THE TREATMENT OF MEDITERRANEAN FEVER BY MEANS OF VACCINES, WITH ILLUSTRATIVE CASES.

By Fleet-Surgeon P. W. BASSETT-SMITH, R.N., D.T.M. & H. (CAMB.) etc.,
Royal Naval Hospital, Haslar.

FOR years we have been vainly striving to find some method of treatment capable of preventing or cutting short the relapses so constantly present in Mediterranean fever. Though certain drugs have for a time seemed successful, their beneficial effects are usually uncertain and transient, so that the treatment of the disease has come to be almost entirely symptomatic. Many germicides have been tried; among these "cyllin" has lately been said to shorten the course of the fever. In my hands this has not been the case. Burney Yeo's chlorine and quinine mixture has here occasionally produced very satisfactory results when given with a rising temperature, apparently cutting short a wave of pyrexia, which from experience we had expected to last ten days or more; but its action is very uncertain.

In this disease a constant intoxication is going on, due to the presence of a specific micro-organism which may apparently flourish in the internal organs for years, rapidly increasing at any favourable opportunity when presumably vitality is lowered, and giving rise to fresh attacks of pyrexia. During these recurrent periods of fever the specific organism is easily isolated from the peripheral blood.

Failing an effective antitoxin, the most successful and rational line of treatment would seem to be the employment of means for increasing and stimulating the "phagocytes" rendering them able to combat this influx of toxin-producing germs. Unfortunately, as I have shown (1902), one of the most constant features of the disease is a relative reduction of the number, as well as the phagocytic activity, of the polymorphonuclear white cells.

In the hope of increasing the number of these polymorphonuclear leucocytes, fresh yeast was given in milk, daily for a month, to the patients in a ward occupied by cases of Mediterranean fever, but with no marked success, and without beneficial effect on the temperature. Koch and Petruschky employed vaccines prepared from the bacilli of tuberculosis and typhoid fever in the treatment of these diseases, and Wright has recently extended and elaborated this method of treatment, having shown that vaccines prepared from the causal organism of a disease, when injected in suitable quantities, are capable of stimulating the body to produce protective substances which are thrown into the blood and modify the clinical manifestations of the disease.

Staff-surgeon S. T. Reid, R.N., in 1905, by applying Wright's method to a small number of cases of Mediterranean fever (9) was apparently so successful that an extensive trial of this treatment was considered desirable in the wards at Haslar, where the cases of this disease are fairly numerous. This trial has now been made during the last eight months on almost all the cases, to the exclusion of any active drug treatment.

In every case the dosage and the nature of the reaction was calculated by recording the opsonic index, as devised by Wright and Douglas (1904). At the same time the amount of the agglutination reaction was also determined.

Preparation of the Vaccine.

A freshly isolated strain of the *Micrococcus melitensis* was used. The culture was taken during life from the spleen of a patient in this hospital.

Agar cultures of ten days' growth were employed, the organisms being scraped off and emulsified in 100 c.c. of distilled water. The emulsion was heated to 60° C. for half-an-hour, and after 0·5 % carbolic acid had been added, sealed up and kept as a stock solution. After twenty-four hours 10 c.mm. of the vaccine were spread on an agar tube to make sure of its sterility.

Estimation of the strength of the Vaccine.

1. *By opacity.* A sample of the vaccine was run out into a square glass bottle, giving a thickness of fluid of 1·5 cm. This just obscured 0·5 Snellen's type.

2. *By dilutions and culture.* By means of a measuring pipette, 5 c.mm. of the freshly prepared emulsion were diluted 50 times, and this again on three occasions, giving a dilution of 1 in 6,250,000. Five c.mm. of this high dilution were plated out, and showed on the fifth day 150 colonies of the *Micrococcus melitensis*, which works out at 937,500,000 organisms per c.mm., or 187,500,000 for each c.mm. of the vaccine.

3. *By weight of the dried organisms.* 20 c.c. of the freshly prepared emulsion, after heating, but before the addition of the carbolic acid, were run off into a long tube. The estimation was kindly undertaken by Dr C. J. Martin, F.R.S., who found that the 20 c.c. of vaccine contained 4 mg. of dried *Micrococcus melitensis*, or 0·2 mg. per c.c.

Method of injection.

About 10 c.c. of the stock vaccine were periodically run off into a sterile bottle with two rubber caps. 0·5 c.c. doses were drawn up into freshly made glass tubes by means of a sterile teat, and then the tubes were sealed in the flame. These were taken to the ward for use, where each dose was expressed into a sterile glass capsule as required.

A 1 c.c. all-glass sterilised syringe was employed for injecting the vaccine. The injections were made in the region of the loin, the part having been prepared with spirit soap, ether and biniodide solution.

Method of noting the effects of the vaccine by the opsonic indices.

Blood from all the cases under treatment in the ward was periodically taken at the same time, at first twice a week and later once a week, and the opsonic index estimated. A few cases were examined daily, the results of these cases being given in the special charts.

