

Communications on the examinations for the diagnosis of whooping cough at the Serum Institute of the State.

By

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Since the opening in February 1916 of the whooping cough diagnosis department of the Serum Institute of the State there have been sent in for examination up to September 1920 — besides some expectorations and blood tests for examination of complement-fixation — 1665 tests in which the cough sowing method which was published by me in 1916¹ was used.

At the request of Dr. TH. MADSEN, I have as a continuation of my previous publications^{2, 3} sent schedules con-

¹ Lecture delivered at the Medical Association at Copenhagen on the 4th april 1916, and published by Dr. CHIEVITZ and myself in the *Ugeskrift for Læger* 1916, No. 34 and in the '*Arch. für Kinderheilkunde*' Bd. LXVI Heft 3—4 p. 186; also in a somewhat different form in the '*Annales de l'Institut Pasteur*' October 1916 Tome XXIX p. 503.

² 'A method for the early diagnosis of whooping cough' published together with Dr. CHIEVITZ in the commemoration paper of the Serum Institute for Prof. CARL JUL. SALOMONSEN 1917; also in the *Münchener mediz. Wochenschr.* 1918, No. 27, p. 729.

³ The cough sowing method for the early diagnosis of whooping cough, 'Lecture delivered at the First Northern Congress on Pediatrics' August 1918. (It was issued with the Congress discussions 1919.)

taining a list of questions, concerning the tests in which the whooping cough bacilli were not found on examination at the Serum Institute. In the following table will be found the results of all the 1665 tests.

Table I.

Stage of the cough	Number of tests	Whooping cough bacilli were present (+)
Catarrhal Stage	134	100 = 75 %
Convulsive Stage 1st week	277	158 = 57 %
» » 2nd »	201	122 = 61 %
» » 3rd »	121	55 = 45 %
» » 4th »	74	30 = 40.5 %
» » 5th » and over	107	10 = 9 %
Unknown Stage	56	
Doubtful whooping cough	45	
Information wanting	41	
Not whooping cough	559	
Unusable tests (overgrown with other organisms, or shaken)	50	
	1665	

As will be seen, it treats of 970 cases of tests from patients suffering from whooping cough. In 914 of these cases, the stage of the disease can be almost stated to a certain extent, and it proves, that of 134 patients in the catarrhal stage, whooping cough bacilli were found in 100, namely 75%; of 107 patients who have had the cough for more than 5 weeks, the whooping cough bacilli were found in 10 cases namely 9%; none of these latter had coughed more than 5 to 6 weeks, therefore none of them had the typical

whooping cough attacks longer than 5 weeks. Patients about whom we have made inquiries, and found that they have coughed for 1, 2, 3 weeks etc. are noted in the columns of the tables, under the headings, convulsive stage 1 week, convulsive stage 2 weeks, convulsive stage 3 weeks etc. Under the heading catarrhal stage are only noted the patients who have been stated to cough less than 1 week.

The longer the whooping cough has lasted, the less often the bacilli have been found by the cough sowing method, which corresponds well with the diminishing period of infection. When the whooping cough attacks have lasted 5 weeks, the patient is practically free of infection, although continued examinations perhaps may show rare cases where patients are infectious even past that stage.

The more the staff of the Serum Institute has acquired practice in proving the whooping cough bacilli in the boxes, the oftener they have succeeded in proving the bacilli after the first weeks in the stages, where the colonies as a rule arise in smaller quantities which will be seen from the following table.

Table II.

The first 209 cases of whooping cough examined <i>Feb. 1916—April 1917</i>				The next 196 cases of whooping cough examined <i>April 1917—June 1919</i>		The last 509 cases of whooping cough examined <i>June 1919—Sept. 1920</i>		
Number of tests				Whooping cough bacilli were present in	Number of tests	Whooping cough bacilli were present in	Number of tests	Whooping cough bacilli were present in
Catarrhal stage 63				49=77 %	16	10=63 %	55	41=75 %
Convulsive stage 1st week 58				30=52 %	75	39=52 %	144	89=62.5 %
" " 2nd " 32				16=50 %	40	15=38 %	129	91=70 %
" " 3rd " 13				4=30 %	33	11=33 %	75	40=53 %
" " 4th " 10				1=10 %	15	6=40 %	49	23=47 %
" " 5th " 33 and over				0	17	1= 6 %	57	9=16 %

On the basis of publication of the first examinations permission was given by the Board of Education in a pamphlet of March 15th 1916 for children to go to school after the whooping cough attacks had lasted for 4 weeks, although apparently no bad effects have followed, it might probably be wiser, as a consequence of continued examination, on a future revision of the question to prolong the isolation to 5 weeks from the commencement of the attacks.

