fully conscious 12 hours after admission there was gross abdominal distension, bowel sounds were poor, and he was passing frank blood per rectum. At laparotomy an oedematous reddened area was observed at the rectosigmoid junction, the histology of which showed gangrenous change in the mucosa. As no definite obstruction was found and the small bowel appeared healthy, the abdomen was closed. Postoperatively his diabetes was well controlled (see Table). The abdomen, however, remained distended and signs of intestinal obstruction persisted. A second laparotomy was carried out on the evening of the third day of admission. The rectum was inflamed and oedematous and a left iliac loop colostomy was performed. His condition deteriorated postoperatively and he died early on the fourth hospital day.

At necropsy there was a haemoperitoneum. Both large and small intestines were oedematous and many small haemorrhagic lesions were scattered in the mesentery of the small bowel. The liver showed evidence of early cirrhosis. The large bowel from the mid-transverse colon to the rectum showed established post-mortem autolytic changes.

Discussion

While abdominal complications are not uncommon in diabetic ketosis (McKittrick, 1933; Hirsch, 1960; Katz and Spiro, 1966) apart from pancreatitis (Davidson, 1964) they do not appear to have been reported with any frequency in hyperosmolar coma. Thrombotic complications were noted in two of the 16 patients with hyperosmolar coma reported by Pyke (1969); in one a femoral artery thrombus was removed surgically, but the other died with pulmonary artery thrombosis. Both of our patients developed intra-abdominal complications due to mesenteric thromboses. One developed an acute abdomen with massive gastrointestinal bleeding. At necropsy the jejunum and the upper third of the ileum were haemorrhagic and showed evidence of recent infarction due to venous thrombosis. In addition, a pulmonary embolism was present. The other developed an acute abdomen with moderate gastrointestinal bleeding. At the initial laparotomy a gangrenous area was found in the rectum, but as there was no apparent mechanical obstruction no bypass procedure was carried out. At a second laparotomy 36 hours later the area of infarction was more extensive and a colostomy was constructed. Despite this he continued to deteriorate. At necropsy multiple haemorrhages were present in the mesentery of the small intestine in addition to a moderate haemoperitoneum. There is little doubt that in both these patients the intra-abdominal catastrophes contributed to death.

When thromboses occur in intra-abdominal vessels they pose particularly difficult clinical problems in these patients. Laparotomy, though it carries a high risk, may need to be considered much more seriously than in patients with ketotic diabetic coma. The pathological features of the thrombosed vessels are non-specific. It is of interest that both the above patients were West Indians, as hyperosmolar coma appears to be relatively common in Jamaicans (Pyke, 1969).

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References

Di Benedetto, R. J., Crocco, J. A., and Soscia, J. L. (1965). *Archives of Internal Medicine*, **116**, 74.
Davidson, A. I. G. (1964). *British Medical Journal*, **1**, 356.
Ehrlich, R. M., and Bain, H. W. (1967). *New England Journal of Medicine*, **276**, 683.
Hirsch, M. L. (1960). *Diabetes*, **9**, 94.
Katz, L. A., and Spiro, H. M. (1966). *New England Journal of Medicine*, **275**, 1350.
Lucas, C. P., Grant, N., Daily, W. J., and Reaven, G. M. (1963). *Lancet*, **1**, 75.
McKittrick, L. S. (1933). *New England Journal of Medicine*, **209**, 1033.
Oakley, W. G., Pyke, D. A., and Taylor, K. W. (1968). *Clinical Diabetes and its Biochemical Basis*, p. 420. Oxford, Blackwell.
Pyke, D. A. (1969). *Journal of Clinical Pathology*, **22**, Suppl. No. 2, p. 57.
Tyler, F. H. (1968). *American Journal of Medicine*, **45**, 485.

PRELIMINARY COMMUNICATIONS

Agglutinin Response to Pertussis Vaccination in the Child

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Summary

Children were immunized with plain pertussis vaccine made by three manufacturers in 1967. After a primary course of three injections at monthly intervals, starting at 3-4 months of age, the agglutinin response was poor. Even after a "booster" dose, given five months later, not all of the vaccines had stimulated a response to all three pertussis agglutinogens.