My own leucocytes were used throughout for the estimation. The plasma was removed and the cells were well washed four times in 0·9 % normal saline solution. A freshly prepared weak emulsion of a young agar culture of *M. melitensis* was made for each operation. One part of the patients' unheated serum, one part of my washed blood cells, and a half part of the fresh emulsion, were then well mixed and drawn up into small tubes, sealed, and placed in the incubator at 37° C. for 20 minutes. This mixture was then blown out on to slides, films were made, and these, when dry, were fixed in alcohol for ten

minutes, and stained with carbol-toluidin blue for half-an-hour. The opsonic indices of the serum of six surgeons under instruction were ascertained, and as the average of their sera was found to be identical with the index of my own serum, the latter for reasons of convenience, was assumed to be normal, and was employed as the control in the tests.

At least, twenty consecutive normal polymorphonuclear leucocytes were generally counted in each film and the average number of ingested cocci taken.

Agglutination reactions.

Besides testing the serum for the opsonic index, capillary tubes were put up on each occasion with the dilutions 1 in 20, 1 in 200, and 1 in 2000, to demonstrate the quantity of agglutinins present. In the cases that were examined daily, the frequency with which the dilutions were made was much greater, as is seen in the special charts.

Frequency of the injection of the Vaccines.

The vaccines were generally given at ten day intervals, unless the clinical symptoms contraindicated their use.

Number of cases and injections given.

Altogether 224 injections in 61 cases have been given here since the commencement of the treatment. Beyond a certain amount of temporary tenderness in the loin, there were no unpleasant local effects, except some heat and redness in two cases which passed off in twenty-four hours. Occasionally there was a slight rise in the temperature indicating some general reaction. In one case the temperature shot up to 103·6° F. with a slight shiver and headache. In the majority of cases nothing was noted. Most of the men believed strongly in the efficacy of the injections and asked for their repetition; in no case was the vaccine given if the patient raised any objection.

Records of cases treated.

In this paper I have tried to record as far as possible the general clinical characters of the cases under treatment with the marked features in each, and in the charts are shown the temperature, opsonic, and agglutination curves. It is essential to follow the course of fever both

before and after the administration of the vaccines if any deductions are to be drawn, and as the cases lasted over such long periods, I have with this object in view reduced the charts into small dimensions.

Epitome of Cases.

1. *Severe Undulant type.*

No. 1. *Chart* 7. W. E. G. Severe case with high continued type of fever, followed by slight undulations. Vaccines were administered commencing in the 6th week, and were repeated for six weeks without appearing to do any good, they were therefore stopped, but two further doses were given in the 24th and 25th weeks. The opsonic index, which had remained about normal, rose considerably after the two last doses of the vaccine. The agglutinins were very high throughout (1 in 2000).

No. 3. *Chart* 8. V. H. A severe continued type of fever passing into the undulant form. The first vaccines were given in the 12th week. The opsonic index at first rose, then fell rapidly; vaccines stopped. After the next relapse, three injections were given, there being a normal morning temperature; this was followed by a further wave of pyrexia, in spite of a high opsonic index. He however gained two stones in weight and felt much stronger during this four week relapse. He was discharged on sick leave. His agglutinins rose and remained high (1 in 2000).

No. 5. *Chart* 9. C. N. A long undulant type of fever. Vaccines were commenced in the 33rd week of the fever and were given weekly. During the treatment he had three relapses, the last being the most severe. The vaccines were powerless to prevent the reinfections, though the opsonic curve steadily rose. The agglutinins fell. He was invalided in the 42nd week.

No. 7. *Chart* 10. P. A. Admitted for phthisis. Fever of an undulant type with great diurnal variations, (there was cough etc., no tubercle bacilli in sputum). Vaccines were commenced in the third week, repeated about every ten days with apparently no effect on the temperature. After a final severe wave he made steady improvement and gained weight rapidly. The opsonic index though high slowly fell during the long fever, rising again when the temperature was normal. The agglutinins were at first high, fell low during the course of the disease, but rose again in convalescence.

No. 10. *Chart* 11. J. W. Severe undulant type of fever. Vaccines were commenced in the 11th week. The opsonic index rose after the second injection, the temperature fell after the fourth, and remained down, with improvement in nutrition and power. The agglutinins remained very low. The case was finally invalided.

No. 11. *Chart* 12. T. L. Severe undulant type of fever. Vaccines were commenced in the 10th week, a wave of pyrexia having started. This aborted, and there was no further rise. His general condition rapidly improved, gaining 25 lbs. in 21 days. The opsonins rose and the agglutinins were fairly high.

No. 12. *Chart* 13. J. M. Late severe undulant type of fever. Vaccines were commenced in the 33rd week, when a wave had well started, the temperature suddenly fell, remaining down. The opsonic index mounted very high, but the agglutinins were low. This was a brilliant result for the time.