It is of great importance, that the results of the first examinations, concerning the frequency of the bacilli of whooping cough in the catarrhal stage, have been confirmed. By help of the cough sowing method, one is able in $\frac{3}{4}$ of the cases, to diagnose whooping cough in that stage which is of practical importance especially in children's homes, asylums and children's hospitals, as it is in this stage that the infection is the greatest, and there is no other sure method of diagnosis. Of course only a positive finding of the bacilli is of diagnostic importance.

The Serum Institute sends on request by return either by letter or telephone aluminium cough boxes, containing Bordet's nutritive substrate, together with directions how to use same and encloses envelopes for returning them.

The production of the nutritive substrate, which is slightly modified from the method which is given by Bordet and Gengou, is as follows; 500 grammes of peeled potatoes are cut in slices, and put in a pot of one litre of distilled water and 40 cubic-centimetres of glycerine, and boiled to a mash. The mash is then rubbed through a sieve, and after that is wrung in a cloth. The potato extract is then diluted, in 3 times as much distilled water, to which is added 6 promille Na Cl. The extract is then boiled and poured into retorts, 300 cubic centimetres extract in each. To each retort is added 3 % agar. The retorts are heated in the autoclave and the completed potato agar can now be preserved in an ice cellar for a very long time, at any rate for about 1 year.

The final nutritive substrate is now prepared in the following manner. One retort, containing 300 cubic centimetres of potato agar, is melted in water bath or autoclave, and is cooled down to about 45° Cels.; 300 cubic centimetres of sterile defibrinated horse blood is heated in water bath to the same temperature, 45° Cels. and is then poured with careful shaking into the melted potato agar. The potato blood agar is put into round sterilized slightly grooved aluminium boxes, of 8 centimetres in diameter and 1.5 centimeter high, with a lid like Petri's cups. The boxes are sterilized in the same way as the Petri's cups, with a layer of paper between the box and the lid, and before pouring the warm liquid substrate in, the lid of the box is removed, but not the layer of paper. It is only a corner of the layer of paper which must be lifted very carefully, when pouring in the substrate. In order to avoid condensation water, the layer of paper is not removed and replaced by the aluminium lid, until the substrate has stiffened, and is of the same temperature as the room. The finished substrate must be a solid red mass. If the substrate is too soft, then the potato extract must contain 4 or 5 % agar instead of 3 %.

The cough sowing method is carried out, as described viz. the lid must be removed, and the patient made to cough towards, but *not* expectorate in the box. The lid is then placed on the box, is packed and sent to the Institute, where it is immediately placed in the thermostat. Then in certain stages of whooping cough, colonies of whooping cough bacilli appear in the course of 2 or 3 days, they are, as a rule, easily recognized from the other colonies, by their shining appearance. They appear in different numbers, from very few colonies up to a very large number.

Although practising doctors have to a great extent made use of the cough sowing method for diagnostic purposes, and have also sent in expectorations to be examined for whooping cough bacilli, very few blood tests have been sent in, for examination of complement-fixation. There are not so few cases, where this method of examination is of diagnostic importance, and can be of great help to the doctors in practice, as in such cases where a cough has lasted for a long time, one would like to know, if it has been whooping cough. It is

known from former examination¹ that the complement-fixation during the whooping cough appears in the 3rd week of the disease, and may last for months even for several years.

¹ In the Hospitalstidende 1915 No. 25, Ugeskrift for Læger l. c., Arch. f. Kinderheilk. Bd. LXIV, Heft. 5—6 and Bd. LXVI l. c. Annales de l'Institut Pasteur, l. c.