

## Further studies with a diphtheria-tetanus-poliomyelitis vaccine

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In 1965 we reported a trial of a combined diphtheria, tetanus and poliomyelitis (dip./tet./pol.) vaccine which had been designed for reinforcing the immunity of children at the time they entered school (Dane *et al.* 1965). One dose was found adequate for this purpose in children who had been previously immunized in infancy. The Ministry of Health (1965) recommended that either a single dose of dip./tet./pol. vaccine or a dose of oral poliovaccine and a dose of diphtheria/tetanus vaccine be used for reinforcing immunity at school entry.

From an administrative point of view there would be obvious advantages if *all* children were given one dose of dip./tet./pol. vaccine at school entry, irrespective of their past immunization history. Those with no history of previous immunization could then be given two further doses of the same vaccine to complete their primary course. The trial described here was designed to investigate the use of dip./tet./pol. vaccine for primary immunization in this way. We did not initially recommend this procedure because of doubts about the efficiency of diphtheria formol toxoid when used in a vaccine containing no adjuvant, such as *Bordetella pertussis* or a mineral carrier, though we considered the other two components would provide adequate immunity.

### MATERIALS AND METHODS

In co-operation with parents and the Belfast County Borough Health Department we immunized thirty-nine school-children aged 5-6 years selected because they were thought not to have had any previous immunization against diphtheria or tetanus. Blood samples were taken at the time of the first injection and again one month after the third.

*The vaccine.* Commercial dip./tet./pol. vaccine prepared by Glaxo Laboratories was used. Each 1 ml. dose contained 56 Lf of diphtheria formol toxoid and 10 Lf of tetanus toxoid. The poliovirus D-antigen content (Beale & Mason, 1962) was: type 1, 75 units; type 2, 3 units; type 3, 6 units. The vaccine was given in three intramuscular doses with intervals of 6 weeks between the first and second and 6 months between the second and third doses.

*Diphtheria and tetanus antitoxin.* Assay was by the methods described in the British Pharmacopoeia (1963 ed., pp. 1107, 1118).

*Poliovirus-neutralizing antibody.* A standard cytopathic test employing approxi-

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mately 100 TCD<sub>50</sub> of virus was used. The titres of British Standard antisera (Perkins & Evans, 1959) were: type 1, 1/1500; type 2, 1/650; type 3, 1/3000.

## RESULTS

*Diphtheria and tetanus*

Five of the thirty-nine children were found to have low levels of either diphtheria or tetanus antitoxin in their pre-immunization blood samples. The antitoxin levels in the blood of the remaining thirty-four children 1 month after the third dose of vaccine are shown in Tables 1 and 2.

Table 1. *Diphtheria antitoxin levels in thirty-four children after three spaced doses of dip./tet./pol. vaccine*

(The children had no previous history of immunization, and all had titres of < 0.001 units per ml. serum before receiving the vaccine.)

No. of children	Antitoxin units per ml. serum		
	0.05-0.10	0.10-1.0	1.0-10.0
	3	13	18

Table 2. *Tetanus antitoxin levels in thirty-four children after three spaced doses of dip./tet./pol. vaccine*

(The children had no previous history of immunization, and all had titres of < 0.02 units per ml. serum before receiving the vaccine.)

No. of children	International antitoxin units per ml. serum					
	0.4-0.8	0.8-1.6	1.6-3.2	3.2-6.4	6.4-12.8	> 12.8
	1	1	9	10	10	3

Table 3. *Poliovirus-neutralizing antibody levels in thirty-nine children who received three spaced doses of dip./tet./pol. vaccine*

(A = Pre-immunization serum; B = post-immunization serum.)