2. *Mild Undulant types.*

No. 21. *Chart* 14. B. C. Undulant type of fever. Vaccines were commenced in the 6th week, when he had a marked purpuric eruption over the body. Four injections were given, his opsonins rose very high and the temperature fell. The agglutinins were moderately high. He steadily gained weight and was discharged cured.

No. 22. *Chart* 15. T. B. Late undulant type with neuritis. Vaccines were commenced in the 22nd week. There was no further rise in the temperature and the opsonic index steadily rose.

3. *Intermittent types of fever.*

No. 46. *Chart* 16. H. H. Intermittent type of fever. Vaccines were commenced in the 16th week. Eight injections were given, which had apparently no effect on the fever. The opsonic index made an irregular but gradual ascent, the agglutination curve steadily fell. The patient was invalided.

No. 47. *Chart* 17. G. G. Slight irregular intermittent fever, with neuritis and effusion into the left knee. Vaccines were commenced in the 26th week. The opsonins rose high and the temperature fell. He gained weight and his general condition improved markedly.

It is not possible to give the details of all the cases referred to, or the charts compiled from them, I have therefore made a short summary of the main facts relating to them.

Severe undulant types of fever. In numbers 2, 4, 6, 8, and 15 relapses were not prevented, and the vaccine did not appear to influence the course of the fever for good. In most of them the opsonic index was very variable, though there was a tendency generally for it to rise in convalescence. Numbers 9, 16, and 17 all showed marked improvement in the course of the temperature, opsonic index, and weight.

Mild undulant types. In numbers 19, 20, 28, 30, 31, 34, and 35 the temperature fell almost at once, and both opsonins and agglutinins rose; in most there was a rapid gain in weight, as much as 14 lbs. in 14 days in one case.

In numbers 18, 23, 26, 29, 33, 37 the temperature fell more slowly, and the opsonic index rose; but the agglutinins remained low. All were discharged to duty.

Case No. 25 was a relapse after one year's apparent freedom from the fever. Vaccines appeared to lower his temperature and hasten his recovery.

Case No. 27 was peculiar in that each injection caused the temperature to rise, on one occasion to 103·6°, but the man steadily improved.

Case No. 32 was of rather special interest. It was an irregular undulant type of fever in an oldish man. During a slight wave of pyrexia one injection of 0·3 c.c. of the vaccine was given, but it caused such marked local and constitutional reaction for a few hours that he refused any further. For 14 days the opsonic index steadily fell, then slowly rose and remained above normal. It is difficult to say how long the one dose of vaccine influenced this curve. The general symptoms slowly improved, but the agglutinins were very low throughout.

In case No. 36 vaccine was given in the 11th week, the temperature abruptly dropped on the second day, but there was no rise in the opsonic index.

Intermittent type of fever. In case No. 48, the temperature fell, the opsonic index rose, and general improvement was marked.

In case No. 49 although the opsonic index rose, there was no marked improvement in his general condition, and his agglutinins kept constantly low.

The total of 47 cases show an improvement in the temperature curve in 28 cases.

VACCINE RECORD.

Table of cases treated.