A further investigation with current vaccines of different kinds administered according to more than one schedule is recommended.

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Introduction

The effectiveness of pertussis vaccination has been questioned in recent years. An investigation by the Public Health Laboratory Service (1969) showed that some of the vaccines used before or during 1967 were not very effective, as shown by a comparison of the attack rates in vaccinated and unvaccinated children. Perkins (1969) pointed out that these vaccines would "have been made, as bulk components, before 1966," and that vaccines made after 1966 have been modified by increasing their minimal potency requirement, as indicated by the mouse protection test, and also by including some of the prevalent type 1,3 strains in the vaccine.

We are reporting here on the agglutinin response of the child to the vaccines of three manufacturers which were made in April, June, and July of 1967, and each of which was stated to contain antigens 1, 2, and 3.

Materials and Methods

Pertussis Vaccines.—The vaccines of three manufacturers (X, Y, and Z) were used. The batches had been manufac-

tured in 1967 (July, June, and April respectively) and were freely available in this country. All were plain vaccines, without adjuvant, and were in the form of triple antigen (diphtheria and tetanus toxoids together with *Bordetella pertussis*). In each case the pertussis component consisted of 20,000 million organisms per 0·5-ml dose, and it was said to include antigens 1, 2, and 3. All the vaccines were stored at 4° C. The first children in this series were vaccinated in August 1967 and the last received their "booster" doses in January 1969.

Immunization of Children.—The mothers of children attending the neonatal assessment clinic in Hope Hospital, Salford, were offered a course of injections for their children, to be followed by a laboratory check on the response. The children had no obvious physical disorder, they had not been born prematurely, and they had no known pertussis contact. They were assigned, in rotation, to three groups for immunization with the vaccines of manufacturers X, Y, and Z. At subsequent visits each child received the same vaccine as previously. At the first visit, when the children's ages ranged from 9 to 21 weeks, samples of blood were taken from mother and child, and vaccine was injected intramuscularly into the outer thigh of the child. At intervals of four to six weeks second and third doses of vaccine were given. Six weeks after the third dose of vaccine a second sample of blood was taken from the antecubital vein or a scalp vein of the child. A fourth (booster) dose of vaccine was given five months after the third, and a further sample of blood was taken six weeks later.

Immunization of Rabbits.—Three groups of five rabbits were immunized by intravenous injection of the vaccines of the three manufacturers. Three rabbits in each group, previously shown to have bordetella agglutinin titres of less than 20, were immunized by giving each rabbit three doses of 12,000 million organisms in the first week and three doses of 24,000 million in the second week—a total of 108,000 million. With two rabbits in each group previously shown to have bordetella agglutinin titres of less than 4 the course of injections consisted of only two doses of 12,000 million organisms in the first week and the same in the second week—a total of 48,000 million per rabbit. A further sample of blood was taken from each rabbit 10 days after the last injection.

Estimation of Pertussis Agglutinins in Sera.—The content of agglutinin 1 was estimated by titration of serum against the type 1 strain of *Bord. pertussis*, GL353. In the samples which failed to agglutinate this strain the titres of the sera against strains 36OE (type 1,2) and H36 (type 1,3) were taken to indicate the content of agglutinins 2 and 3 respectively. Sera that contained agglutinin 1 were absorbed with strain GL353 (type 1) until they no longer agglutinated it, and their content of agglutinins 2 and 3 were then estimated by titration of the absorbed serum against strains 36OE and H36. The details of these techniques have been recorded by Preston (1966, 1970).

Results

Pertussis Agglutinins in Normal Sera.—The sera of most of the mothers contained agglutinin 1 but only a minority had agglutinin 2 or 3 (Table I). The presence of these antibodies did not appear to be in any way related to the mothers' ages, which ranged from 18 to 35 years. Before vaccination only six of the children's sera contained pertussis agglutinin. There was a clear relation between the agglutinin titres of the children's sera and those of their mothers' sera, which suggested that the agglutinin in the child was residual maternal antibody which had not yet been eliminated completely.