Reciprocal of serum antibody titre	No. of children					
	Type I		Type II		Type III	
	A	B	A	B	A	B
< 10	6	0	1	0	4	0
10-100	4	0	3	0	3	0
> 100-1000	13	1	14	2	13	2
> 1000	16	38	21	37	19	37

*Poliomyelitis*

Though the majority of children in this trial had no previous immunization against diphtheria or tetanus, many had been immunized against poliomyelitis; therefore little information could be obtained about the effectiveness of dip./tet./

pol. vaccine for *primary* immunization against this disease. The neutralizing antibody levels of the children before and after immunization are shown in Table 3.

#### DISCUSSION

The efficiency of diphtheria formol toxoid as an immunizing agent when given with an adjuvant is well established (Report, 1959). However a report to the Medical Research Council (Report, 1962) indicated that highly purified, plain diphtheria formol toxoid is a relatively poor antigen, unsuitable for primary immunization and its use either alone or in combination with tetanus toxoid has not been recommended by the Ministry of Health since 1963 (Ministry of Health, 1963). In the present trial the post-immunization levels of diphtheria antitoxin were found to be adequate even though the dip./tet./pol. vaccine contained no adjuvant. Two factors may account for this apparent difference in efficiency between plain diphtheria formol toxoid and this antigen as a component of dip./tet./pol. vaccine. First, the diphtheria toxoid in combination with two other components as in dip./tet./pol. vaccine is less pure, and, secondly, the dip./tet./pol. vaccine was given in three doses separated by intervals of 6 weeks and 6 months whereas the diphtheria toxoid was given in a more closely spaced schedule of two or three doses 4-6 weeks apart.

The response to the tetanus component of the vaccine was satisfactory. This had been expected because, even without adjuvant, tetanus toxoid is suitable for primary immunization when given in three suitably spaced doses (Boyd, 1959).

We had no reason to believe that present-day potent inactivated poliovirus antigens would be unsuitable for primary immunization and therefore no attempt was made to select children for the trial who were devoid of poliomyelitis antibody. The majority of children had moderate or high levels of neutralizing antibody to at least two of the poliovirus types before immunization with dip./tet./pol. vaccine. As expected, after the third dose of vaccine all children had high levels of antibody to all three poliovirus types.

Though the serological responses of the children in this trial suggest that dip./tet./pol. vaccine is suitable for primary immunization of children at 5 or 6 years of age, this cannot be taken as evidence that it is also suitable for immunizing very young infants. When, for one reason or another, it is thought undesirable to give an infant a pertussis-containing vaccine then dip./tet./pol. vaccine would probably be effective provided that it is given after the age of 6 months, when the inhibitory influences of maternal antibody and immunological immaturity have waned (Evans & Smith, 1963).

It might be argued that dip./tet./pol. vaccine could be improved by the addition of a mineral-carrier adjuvant. Such an addition would result in a better response to the diphtheria toxoid but it might also lead to more local reactions. In its present form the vaccine causes negligible reactions (Dane *et al.* 1965) and we consider that it would be unwise to alter it in a way which might decrease its acceptability.

The administrative convenience of being able to use a single vaccine for protect-

ing young school-children against diphtheria, tetanus and poliomyelitis, whether they have had previous immunization or not, is considerable. There was a 6 months interval between the second and third doses in the present trial but if this was extended to  $10\frac{1}{2}$  months it would be possible to give third doses, at the same time as first doses were being given to next year's intake of children into a school. In this way a school Medical Officer making two visits to a school a year and using a single vaccine could ensure that adequate continuing immunity was provided against diphtheria, tetanus and poliomyelitis.

#### SUMMARY

The efficiency of a diphtheria, tetanus, poliomyelitis vaccine in inducing a serological response after a three-dose primary course of immunization was tested in thirty-nine children aged 5 and 6 years and found to be satisfactory. This vaccine had previously been shown to be suitable for use as a single dose reinforcing vaccine for children of this age who had been immunized in infancy. It is suggested that all children might receive one dose of the vaccine at the time they enter school, and then those who have not been immunized before should receive a further two suitably spaced doses to complete their course of primary immunization.

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