0·25 c.c. equals 468,750,000 *M. M.* or 0·05 milligram of dried *M. M.*

No. of case	Name	Amounts injected	No. of injections	Subsequent course: Temperature	Subsequent course: Opsonins
1.	W. E. G.	·2 c.c., ·4 c.c., ·25 c.c., ·25 c.c., ·2 c.c., ·2 c.c., ·25 c.c.	7	no improvement	rose at last.
2.	A. R. M.	·25 c.c., ·25 c.c., ·3 c.c., ·25 c.c., ·25 c.c.	5	do.	rose.
3.	V. H.	·25 c.c., ·4 c.c., ·25 c.c., ·4 c.c., ·25 c.c., ·25 c.c.	6	do.	rose at last.
4.	W. M.	·25 c.c., ·3 c.c., ·25 c.c.	3	do.	fell.
5.	C. N.	·3 c.c., ·3 c.c., ·2 c.c., ·2 c.c., ·3 c.c., ·2 c.c., ·5 c.c.[1], ·25 c.c., ·3 c.c.	9	do.	rose.
6.	A. H.	·3 c.c., ·3 c.c., ·5 c.c., ·5 c.c., ·25 c.c., ·3 c.c., ·25 c.c., ·4 c.c., ·3 c.c.	9	do.	irregular.
7.	P. A.	·2 c.c., ·5 c.c., ·3 c.c., ·3 c.c., ·25 c.c., ·25 c.c., ·5 c.c., ·2 c.c., ·25 c.c.	9	do.	steadily fell then finally rose.
8.	G. B.	·25 c.c., ·25 c.c., ·3 c.c., ·25 c.c., ·3 c.c.	5	fell	rose.
9.	C. L.	·25 c.c., ·25 c.c., ·25 c.c., ·25 c.c., ·25 c.c.	5	fell	rose.
10.	J. W.	·1 c.c., ·3 c.c., ·2 c.c., ·25 c.c., ·3 c.c., ·3 c.c.	6	fell	rose.
11.	T. L.	·2 c.c., ·2 c.c., ·25 c.c., ·3 c.c., ·3 c.c.	5	fell	rose.
12.	J. M.	·2 c.c., ·3 c.c.	2	fell	rose.
13.	J. G. B.	·3 c.c., ·3 c.c., ·5 c.c.	3	fell	rose.
14. 39.	T. N.	·3 c.c., ·3 c.c., ·3 c.c., ·2 c.c., ·25 c.c., ·3 c.c., ·25 c.c., ·25 c.c., ·3 c.c.	9	no improvement	irregular.
15.	G. W. M.	·2 c.c., ·25 c.c., ·3 c.c., ·25 c.c., ·5 c.c., ·25 c.c., ·25 c.c.	7	do.	fell.
16.	G. V.	·2 c.c., ·3 c.c., ·25 c.c., ·5 c.c.	4	fell	steady.
17.	W. C.	·25 c.c., ·2 c.c., ·25 c.c., ·3 c.c., ·2 c.c., ·2 c.c., ·25 c.c., ·25 c.c.	9	no improvement	rose slightly.
18.	A. T.	·3 c.c., ·3 c.c.	2	fell	rose.
19.	H. D.	·2 c.c.	1	fell	rose.
20.	F. B.	·2 c.c.	1	fell	rose.
21.	B. C.	·25 c.c., ·25 c.c., ·25 c.c., ·25 c.c.	4	fell	rose.
22.	T. B.	·25 c.c., ·25 c.c.	2	fell	rose.
23.	E. J.	·25 c.c., ·25 c.c., ·25 c.c., ·3 c.c., ·3 c.c.	5	no improvement	rose slightly.
24. 43.	H. F. C.	·25 c.c., ·25 c.c., ·25 c.c., ·3 c.c.	4	fell	rose.
25.	F. M. W.	·25 c.c., ·25 c.c., ·1 c.c., ·4 c.c.	4	improved	rose slightly.
26.	F. H.	·25 c.c., ·25 c.c., ·25 c.c., ·3 c.c., ·3 c.c., ·5 c.c.	6	do.	very regular.
27.	T. D.	·25 c.c., ·25 c.c., ·25 c.c., ·25 c.c.[2], ·3 c.c.	5	no improvement	rose.
28.	W. H.	·25 c.c., ·25 c.c., ·25 c.c., ·3 c.c., ·25 c.c.	5	improved	rose.
29.	J. P.	·25 c.c.	1	kept low	rose.
30.	J. I.	·25 c.c., ·25 c.c.	2	kept low	rose.
31.	T. F.	·25 c.c., ·25 c.c., ·25 c.c.	3	fell	rose.
32.	H. P.	·3 c.c.[3]	1	no improvement	fell for 14 days then rose.
33.	H. H.	·3 c.c., ·3 c.c., ·3 c.c., ·2 c.c., ·4 c.c., ·25 c.c., ·25 c.c.	7	do.	regular.
34.	A. W.	·3 c.c., ·2 c.c., ·25 c.c.	3	do.	rose.
35.	J. S.	·25 c.c., ·5 c.c.	2	fell	steady.
36.	D. G.	·2 c.c., ·4 c.c.	2	fell	rose slightly.
37.	J. W.	·25 c.c., ·3 c.c., ·25 c.c.	3	fell	rose.
38.	W. S.	·25 c.c., ·25 c.c., ·25 c.c.	3	fell then relapsed	rose for a short time.
40.	Mr S.	·25 c.c., ·25 c.c.	2	no improvement	rose.
41.	F. T.	·25 c.c.	1	fell	rose.
42.	Mr C.	·25 c.c.	1	fell	rose.
44.	H. B.	·25 c.c., ·3 c.c., ·35 c.c.	3	fell slowly	rose.
45.	H.	·25 c.c., ·3 c.c.	2	fell	rose.
46.	H. H.	·3 c.c., ·1 c.c., ·2 c.c., ·25 c.c., ·3 c.c., ·3 c.c., ·3 c.c., ·25 c.c.	8	no improvement	irregular rise.
47.	G. G.	·25 c.c., ·3 c.c., ·3 c.c.	3	fell	rose.
48.	G. F.	·25 c.c., ·25 c.c., ·25 c.c.	3	kept low	irregular.
49.	J. C.	·3 c.c., ·3 c.c., ·3 c.c., ·3 c.c., ·3 c.c.	5	no improvement	irregular.

[1] Marked local reaction for 24 hours.
[2] Marked general reaction for 24 hours.
[3] Evening temperature ran up to 103·6° F.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press

General considerations in relation to the temperature.

In Mediterranean fever the course of the temperature is very variable.

