NOTES ON DYSENTERY VACCINATION.

BY

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DYSENTERY vaccination is of such great practical importance that one wonders that so little attention, comparatively speaking, has been paid to it since the pioneer work of Kruse and Shiga. Such vaccination has never been used to any large extent, owing, I believe, to the following reasons:

1. Vaccines containing the Shiga-Kruse bacillus, prepared according to the usual technique (broth cultures killed by heat), give an extremely severe local and general reaction. I have seen persons incapacitated for work for more than two weeks after such inoculation. In the lower animals, too (rabbits), such vaccines produce very severe symptoms and sometimes death.

2. A vaccine prepared with Shiga-Kruse bacillus does not give immunity for forms of dysentery due to Flexner, Flexner-like, and other dysentery germs.

Peptone-Water Mixed Vaccine.

The only way to obviate these two drawbacks is to prepare the Shiga-Kruse vaccine by a method which will decrease its irritating properties, and to use a mixed vaccine instead of a mono-vaccine. I have carried out experiments on the subject since 1903 and 1904. In the *Ceylon Medical Reports* for 1904 I published the results I arrived at in that year. I stated that as the anti-dysentery vaccine prepared with broth cultures gives rise to very severe symptoms, great tenderness and redness of the skin, very often enlargement of the neighbouring glands, and fever, which may be very high, it had occurred to me to use peptone water cultures instead of broth cultures. This simple modification of the technique was most satisfactory; the irritating properties of the vaccine practically disappeared without apparently any distinct decrease in its immunizing power. I did not think this result was merely due to the fact that peptone-water cultures contained a smaller number of bacteria than broth cultures.

Preparation.

The Shiga-Kruse bacillus and three species of dysentery-like germs (paradysentery), common in Ceylon, were grown in peptone-water tubes for three days at blood temperature. They were then kept for one hour at the temperature of 60° C. Using all aseptic precautions, the different cultures were mixed in this proportion:

Dysentery	...	...	3 parts
Paradysentery No. 1	...	...	1 part
Paradysentery No. 2	...	...	1 "
Paradysentery No. 3	...	...	1 "

The resulting mixture was the vaccine, of which I inoculated 1 c.cm. (sometimes $\frac{1}{2}$ c.cm.), the injection being repeated one or two weeks later.

The inoculation was followed in most cases by practically no local symptoms. In other cases it was followed after five or six hours by mild signs of reaction—some little tenderness and redness of the skin at the place of inoculation. In some instances the temperature rose to 100° or 101° F.

The conclusions I came to were that the inoculation of the mixed peptone-water cultures vaccine was harmless and that the blood of the inoculated persons agglutinated the *Bacillus dysenteriae* (Shiga-Kruse) and the various strains of paradysentery with which the vaccine had been prepared, though the agglutinins were in small amount and disappeared after a short time.

Carbolized Mixed Dysentery Vaccine.

Since 1912 I have prepared and used in Ceylon a carbolized mixed dysentery vaccine, consisting of an emulsion of Shiga-Kruse, Hiss Y bacillus, original Flexner bacillus, a Flexner-like bacillus No. 1, isolated in Ceylon, a Flexner-like bacillus No. 2, also isolated in Ceylon.

Preparation.

The individual vaccines are prepared by making emulsion from twenty-four hours agar cultures, in normal salt solution (0.75 per cent.), to which 0.5 per cent. of carbolic acid had been added. All particulars of the technique are given in my

previous papers on the subject of mixed dysentery vaccines. The individual vaccines are standardized as follows per c.cm.:

Shiga-Kruse bacillus	...	...	1,000 million.
Flexner bacillus	...	...	1,000 "
Hiss Y bacillus	...	...	1,000 "
Flexner-like No. 1	...	...	1,000 "
Flexner-like No. 2	...	...	1,000 "

These vaccines are mixed in equal parts, so that 1 c.cm. of the mixed vaccine will contain 125 million of each of the organisms used.

Of this vaccine, 0.5 to 0.6 c.cm. is given hypodermically the first time, and the same amount after a week. The reaction is not very severe, though more marked as a rule than after the typhoid-paratyphoid vaccine. Agglutinins generally develop for all the germs of the dysentery group which have been injected, but their amount is not high, the agglutination limit seldom being higher than 1 in 40, and is somewhat inconstant and irregular; but the same may be said of individuals inoculated with simple mono-vaccines, Shiga-Kruse, Flexner, etc.

Combined Typhoid+Paratyphoid A+Paratyphoid B +Dysentery.

This combined vaccine, on which I have published several previous papers, consists of an emulsion of Shiga-Kruse, Hiss Y bacillus, original Flexner bacillus, a Flexner-like bacillus No. 1, isolated in Ceylon, a Flexner-like bacillus No. 2, also isolated in Ceylon, typhoid bacillus, paratyphoid A bacillus, and paratyphoid B bacillus.

Preparation.

The individual vaccines are prepared by making emulsions from twenty-four hours agar cultures in normal salt solution (0.75 per cent.), to which 0.5 per cent. of carbolic acid has been added.

The individual vaccines are standardized as follows per cubic centimetre:

Typhoid bacillus	...	...	4,000 million.
Paratyphoid A bacillus	...	...	1,000 "
Paratyphoid B bacillus	...	...	1,000 "
Shiga-Kruse bacillus	...	...	1,000 "
Flexner bacillus	...	...	1,000 "
Hiss Y bacillus	...	...	1,000 "
Flexner-like bacillus No. 1	...	...	1,000 "
Flexner-like bacillus No. 2	...	...	1,000 "

These vaccines are mixed in equal parts, so that 1 c.cm. of the mixed vaccine will contain 200 million of the typhoid bacillus and 125 million of each of the other organisms used.

Of this vaccine, 0.5 to 0.6 c.cm. is given hypodermically the first time, and the same or double the amount after a week. The reaction is severer as a rule than after the typhoid-paratyphoid vaccine. As regards agglutinins, the agglutination for the germs of the dysentery group is not high, the agglutination limit seldom being higher than 1 in 40; it is also somewhat irregular and inconstant, but to a certain extent the same may be said of individuals inoculated with simple Shiga-Kruse, Flexner, etc., mono-vaccines. Typhoid, paratyphoid A and paratyphoid B agglutinins, on the other hand, are produced in fair amount, though as a rule distinctly less than in control individuals inoculated with simple typhoid, paratyphoid A and paratyphoid B vaccines.

CONCLUSIONS.

1. A dysentery vaccine should contain several species of dysentery bacteria, and not one only.

2. As I have stated in all my publications since 1904, the vaccine should never be prepared with broth cultures, as such vaccine often gives a most serious reaction with very severe infiltration and occasionally an abscess.

3. The vaccine should be prepared with a carbolic salt emulsion from agar cultures without heating, or peptone-water cultures may be used. Of sensitized dysentery vaccines I have little experience.

4. It is essential to prepare the vaccine with strains which, though rich in antigen, are not virulent, or very little so. It is very rare to come across such a strain of the Shiga-Kruse germ. A large series of strains of this organism should be inoculated in rabbits, and the least virulent, provided it be rich in antigen, should be kept permanently as a stock culture to prepare the vaccine.

REFERENCES.

Castellani: *Ceylon Medical Reports*, 1904; *Centr. f. Bakt.*, 1909; *Journal*, Ceylon Branch British Medical Association, November, 1914; *Reports to the Advisory Committee, Tropical Research Fund*, 1914; *Sperimentale*, April, 1915. Johnson and Milne: *BRITISH MEDICAL JOURNAL*, January 15th, 1916.