VACCINATION AGAINST POLIO- MYELITIS WITH LIVE VIRUS VACCINES

5. NEUTRALIZING ANTIBODY LEVELS ONE YEAR AFTER VACCINATION

BY

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During the first half of 1956 small laboratory-studied trials of TN type II and SM type I oral attenuated poliomyelitis vaccines (Koprowski, Norton, Jervis, Nelson, Chadwick, Nelsen, and Meyer, 1956) were made in Northern Ireland (Dane, Dick, Connolly, Fisher, and McKeown, 1957; Dick, Dane, Fisher, Connolly, and McKeown, 1957). Neither of these vaccines fulfilled what we considered to be the necessary theoretical requirements of safety (Dick and Dane, 1957), but the results of investigations into the duration of immunity following their administration are reported here because they may be of some value in predicting the sort of immunity which is likely to follow the use of the more attenuated virus vaccines now available.

Materials and Methods.—The materials and methods used were as described previously (Dane *et al.*, 1957). Comparative results for levels of neutralizing antibody in any individual are given only when paired sera were titrated in the same test.

Results

Persistence of Neutralizing Antibody After Vaccination with SM type I Poliovirus

A number of volunteers who were fed SM type I virus in 1956 were bled at frequent intervals after vaccination in order to estimate the highest titre of antibody which they would develop. The serum with the highest titre from each of six adult volunteers was tested again with another serum taken about a year later. The results of these tests are shown in Table I.

TABLE I.—Levels of Type I Neutralizing Antibody in Adults at Various Intervals After Vaccination with S.M. Type I Virus

Serial No.	Age	Sex	Pre-vaccination Titre	Weeks After Vaccination	Titre	Weeks After Vaccination	Titre
193	41	M	<4*	7	22	67	<4
218	39	M	<4	6	22	68	11
353	24	F	<4	4	16	68	4
284	20	M	<4	3	179	99	64
212	20	M	<4	5	359	62	89
235	25	M	<4	9	8	72	256

* Reciprocal of serum antibody titre.

The antibody titre had fallen in five of the individuals, but had risen significantly in the other (No. 235). A number of serum specimens taken from No. 235 over the study period were tested in an attempt to explain this rise in anti-

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body. It was found that between the ninth and the fifteenth month after vaccination he had developed a significant rise in neutralizing antibody to both type I (from 1:4 to 1:256) and type II poliovirus (from <1:4 to 1:96). It appears, therefore, that during this time he had had at least one natural poliovirus infection.

Persistence of Neutralizing Antibody After Vaccination with TN type II Poliovirus

Blood samples from 60 children vaccinated orally with TN type II poliovirus in 1956 were taken approximately one year later. The comparative levels of neutralizing antibody in this specimen and one taken about two months after vaccination were estimated by testing the two sera at the same time. The variation in antibody levels during this period is shown in the Chart.

Individual variation of type II neutralizing antibody titre in 60 children following vaccination with TN type II poliovirus. All the children were without type II antibody at the time of vaccination.

In a proportion of the children a significant antibody rise occurred after the first post-vaccination blood specimen had been taken. It is impossible to prove that the antibody rises in these children were not the result of infections with naturally occurring type II polioviruses, but the most satisfactory explanation is that vaccine virus multiplication in the gut (and therefore the antigenic stimulus) continued in these children beyond the date when the first post-vaccination blood sample was taken (approximately the ninth week after vaccination). We have no laboratory data to support this hypothesis, because the trial was originally planned on the basis of observations by Koprowski *et al.* (1956) that TN type II vaccine virus was excreted in scarcely detectable amounts over a short period, and for this reason collection of faecal specimens was not continued long enough. However, a number of children were excreting faecal virus during the fourth week after vaccination, when specimen collection ceased, and there seems to be no reason why they should not have continued doing so for several months, as was the case in children vaccinated with SM type I virus (Koprowski *et al.*, 1956).

The possibility that some of the rises in type II antibody might have been due to natural infections with heterologous poliovirus types was also considered. Serum specimens taken at the time of vaccination and one year later were tested at a 1:4 dilution against types I and III poliovirus to find out whether any children had developed antibody against

these poliovirus types. Titrations of the paired sera from children found to have developed antibody showed that six natural infections had occurred—three with type I poliovirus and three with type III poliovirus. In the course of another investigation type I poliovirus had in fact been isolated from two of the children who showed serological evidence of a type I infection.

