of dilated and confluent alveoli. The alveolar spaces were completely free in some areas; others contained scattered erythrocytes and macrophages and some precipitated albuminous material; while still others were filled with an acute inflammatory exudate composed mostly of polymorphonuclear leucocytes, with erythrocytes, macrophages, and definite fibrin webs.

Discussion

Because of the patient's general condition—his age was 76; he had suffered a recent weight loss of 50 lb. (22.7 kg.); he had moderately severe fibrosis and emphysema of the lungs and generalized arteriosclerosis—and since it was anticipated that operation would last six hours, it was felt that this case was suitable for artificial hibernation with hypothermia. Also, Huguenard (1953) has stated that chronic pulmonary disease does not constitute a contraindication but rather an indication for this procedure. Laboratory findings and an electrokymogram were similar to those reported by Huguenard. The only electrocardiographic changes noted were non-specific T-wave changes in the immediate post-operative period. These were interpreted as probably positional in origin. The electrolyte values remained within normal limits throughout. A transient hyperglycaemia (up to 250 mg. per 100 ml.) occurred at the end of the operation, but the blood sugar returned to a normal level on the first post-operative day. Dundee (1954) questioned whether this could be due to a direct toxic effect of chlorpromazine on the liver.

The microscopic findings would seem to indicate that pulmonary infarction had occurred some time previously as well as recently. It is interesting to speculate on what may have been the cause of these infarcts. A systolic blood pressure of 60 mm. Hg may be a factor. However, Zeavin *et al.* (1954) are of the opinion that this hypotension is physiological in that it is accompanied by a marked reduction in oxygen requirement. Another factor may have been the prolonged collapse of the left lung during surgery. Deterling *et al.* (1955) have shown that during hypothermia there is an increase in the clotting time of the blood, but no increase in bleeding because of the slowed peripheral circulation. From the findings of the above authors it would appear that slowing of the circulation does not predispose to the formation of emboli. Huguenard warns of the "rebound" that takes place during the rewarming period, and urges the use of heparin. In the present case heparin was not used in the rewarming period, as it was considered that bleeding might occur from the site of anastomosis. Nevertheless recovery was uneventful until the sixteenth day after operation, when signs and symptoms of shock appeared. Despite energetic treatment the patient died 36 hours later.

At necropsy pulmonary emboli were found, but it is difficult to be sure what were the factors, if any, predisposing to this complication. Therefore it is suggested that to guard against such an eventuality anticoagulant therapy be used, not only in the rewarming period, but also post-operatively.

Summary

A case is presented in which oesophago-gastrectomy was performed with the use of "artificial hibernation" with hypothermia. This procedure was complicated post-operatively by the development of multiple pulmonary emboli. It is suggested that anticoagulant therapy should be used in the post-operative period as well as in the rewarming period to prevent this complication.

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EFFECT OF ADDED TOXOIDS ON THE ANTIGENICITY OF *H. PERTUSSIS* VACCINES

BY

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It has been firmly established that the addition of *Haemophilus pertussis* vaccine to diphtheria and tetanus toxoids enhances the activities of both antigens (Greenberg and Fleming, 1947). Addition of mineral carriers—for example, aluminium phosphate—to the toxoids for the immunization of children can be replaced by the addition of *H. pertussis* (Ungar, 1952, 1954). The pronounced local reaction after inoculations with antigens containing mineral carrier is thus avoided. Earlier work by Glenney and Waddington (1928), and more recent work by Barr and Llewellyn-Jones (1953), indicated that the qualitative and quantitative composition of the components in a combined prophylactic might influence in animals the degree of immunological response to the different components. Under a particular set of conditions those authors noticed that one component had interfered with the immunity response to the other antigens present in a combined product.

In view of the present general interest in combined prophylactics for the immunization of children we have investigated whether or not the presence of either toxoid, diphtheria or tetanus, or of both, reduced the immunity response of mice to *H. pertussis* vaccines. Combined antigens that had been used for the immunization of children, with compositions based on earlier experiments on animals, were chosen.

Experimental

Two series of prophylactics were prepared by adding toxoids to each of two *H. pertussis* vaccines A and B, prepared on different occasions but otherwise similar. The following prophylactics were used:

- (1) *H. pertussis* vaccine A $20,000 \times 10^6$ cells/ml.
- (2) *H. pertussis* vaccine A plus diphtheria toxoid 25 Lf/ml.
- (3) *H. pertussis* vaccine A plus diphtheria toxoid 25 Lf/ml., plus tetanus toxoid, 2.5 Lf/ml.
- (4) *H. pertussis* vaccine B $20,000 \times 10^6$ cells/ml.
- (5) *H. pertussis* vaccine B plus diphtheria toxoid 25 Lf/ml.
- (6) *H. pertussis* vaccine B plus diphtheria toxoid 25 Lf/ml., plus tetanus toxoid, 2.5 Lf/ml.