1. In some cases we may have a long wave of continued fever ranging from 105° to 102° F. lasting perhaps for 7 or 8 weeks, the condition of the patient closely simulating that seen in typhoid fever. After short apyrexial intervals, fresh waves of fever follow, each becoming as a rule shorter and less severe.

2. Cases where from the commencement the undulations of pyrexia are more moderate in intensity and duration, but pyrexia recurs over and over again, and the late relapses may be of any severity.

3. Cases which have lasted for months (4 to 12 or more) having an irregular intermittent type of fever, generally accompanied with neuralgic pains, chronic joint affections, and cachexia. In the majority of cases there is a great tendency for the waves of pyrexia gradually to die out as the duration of the disease extends.

Importance of the temperature curve as an index of the clinical condition.

No case of Mediterranean fever can be correctly understood without having a continuous temperature chart of the case to study. By following the course of the fever month by month a fair index is given of the intensity of the toxaemia from which the patient has suffered, and we gather a slight idea of the probable sequelae.

Effects of the Vaccine on the temperature record.

In a large proportion of the cases, the immediate effect of the vaccine was not appreciable; in a few, as seen in charts 5 and 9, a slight rise occurred the same evening, and in case 32 the evening temperature rose to 103·6° F.

In case 12, chart 13, an injection was given on the fourth day of a relapse (33rd week), the wave was checked, the temperature fell rapidly and did not rise again.

In case 11, chart 12, the first injection was given on the third day of a rising wave in the 10th week of the fever; the wave stopped abruptly and there was no relapse.

In case 10, chart 11, after the fourth injection, a prolonged wave suddenly ended, and no relapse occurred.

Similar favourable influences on the temperature are seen in case 13, case 18, case 19, case 20, case 21 chart 14, case 22 chart 15, and cases 28, 31, 35, 36, and 37.

Out of these eleven last mentioned cases, ten were treated before or during the 13th week with the vaccine.

In cases 29, 30, 48, 49 the temperature at the time of giving the vaccine was low, and no subsequent rise took place.

It would appear that in mild undulatory relapses or with a chronic intermittent type of fever, the fall to normal is frequently accelerated, judging from previous experience in the treatment in so many cases.

In very acute cases with high fever no benefit could be observed either in the clinical symptoms or on the temperature curve. *Vide* case 1 chart 7, case 2, case 7 chart 10, and cases 14, 15, and 16.

The *prevention of relapses* is perhaps the greatest desire of any one treating this trying disease.

In the administration of vaccines we seemed to have a possible means of doing so. At the commencement of this investigation, some very successful cases led me to think that at last there was a reliable method. Vide charts 12, 13, 15. Further experience has proved this hope to be without foundation, for over and over again cases under treatment, when appearing to be doing favourably have suddenly developed another wave of pyrexia, not differing from those met with in cases otherwise treated. Vide charts 8 and 9.

Relation of the temperature curve to the opsonic curve.

In thirty of the forty-seven cases here charted which were treated with the vaccine, the opsonic index rose markedly in convalescence. In others who had not received any vaccine (see chart 6) or who had not had any for a prolonged period, the index ran high during severe pyrexial periods.

Agglutination Reaction.

Magnitude of agglutination reaction in cases of Mediterranean fever.

In the paper on this subject by Birt and Lamb, *Lancet*, Sept. 9th, 1899, complete reactions were found in 6000 fold dilutions of the serum. In the present investigation, the maximum obtained and highest tried for was 1 in 10,000, chart 7. In some chronic cases with much cachexia it falls as low as 1 in 10.

Course of the curve without Vaccines.

In the above mentioned paper by Birt and Lamb, it was demonstrated that as a rule with acute fever the agglutination reaction was at first high, falling rapidly before death, or remaining fairly constant till convalescence set in, when it fell. In cases running a chronic course though at first high, it became low and variable. A rapid fall from a high level was a bad sign, if accompanied by acute symptoms, and a consistently low agglutination curve made prognosis unfavourable.

In my observations on the blood reaction in Mediterranean fever previously referred to, the same general characters were demonstrated, but it was noticed that after the fall in convalescence, the agglutinins again rose considerably in the cases which remained free from relapses.

Further experience has shown more clearly than before that in prolonged cases the agglutination reaction may only be obtained in very low dilutions. This occurred recently in a patient at Haslar, when for some months before death 1 in 10 was the highest dilution giving a complete reaction, though on the day before he died the *Micrococcus melitensis* was isolated from his blood. (*Med. Fever Com. Report Royal Society*, part 4, p. 104.)

Duration of agglutination reaction.

Birt and Lamb found a complete reaction in a 1 in 50 dilution after five years. I have not found any to exceed this period, but have frequently obtained good reactions in a 1 in 40 dilution after three years.

Effect of the Vaccine on the agglutination curve.

