 50 cc. of virus, followed immediately by the intraspinal
injection of 2.5 cc. of Rosenow's serum. The intraspinal injections were re-
peated on Nov. 28, 29, Dec. 2, and 3. In each instance the activated serum was
injected. Dec. 3. The animal developed a marked tremor of the head, ataxia,
and right facial paralysis; also, the deltoid muscles were weak. Dec. 4. The
monkey died in the early morning.

Autopsy.—Macroscopic lesions of poliomyelitis throughout brain and cord.
Microscopic examination of the central nervous system showed marked congestion
and perivascular infiltration, slight cell degeneration, and neurophagocytosis in
medulla and cervical enlargement (Fig. 4); slight meningeal infiltration in lumbar
enlargement, and focal infiltration of lymphocytes, cell degeneration, and neuro-
phagocytosis in posterior root ganglia (Fig. 5).

Monkey G, immune monkey serum. Nov. 26, 1917, 5.15 p.m. 2 cc. of pooled
immune serum injected intraspinally. Nov. 27, 12.25 p.m. Intravenous injec-
tion of 50 cc. of the virus, followed by the intraspinal injection of 2.5 cc. of pooled
immune serum. The intraspinal injections of the pooled serum were repeated on
Nov. 28, 29, Dec. 2, and 3. At no time were any symptoms detected, and the
animal is normal at this time (Jan. 1, 1918).

DISCUSSION.

The preceding experiments accomplish two purposes directly. First, they test the ability of Rosenow's serum, which was prepared
by injecting a horse with cultures of the streptococci derived from
poliomyelitic nervous organs, to prevent a poliomyelitic infection
arising in the monkey after an intravenous injection of the virus.
This is readily accomplished by means of the immune serum obtained
from convalescent and recovered monkeys. Second, they compare directly under these favorable therapeutic conditions the Rosenow serum with the serum of immune monkeys.

The results of the experiments are unequivocal. They show the Rosenow serum to be devoid of protective power. Moreover, they show that the Rosenow serum acts in the manner of normal horse serum in promoting infection in monkeys from an intravenous injection of the virus, in itself incapable of inducing paralysis.

The immune monkey serum possesses, under the same conditions of administration, perfect protective power, as has been shown previously by Flexner and Amoss.¹¹

A further conclusion may be drawn from the experiments. Rosenow states that the horse serum prepared by him contains demonstrable antibodies for the streptococci employed in its production. It is assumed that these antibodies are identical with the antibodies, demonstrable by neutralization experiments with the virus, contained within human and monkey serum derived from recovered cases of poliomyelitis in man and the monkey. This supposition is rendered untenable by the results of our experiments. The antibodies induced in the horse by immunization with the streptococci have proven incapable of neutralizing the virus of poliomyelitis introduced into the blood of monkeys in its passage to the central nervous system—a neutralization which the immune monkey serum readily effects. The two classes of antibodies or immunity principles, those present in the blood derived from recovered cases of poliomyelitis and those induced in the horse by treatment with streptococci, are therefore to be regarded as distinct.

There is a further corollary to this general deduction. Once it is established that the antibodies yielded by the streptococci differ essentially from those induced by the virus of poliomyelitis, the contention that virus and streptococci are identical becomes untenable. In other words, the experiments reported in this paper tend also to refute the claim that certain streptococci are the microbial cause of epidemic poliomyelitis.

Rosenow and Nuzum and Willy assert that their serums possess striking therapeutic activity in man. Their conclusions are based on the treatment of relatively small numbers of cases of epidemic polio-
myelitis during the past summer and autumn. We have already
drawn attention to the difficulties surrounding a statistical study, of
limited extent, of the questions here involved. Hence we venture
to place, in this instance, the greater weight on decisive animal ex-
periments, and those reported in this paper clearly show that Rose-
now's horse serum injected intraspinally into monkeys is without
specific protective power against the virus of poliomyelitis.