Agglutinin Response to Vaccination.—Of the 24 children who were included in this investigation 18 completed the

TABLE I—*Pertussis Agglutinins in Normal Children and Their Mothers*

Age of Child (in weeks)	Titres of Three Pertussis Agglutinins in Serum of					
	Mother			Child		
	1	2	3	1	2	3
9	8	0	(20)	0	0	(4)
12	16	0	0	0	0	0
13 ..	0	0	0	0	0	0
	8	0	0	0	0	0
	16	0	0	0	0	0
	32	0	(10)	0	0	0
	64	0	(10)	(8)	0	0
14 ..	0	0	0	0	0	0
	0	0	0	0	0	0
	128	0	(10)	16	0	0
15 ..	16	0	0	0	0	0
	16	0	0	0	0	0
	0	0	16	0	0	0
	32	0	0	0	0	0
	32	(20)	10	0	0	0
	16	160	40	0	32	8
16 ..	8	0	0	0	0	0
	16	10	0	0	0	0
	64	10	10	0	0	0
20 ..	32	0	0	0	0	0
	32	0	80	0	0	4
	640	80	0	32	(10)	0
21 ..	8	0	(10)	0	0	0
	16	(10)	0	0	0	0

( ) = Weak reaction. 0 = Less than 4 (less than 10 for agglutinin 2 or 3 if agglutinin 1 present).

TABLE II—*Pertussis Agglutinins in Sera of Vaccinated Children*

Vaccine	Age of Child at First Injection (in weeks)	Titres of Three Pertussis Agglutinins								
		Before First Injection			After Three Doses at Monthly Intervals			After a Fourth (Booster) Dose		
		1	2	3	1	2	3	1	2	3
X	12	0	0	0	0	8	4	—	—	—
	13	0	0	0	0	0	8	4	32	32
	13	0	0	0	0	0	0	—	—	—
	15	0	0	0	8	40	(10)	16	640	320
	15	0	0	0	0	0	0	16	0	0
	15	0	32	8	0	8	0	—	—	—
	20	32	(10)	0	16	40	40	16	80	160
Y	14	0	0	0	0	8	0	32	80	(10)
	15	0	0	0	16	10	10	16	80	0
	16	0	0	0	0	16	0	32	40	0
	21	0	0	0	0	32	0	16	80	0
Z	13	0	0	0	0	32	0	16	80	0
	14	0	0	0	8	0	0	64	40	0
	15	0	0	0	32	0	0	—	—	—
	16	0	0	0	0	32	0	16	320	0
	16	0	0	0	16	0	0	16	0	0
	20	0	0	4	0	64	0	—	—	—

( ) = Weak reaction. 0 = Less than 4 (less than 10 for agglutinin 2 or 3 if agglutinin 1 present). — = Not tested (booster dose not given, or blood sample not obtained subsequently).

TABLE III—*Pertussis Agglutinins in Sera of Vaccinated Rabbits*

Vaccine	Rabbit	Titre of Three Pertussis Agglutinins								
		Before First Injection			After a Total Dose of					
					48,000 million Bacteria			108,000 million Bacteria		
		1	2	3	1	2	3	1	2	3
X	1	0	0	0				320	1,600	1,600
	2	0	0	0				1,280	800	400
	3	0	0	0				2,560	3,200	1,600
	10	0	0	0	80	640	80			
	11	0	0	0	80	320	160			
Y	4	0	0	0				160	1,600	200
	5	0	0	0				640	3,200	400
	6	0	0	0				320	3,200	400
	12	0	0	0	40	320	80			
	13	0	0	0	40	2,560	40			
Z	7	0	0	0				320	1,600	0
	8	0	0	0				160	1,600	0
	9	0	0	0				40	800	0
	14	0	0	0	80	640	0			
	15	0	0	0	80	320	0			

0 = Less than 20 (less than 10 for Agglutinin 3 in rabbits 14 and 15).

primary course of three injections, and samples of blood were obtained from 17 both before and after the primary course. Since the agglutinin response in most children was poor, a booster dose was offered and a final sample of blood was obtained in 12 cases. From Table II it can be seen that after the primary course most of the children did not have detectable antibody to all of the three pertussis agglutinogens, and there was no clear distinction between the three batches of vaccine. After a booster dose, however, most of the children had responded to antigens 1 and 2, but only vaccine X had stimulated a response to antigen 3. Corresponding differences between the three vaccines were found in vaccinated rabbits (Table III). Agglutinins 1 and 2 were produced in all three groups of rabbits; but agglutinin 3 was produced in high titre by vaccine X, in lower titre by vaccine Y, and not at all by vaccine Z.