Charts 4 and 5 show a rise in the agglutination curve after the injections of the vaccines, generally attaining its maximum about the third day. In chart 4 the rise was very great, from 1 in 1000 to 1 in 10,000. In chart 5 after the first injection it rose to 1 in 6000, and after the second to 1 in 8000, but after the third there was very little response.

Relation of the agglutination to the opsonic curve.

The relationship of the two curves seems to vary very considerably. At one time it was believed that the registration of the agglutination curve would give a correct indication of the amount of protective substances in the blood, a view put forward by Wright (1902), and since held by others.

It has since been shown by French (1906), that the amount of agglutinins are not indicative of the amount of opsonins present.

On the addition of the emulsion of the *Micrococcus melitensis* to the mixture of normal washed cells and the serum of patients suffering from Mediterranean fever, we have to reckon with the possibility of the specific agglutinins causing clumping of the micrococcus and rendering their ingestion in large numbers into the phagocytes easier than normal. In those sera which show high agglutination values, as is often the case in the early stages of the disease with high fever, one would expect that clumping would be very marked. That this is so is evident in some of the stained films showing high opsonic indices, for not only are the phagocytes full of organisms, but the latter are seen distinctly agglutinated throughout the film. From the study of a very large number of films it is however evident that this expected agglutination is *rarely present* sufficiently to explain the high phagocytic index of the blood under examination.

In chronic cases, after the injection of the vaccine, there was a slight rise in the agglutinins quickly falling to the usual low level.

To test the relationship of the two curves the blood was examined daily for both agglutinins and opsonins, the results being shown in charts 1 to 6.

A critical examination of these, as well as of all the accompanying charts of cases extending over many weeks, shows that there is no real relationship between the agglutination and the opsonic curve. Many of those showing evidence of low agglutinins having a high opsonic index. These observations are more in conformity with the experiments of Leishman (1905), than with those of some other observers.

Experiments relating to Opsonins in Mediterranean Fever, etc.

Comparison of the amount of sensitising substances in heated and unheated serum.

The serum was heated to a temperature of 60° C. for half-an-hour, then added to my washed blood cells, and finally to the emulsion.

		Average cocci ingested	Index
No. 1. *Acute Mediterranean fever case.*			
Unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	11	1·2
Heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	9	1·0
My unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	9	1·0

The heated serum of the patient still contained substances favourable to phagocytosis.

No. 2. *Chronic Mediterranean fever case.*			
Unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	7	1·2
Heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	2·5	·41
My unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	6	1·0
No. 3. *Chronic Mediterranean fever case.*			
Unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	10·2	·94
Heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	2·8	·16
My unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	10·8	1·0
No. 4. *Chronic Mediterranean fever case.*			
Unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	7·2	1·2
Heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	1·9	·33
My unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	5·7	1·0

Controls.		Average cocci ingested	Index
No. 5. *Healthy surgeon.*			
Unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	13·6	1·04
Heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	3·0	0·23
My unheated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	13·0	1·0
My heated serum	1 part		
My washed blood cells	1 part		
Emulsion	1 part	2·6	·2

In all these last the sensitising substance was almost destroyed by heat.

Experiment to ascertain whether the removal of the serum and replacing the content with normal saline solution would cause an equal diminution in the phagocytic action of the washed blood cells.

			Average cocci ingested	Index
No. 1.	My washed blood cells	1 part		
	Normal saline solution	1 part		
	Emulsion	1 part	5	·37
	My washed blood cells	1 part		
	My unheated serum	1 part		
	Emulsion	1 part	13·5	1·0
No. 2.	My washed blood cells	1 part		
	Normal saline solution	1 part		
	Emulsion	1 part	2·9	0·78
	My washed blood cells	1 part		
	My unheated serum	1 part		
	Emulsion	1 part	3·7	1·0
No. 3.	My washed blood cells	1 part		
	Normal saline solution	1 part		
	Emulsion	1 part	2·2	0·5
	My unheated serum	1 part		
	My washed blood cells	1 part		
	Emulsion	1 part	4·0	1·0
No. 4.	My washed blood cells	1 part		
	Normal saline solution	1 part		
	Emulsion	1 part	7·0	·7
	My washed blood cells	1 part		
	My unheated serum	1 part		
	Emulsion	1 part	10·0	1·0

These results indicate that moderate phagocytosis still occurs in the absence of serum.

Relative importance of the serum and of the leucocytes in phagocytosis.

The following experiment was carried out to show the relative phagocytic values of the serum and the blood cells of a patient suffering from chronic Mediterranean fever, and that of normal blood mixed in different ways.

		Average no. in each PMN cell	
		Case 1	Case 2
1.	Patient's unheated serum 1 part Patient's washed blood cells 1 part Emulsion of living *M. M.* ½ part	23	46
2.	Patient's unheated serum 1 part Normal washed blood cells 1 part Emulsion ½ part	8·6	25
3.	Normal unheated serum 1 part Patient's washed blood cells 1 part Emulsion 1 part	16	30
4.	Normal unheated serum 1 part Normal washed blood cells 1 part Emulsion ½ part	19	29

The same mixtures with dead cultures gave proportionally similar results.