The *H. pertussis* vaccines were prepared from freshly isolated strains grown on Bordet-Gengou medium, killed with formalin and preserved in thiomersalate ("merthiolate") (1:7,500). The diphtheria and tetanus components added were plain toxoids, purified by ammonium sulphate precipitation.

The six products were tested three times concurrently for their ability to protect mice against intracerebral challenge by Kendrick's method (Kendrick *et al.*, 1947), the antigenic ratios being assessed by the method of Weiss (1948). The mice used were of both sexes, from the colony of strain A2G mice inbred in these laboratories.

Similar mouse protection tests were carried out on a series of 12 plain *H. pertussis* vaccines, prepared from organisms grown for 48 hours on Bordet-Gengou medium, killed with formalin and standardized to $20,000 \times 10^6$ cells per ml. by opacity measurement. The results of these tests were compared with those on 16 different batches of a diphtheria-pertussis-tetanus prophylactic ("triple antigen" D.P.T.), prepared by the addition of both toxoids to

H. pertussis vaccine suspensions similar to the 12 just mentioned. Several of these tests on the two prophylactics were carried out concurrently, the animals being "challenged" at the same time with the same suspensions of virulent organisms. These are indicated by dates in Tables II and III, where the results are recorded.

Results

The results given in Table I indicate that the antigenic response of mice to the two different *H. pertussis* vaccines investigated was not reduced by the addition of diphtheria toxoid or of diphtheria and tetanus toxoids combined. This is in agreement with the further results given in Tables II and III. Statistically, the batches of *H. pertussis* plain

TABLE I.—Results of Mouse Protection Tests of *H. pertussis* Vaccines A and B With and Without the Addition of Diphtheria and Tetanus Toxoids

Vaccine	Test No.	No. of Mice Surviving 14 Days at Immunization Levels			ED ₅₀	ED ₅₀ Mean of Three Tests
		1,000 × 10 ⁶	200 × 10 ⁶	40 × 10 ⁶		
<i>H. pertussis</i> plain vaccine (A)	1	20/20	8/20	5/20	125 × 10 ⁶	133 × 10 ⁶
	2	19/20	9/20	0/20	220 × 10 ⁶	
	3	20/20	14/20	9/20	55 × 10 ⁶	
<i>H. pertussis</i> plain vaccine (B)	1	16/20	11/20	7/20	110 × 10 ⁶	400 × 10 ⁶
	2	10/20	8/20	2/20	1,000 × 10 ⁶	
	3	18/20	15/20	5/20	90 × 10 ⁶	
Diphtheria/ <i>pertussis</i> (A)	1	18/19	18/20	3/20	60 × 10 ⁶	168 × 10 ⁶
	2	17/20	5/20	3/20	400 × 10 ⁶	
	3	18/20	14/20	9/20	45 × 10 ⁶	
Diphtheria/ <i>pertussis</i> (B)	1	17/20	12/19	9/18	50 × 10 ⁶	253 × 10 ⁶
	2	12/20	6/20	1/20	600 × 10 ⁶	
	3	19/19	14/20	3/20	110 × 10 ⁶	
Diphtheria/ <i>pertussis</i> / tetanus (A)	1	20/20	16/20	9/20	50 × 10 ⁶	73 × 10 ⁶
	2	19/20	19/20	9/20	50 × 10 ⁶	
	3	17/20	16/20	4/20	120 × 10 ⁶	
Diphtheria/ <i>pertussis</i> / tetanus (B)	1	19/20	17/20	10/20	50 × 10 ⁶	55 × 10 ⁶
	2	19/20	18/20	11/20	50 × 10 ⁶	
	3	17/19	15/19	8/20	65 × 10 ⁶	

TABLE II.—Results of Mouse Protection Tests on 12 Batches of *H. pertussis* Vaccines