We conclude that heterotypic poliovirus infections could only have contributed to the late rise in type II antibody in these six instances.

After testing the first post-vaccination blood sample (taken after about two months) we reported that only 77% of 124 children had developed neutralizing antibody following vaccination with TN type II virus (Dane *et al.*, 1957). We were able to obtain blood samples one year later from 15 of the 28 children who did not develop antibody. None were found to have developed type II antibody during this interval, thus confirming our original conclusion that the vaccine had failed to "take" in these individuals.

Influence of Heterotypic Antibody on Vaccination with TN Type II Virus

Because of the possibility that previous natural heterotypic poliovirus infections might have influenced the response of children to vaccination with TN type II virus, pre-vaccination samples of blood from 110 children who had been vaccinated with TN type II virus were tested for neutralizing antibody at a 1:4 dilution against types I and III poliovirus. The pre-vaccination antibody patterns found are shown in Table II. Children possessing different pre-vaccination heterotypic antibody patterns showed no significant difference in their response to vaccination.

TABLE II.—Poliomyelitis Neutralizing Antibody Patterns at the Time of Vaccination in 110 Children Fed TN Type II Virus in 1956

No. of Children	Type I Antibody	Type II Antibody	Type III Antibody
63	—	—	—
20	+	—	—
19	—	—	+
8	+	—	+

— = Titre of < 1 in 4. + = Titre of > 1 in 4.

Discussion and Conclusions

The neutralizing antibody levels which we found in children one year after vaccination with TN type II virus were of the same order as those reported in non-immune children who had received two injections of formalinized vaccine a year before (Report, 1957). A number of the TN-vaccinated children were found to have had a marked decline in their neutralizing antibody level over the year, and our previous finding that 23% of children failed to develop antibody after vaccination was confirmed.

One of the advantages claimed for oral attenuated poliomyelitis vaccines is that they will produce a more durable immunity than formalinized vaccines. We can find no evidence that this was true in a proportion of children vaccinated with TN type II virus. Admittedly our yardstick for the measurement of immunity has been circulating neutralizing antibody. Some sort of local gut immunity follows infection with oral poliovirus vaccines, but in the absence of more precise information about this local immunity it would be unwise to assume that it is more durable than circulating antibody.

Further trials of Koprowski's TN type II vaccine have been abandoned because the virus is liable to change its character and become virulent after growth in the human gut. If it can be shown that the recently developed more highly attenuated poliovirus vaccines do not suffer from this drawback, then it would seem desirable to show also that they can produce a reasonably durable immunity in the majority of vaccinated subjects before they are used on a large scale. On the basis of the results reported here we do not think that durability of immunity should be considered as an automatic benefit associated with living vaccines.

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A SMALL-SCALE TRIAL OF TYPE III ATTENUATED LIVING POLIOVIRUS VACCINE

BY

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The purpose of this small-scale trial of a single strain of living attenuated poliovirus vaccine was to obtain information concerning the capacity of the virus to induce infection in children previously immunized with inactivated Salk-type vaccine, to measure the antibody formation after the feeding of the virus, and to compare the virulence for monkeys of excreted virus with that of the strain composing the vaccine. In addition, evidence was sought concerning the spread of virus from vaccinated children to their contacts. The virus vaccine selected for use was the type III attenuated poliovirus from culture of monkey-kidney tissue developed by Dr. Albert B. Sabin and obtained as virus-containing culture fluid from Dr. J. D. Verlinde at Leyden. The virus was the highly purified Leon strain previously subjected to many kidney-tissue-culture passages and plaque-selection, and designated the 12a,b (KP4) virus (Sabin, 1957). The strain pool was freshly titrated by one of us (A. P. G.) upon receipt and found to have a titre of $10^{7.35}$ 50% tissue-culture infective doses per ml.

Control Period of Study

The test was carried out in a long-stay orthopaedic institution where the children were under treatment for lesions such as those due to Perthes's disease or tuberculous arthritis. A control period of study began in September, 1957, when immunization with two doses of British-type Salk vaccine was given to certain boys whose parents volunteered for their inoculation. Nineteen boys aged 6 to 15 years received vaccine intramuscularly after a preliminary withdrawal of 10 ml. of blood. The second dose of vaccine was given one month after the first dose, and all inoculations were completed by December 4, 1957. A second specimen of blood was obtained two weeks after the second

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