From the above it would appear that the specific serum with its own washed blood cells was more powerfully phagocytic than when mixed with blood cells from another individual, and that the specific serum with blood cells from an individual not suffering from Mediterranean fever was less active than when the patient's blood cells were mixed with non-specific serum. This seems to indicate that the increased phagocytosis was due in part to an increased capacity of the patient's white blood cells to ingest the organisms of Mediterranean fever.

These experiments as a whole demonstrate that though sensitising thermolabile substances for the *Micrococcus melitensis* are found in the serum in varying quantities, yet when these have been removed by heat, or by replacing the serum with normal saline solution, phagocytosis still takes place to a considerable extent.

It is possible, as suggested by Metchnikoff in his Harben Lectures, that "the absorption of the microbes may be effected without the help of opsonins, or that, should such help be indispensable, the opsonins may be supplied by the leucocytes themselves."

The course of the opsonic curve in patients not treated by vaccine, or not having received any for long periods.

It is very difficult in such a disease as Mediterranean fever to estimate how much of the variation observed in the opsonic index is due to the addition of small artificially introduced doses of the toxic microbe, when at any time fresh natural periods of intoxication may occur. To illustrate these variations in the index, charts of the opsonic curves of two patients in very similar conditions of fever, cachexia, and periods of illness were carefully recorded day by day. One (chart 5) being treated with vaccines from the first, the second (chart 6) only having the vaccine after a considerable time. In the latter, we found a slightly raised and irregular index, followed by a rather high curve preceding a fresh wave of pyrexia. When the vaccine was given there was a further rise in the opsonic index and a fall in the temperature.

Chart 8 shows also a high opsonic curve with a severe wave of fever.

Effect of the Vaccine on the opsonic curve.

Charts 1 to 6 show from daily observations the opsonic curves in Mediterranean fever cases (the remainder were estimated weekly).

Chart No. 1. After the first dose of the vaccine, there was a slight negative phase followed by a rapid rise, this positive phase lasting over a week. When it reached the normal line, a second dose was given, which produced no negative phase and only a slight positive one.

Chart No. 2. The first injection of vaccine was given at the end of the third month of the fever, in a very anaemic and cachectic case.

After the first injection there was a short well marked negative phase, and a slight irregular positive one. With a falling opsonic curve and a rising temperature a second dose failed to elicit any response (the case was severe and ran a long course).

Chart No. 3. These observations were taken after the fourth injection on the 55th day of the disease, the temperature being slightly irregular. The opsonins from a low level rose steadily reaching a maximum on the third day, falling to the normal line on the sixth.

Chart No. 4. The first dose of the vaccine was given in the 10th week, with an irregular temperature, the opsonic curve rose rapidly but was very irregular. A second rather larger dose was followed by a more marked response, and a fall in the temperature.

Chart No. 5. Vaccines were commenced about the end of the third month of the fever, which was of an undulant type. Three injections were given, the response after the first and third being very marked.

Chart No. 6. This case was admitted also about the end of the third month of the disease, and was used for a control case to show the variations in the opsonic index without the influence of vaccines, quoted before.

In the long charts the general effect of the vaccines is seen to be a constant raising of the opsonic index.

Presence of a negative phase.

Col. Leishman in his paper on anti-typhoid inoculations, *Journ. Hygiene*, Vol. v. 1905, is sceptical as regards typhoid fever of the well marked negative phase described by Wright. A study of these charts shows that this phase is frequently absent, or very short in cases of Mediterranean fever, especially when the initial reading was below normal, the rise being steady from the time of giving the vaccine.

Relief of pain.

In the more chronic cases, having the very common secondary nerve and arthritic pains so frequently met with, the vaccines appeared to produce at least temporary improvement in some instances.

Gain in weight.

The gain in weight which occurred in 50 per cent. of his cases is regarded by Staff-surgeon Reid as the most certain indication of the favourable action of the vaccine. In some of my cases it was remarkable, half to one pound a day; at the same time too much value must not be placed on this rapid gain in the body weight, as it has frequently been observed in cases otherwise treated, when convalescence first sets in.

Looking over the total series of cases, the points standing out most clearly are that:

1. The injection of the vaccine, if carried out with care, is productive of little or no local or general disturbance to the patient.

2. The injection of the vaccine in most instances produced a more or less steady rise in the phagocytic power of the blood, as registered by the opsonic index.

3. The earlier the vaccine is commenced the more favourable the result.

4. Though the vaccine is ineffective when given in high fever or with severe clinical symptoms, and is unable with certainty to prevent relapses, yet in many cases the fever undulations were curtailed or reduced in severity after its administration.

In a disease like Mediterranean fever with a fatality of only 4 per cent. it is very difficult to provide adequate controls to check these results. The severity of an epidemic varies much from year to year, and even during each part of the year, so that comparisons are unsatisfactory.