Batch	Date of Challenge	Mice Surviving to 14th Day after Challenge at Dose (Orgs)			ED ₅₀ (Orgs) × 10 ⁶
		40 × 10 ⁶	200 × 10 ⁶	1,000 × 10 ⁶	
36	12/2/54	2/15	7/15	15/15	150
37	12/3/54	2/15	5/15	14/15	180
39	19/3/54	1/15	2/12	8/11	550
40	19/3/54	2/15	7/15	8/14	420
44	11/6/54	1/15	5/15	12/15	320
45	11/6/54	3/15	4/13	9/13	380
51	23/3/55	2/15	5/15	12/15	290
52	23/3/55	1/15	8/15	10/15	300
53	23/3/55	2/14	8/15	13/15	180
54	20/5/55	0/14	2/15	8/12	620
55	20/5/55	0/14	3/14	10/15	560
56	20/5/55	0/15	7/15	10/14	240
Total		16/177	63/174	129/169	

TABLE III.—Results of Mouse Protection Tests on 16 Batches of Diphtheria-Pertussis-Tetanus Prophylactic

Batch	Date of Challenge	Mice Surviving to 14th Day after Challenge at Dose (Orgs)			ED ₅₀ (Orgs) × 10 ⁶
		40 × 10 ⁶	200 × 10 ⁶	1,000 × 10 ⁶	
1	5/8/53	6/14	10/14	14/14	64
2	11/12/53	2/12	6/10	12/12	130
3	11/12/53	1/12	7/12	12/12	140
4	11/12/53	0/12	6/10	9/10	140
5	12/3/54	6/15	11/15	11/15	65
6	12/3/54	6/15	10/15	15/15	72
7	19/3/54	0/16	9/16	10/13	120
8	19/3/54	4/15	7/16	14/16	160
9	11/6/54	4/15	7/16	11/16	270
10	10/12/54	0/10	2/10	10/15	580
11	10/12/54	0/14	4/8	10/11	200
12	10/12/54	0/10	4/14	6/12	720
13	10/12/54	0/12	3/13	12/14	380
14	23/3/55	0/13	2/15	12/15	530
15	23/3/55	1/13	7/14	13/15	230
16	20/5/55	1/7	4/12	9/14	470
Total		31/205	99/210	180/219	

vaccine and those of D.P.T. do not vary among themselves more than could be attributed to chance. Thus for *H. pertussis* vaccine $\chi^2_{n=11}=9.00$, and for D.P.T. $\chi^2_{n=15}=19.95$, neither being significant.

If the total mortalities for the batches of the two prophylactics shown in Tables II and III are regarded as assays of relative activity, the activity of D.P.T. in terms of plain *H. pertussis* vaccine is 1.52 times, with a fiducial range ($P=0.95$) of 1.17 to 2.00. Since this lower limit is greater than 1.00 the activity of the pertussis antigen seems to have been significantly enhanced by the addition of the toxoids: at least it has not been reduced.

Discussion

The so-called "interference" phenomenon, demonstrated by Glenny and Waddington (1926) on experimental animals, is noticeable only in certain experimental conditions and is not a general one. It was found in earlier experiments (Ungar, 1954) that there was an optimum amount of vaccine or toxoid that could usefully be combined and that any further amount of either component in the combined product would not improve the antigenic response. Barr and Llewellyn-Jones (1953) showed in their animal experiments that the time of successive doses for primary and secondary immunization with combined prophylactics should be carefully considered so as to avoid the "crowding effect" described by Glenny and Waddington (1928); the implication of this finding should be borne in mind when studying the mechanism of antigenic response in children.

The available evidence, based on experience in the U.S.A. (Kendrick *et al.*, 1947; Pittman, 1954) and in this country (Evans and Perkins, 1954; Butler and Ungar, 1956), indicates that the mouse test might serve as a means of differentiating between those vaccines that are effective in children and those that are not.

Our past experiments and those recorded here seem to show that the combined prophylactic, consisting of the three antigens, gives mice a degree of protection against a challenge with *H. pertussis* similar to that given by a plain pertussis vaccine.

Summary

The addition of diphtheria toxoid or of diphtheria and tetanus toxoids to a pertussis vaccine has no adverse effect on the antigenic response of mice to it.

This finding indicates that combined prophylactics will induce in children the same degree of protection against whooping-cough as will a plain suspended vaccine.

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Speaking at St. Paul's Cray, Kent, on April 4, Miss PAT HORNSBY-SMITH, Parliamentary Secretary to the Ministry of Health, said it was sometimes suggested that a child vaccinated against poliomyelitis could be infectious to others by reason of vaccination. But with the dead vaccine being used, and with the stringent safety tests to which it was subjected before being used, this could not happen. "I should like to emphasize," she said, "that it is quite impossible for a vaccinated child to infect others simply because he has been vaccinated."