It has, however, been shown by Flexner and Amoss\textsuperscript{12} that normal
horse serum, when injected intraspinally into monkeys, promotes the
passage of poliomyelitic immune bodies from the blood into the
subarachnoid space. Hence it is possible that under certain circum-
stances in which those bodies are already present in the blood in man,
they may be directed into the subarachnoid space through the in-
creased permeability of the meninges induced by the horse serum and
thus affect the course of the infection. The antibodies have been
detected on the 3rd\textsuperscript{13} and 6th\textsuperscript{12} days of illness in man, or, in other
words, early in the course of the disease.

This, however, is a purely hypothetical consideration, in support of
which normal horse serum should prove as effective as antistrepto-
coccus serum. It is questionable whether this roundabout method
of directing the circulating immunity bodies to the central nervous
organs is advisable in practice. As far as present knowledge, based
on definitive experiments, is concerned, it may be said that only im-
mune serum derived from convalescent and recovered cases of polio-
myelitis in man and the monkey have been determined to be pro-
tective against the infectious power of the poliomyelitic virus.

CONCLUSIONS.

Two series of experiments are described in which Rosenow's anti-
poliomyelitic serum, so called, has been compared with the immune
serum derived from monkeys which have convalesced or recovered
from experimental poliomyelitis.

The experiments consisted in introducing an active virus of polio-

\textsuperscript{12} Flexner, S., and Amoss, H. L., \textit{J. Exp. Med.}, 1917, xxv, 499.
\textsuperscript{13} Kling, C. A., and Levaditi, C., \textit{Études sur la poliomyélite aiguë \textit{épidémique},
myelitis into the blood and of injecting the two kinds of serum into the cerebrospinal meninges according to the method of Flexner and Amoss.

Under the conditions of the experiment, the control monkeys (a) receiving the virus intravenously alone do not develop paralysis, while those (b) receiving the virus intravenously and normal horse serum intraspinally develop paralysis. Moreover, the monkeys (c) receiving the virus intravenously and Rosenow’s antipoliomyelitic serum intraspinally develop paralysis in the manner of those receiving normal horse serum intraspinally. The monkeys (d) which received the virus intravenously and the convalescent or immune monkey serum intraspinally alone did not develop paralysis.

The Rosenow serum acts in the manner of normal horse serum; it promotes the passage of the virus of poliomyelitis from the blood into the nervous organs, and it does not protect from infection.

We have found no evidence that Rosenow’s serum under the conditions of the tests is effective therapeutically in monkeys or possesses antibodies of the same nature as those present in the blood of monkeys which have recovered from experimental poliomyelitis.

Since the antibodies in convalescent poliomyelitic serum in man and the monkey are identical, it follows that any antibodies present in the Rosenow horse serum do not conform to those occurring in human convalescent serum.

EXPLANATION OF PLATES.

The illustrations were taken from monkeys treated with Rosenow’s serum.

PLATE 8.

Fig. 1. Monkey C. 11 days after the intravenous injection of virus. Received seven intraspinal injections of Rosenow’s serum. Arms, legs, and back muscles paralyzed; face muscles active.

PLATE 9.

Fig. 2. Monkey C. Cervical enlargement showing perivascular mononuclear cell infiltration in anterior horn. × 165.

Fig. 3. Monkey C. Cervical enlargement showing perivascular mononuclear cell infiltration in posterior horn. × 165.
PLATE 10.

Fig. 4. Monkey F. Cervical enlargement showing anterior horn with degeneration of ganglion cells and neurophagocytosis. × 240.

Fig. 5. Monkey F. Posterior root ganglion with ganglion cell degeneration, neurophagocytosis, and mononuclear cell infiltration. × 240.
(Amoss and Eherson: Rosenow's antipoliomyelitic serum.)
Fig. 2.

Fig. 3.

(Amos and Eberson: Rosennow's antipoliomyelitic serum.)
FIG. 4.

(Amoss and Eberson: Rosenow's antipoliomyelitic serum.)

FIG. 5.