Discussion

Though the numbers of children and animals in this investigation are small the results give a clear indication of the lines along which further study may be profitable. Perhaps the most striking result was the poor agglutinin response in children who were given three doses (at monthly intervals) of pertussis vaccine made in 1967.

If these agglutinins are important in immunity (Preston, 1966) it would be desirable to determine the agglutinin response to current vaccines by an investigation similar to the one reported here. It would also be possible to compare plain vaccines with those containing adjuvant and to assess the relative efficacies of different immunization schedules. Moreover, it may be possible to predict, from the agglutinin response obtained in the rabbit, the likely response of the child to a batch of vaccine.

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References

Perkins, F. T. (1969). *British Medical Journal*, **4**, 429.
Preston, N. W. (1966). *Journal of Pathology and Bacteriology*, **91**, 173.
Preston, N. W. (1970). *Laboratory Practice*, **19**, 482.
Public Health Laboratory Service Report (1969). *British Medical Journal*, **4**, 329.

MEDICAL MEMORANDA

Hepatitis-associated Antigen in Liver Disease in Kenya

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The presence of hepatitis-associated antigen in the serum of patients with acute viral hepatitis (Blumberg *et al.*, 1967; Prince, 1968) and of patients with cirrhosis (Wright *et al.*, 1969; Sherlock *et al.*, 1970) suggests that there may be an aetiological relationship between these conditions. In Kenya viral hepatitis is a severe disease (Jindani *et al.*, 1970) and cirrhosis is often encountered. The following is a preliminary report of the occurrence of hepatitis-associated antigen in sera from patients with acute and chronic liver disease in Kenya.

Present Study

Sera from 225 patients attending the Kenyatta National Hospital with a variety of liver diseases and from 200 apparently healthy Nairobi blood donors (Table I) were tested for the presence of hepatitis-associated antigen. A diagnosis of viral hepatitis was made in the presence of jaundice of recent onset associated with raised serum alanine aminotransferase and normal or only slightly raised serum alkaline phosphatase (up to 36 King-Armstrong units). The diagnosis was supported by a clinical course consistent with viral hepatitis. Twenty-six patients were tested during the acute illness and 49 after complete recovery. A diagnosis of cirrhosis was confirmed on liver histology in 31 patients; in 18 needle biopsy was contraindicated or was unsuccessful. The diagnosis of primary carcinoma of the liver was confirmed histologically in all cases. Other diseases studied included primary extrahepatic portal hypertension, schistosomal hepatic fibrosis, and idiopathic tropical splenomegaly (Pitney, 1968). All patients with primary extrahepatic portal hypertension had normal liver function and normal liver histology.

Sera were tested for hepatitis-associated antigen by the gel-diffusion technique with tris agarose medium as described by Prince (1968). Antibody used for routine testing came from two Kenyan patients and gave a positive reaction of identity with specific sera. The results of this study are summarized in Table I.

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Discussion

The reported incidence of the antigen in acute viral hepatitis ranges from 12·3% of 358 patients in Japan (Okochi and Murakami, 1968) to as high as 80% of 49 patients in the U.S.A. (Gocke and Kavey, 1969). This variation may reflect

TABLE I—*Occurrence of Hepatitis-associated Antigen in Liver Disease and in Blood Donors in Nairobi. Figures in Parentheses indicate Total Number Tested*

Diagnosis	No. Positive	% Positive
Acute viral hepatitis (26)	14	54
Recovered from viral hepatitis (49)	1	2
Cirrhosis (39)*	8	20·5
Primary liver cancer (22)	3	14
Other conditions (89) (see text)	5	5·6
Nairobi blood donors (200)	12	6

*Including five patients with chronic active hepatitis.