I have however compared 60 cases treated with the vaccines in the early half of this year with a similar number from the same period last year, with the following result:

	Average duration in Haslar Hospital
60 cases 1905, without vaccines	67 days
60 cases 1906, with vaccines	75 days

Among the latter were some very severe cases, ten being over 100 days in the hospital, and one 245 days; some of these were months in the ward before the vaccines were commenced.

In the first series fourteen were over 100 days in the hospital.

Conclusions.

The vaccine treatment of Mediterranean fever appears in a certain number of cases to produce a beneficial effect, the severity of the symptoms being diminished, the general condition improved, and the duration of the disease curtailed.

The disease being characterised by its irregular course, long latent periods and unexpected recrudescences, it is one in which the estimation of the value of any method of treatment is most difficult. Too great weight therefore cannot be put on the above impression.

In the more acute type of case with high fever and evidence of severe intoxication, the method appears to have a deleterious, instead of a favourable action.

REFERENCES.

BASSETT-SMITH (1902). "The agglutinating properties of the blood in cases of Mediterranean fever, and other changes and reactions found in the course of the disease." *Brit. Med. Journ.* Vol. II. p. 861.

BASSETT-SMITH (1906). "Critical examination of blood of patients in hospital to determine if other than Mediterranean fever sera would agglutinate the *Micrococcus melitensis*." *Mediterranean Fever Commission Report to the Royal Society, London*, Part IV. 1906.

BIRT and LAMB (1899). "Malta fever, with special reference to specific agglutinating substances found in the blood." *Lancet*, Vol. II. p. 701.

BULLOCK (1905). "On the principles underlying the treatment of bacterial diseases by the inoculation of the corresponding vaccine." *Practitioner*, November.

DEAN (1905). "An experimental inquiry into the nature of the substance in serum which influences phagocytosis." *Proc. Royal Society*, Vol. B. 76, p. 506.

FRENCH (1906). "On opsonic factors, their estimation and uses in therapeutics." *Practitioner*, July.

HAGGARD and MORLAND (1905). "The action of yeast on tuberculosis and its influence on the opsonic index." *Lancet*, Vol. I. p. 1493.

LEISHMAN (1905). "Experiments in connection with stimulins." *Trans. Pathol. Soc. London*, July.

LEISHMAN (1905). "Anti-typhoid inoculations." *Journ. Hygiene*, Vol. V. p. 380.

METCHNIKOFF (1906). Harben Lecture, 1906. "The hygiene of the tissues." *Journ. R. Inst. Pub. Health*, June.

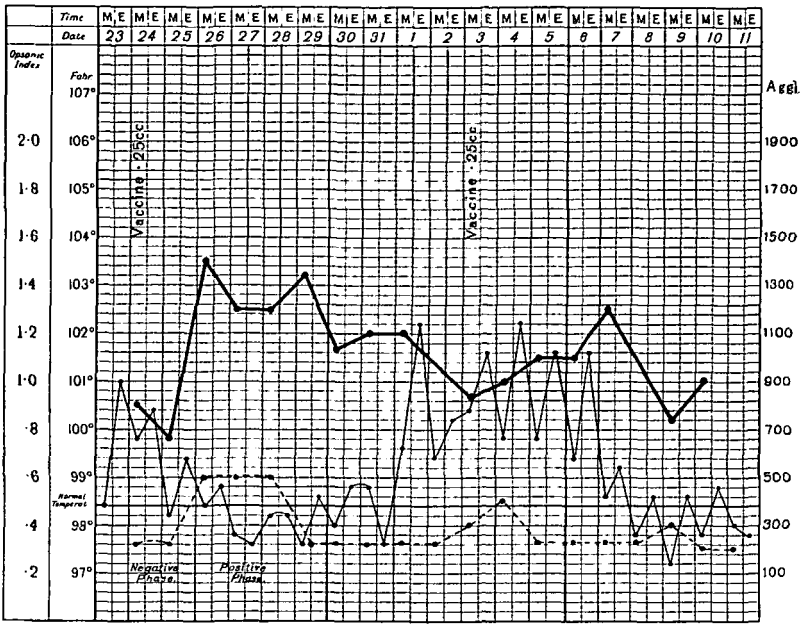
REID (1906). "Malta fever inoculations." *Trans. Pathol. Soc. Lond.* Vol. LVII. p. 463.

WRIGHT (1902). "Notes on the treatment of furunculosis, etc., by the inoculation of vaccines; and generally on the treatment of localised bacterial infections by therapeutic inoculations of corresponding bacterial vaccines." *Lancet*, Vol. I. p. 874.

WRIGHT and DOUGLAS (1903). "An experimental investigation of the rôle of the blood fluids in connection with phagocytosis." *Proc. Royal Soc. London*, Vol. LXXII. p. 357.

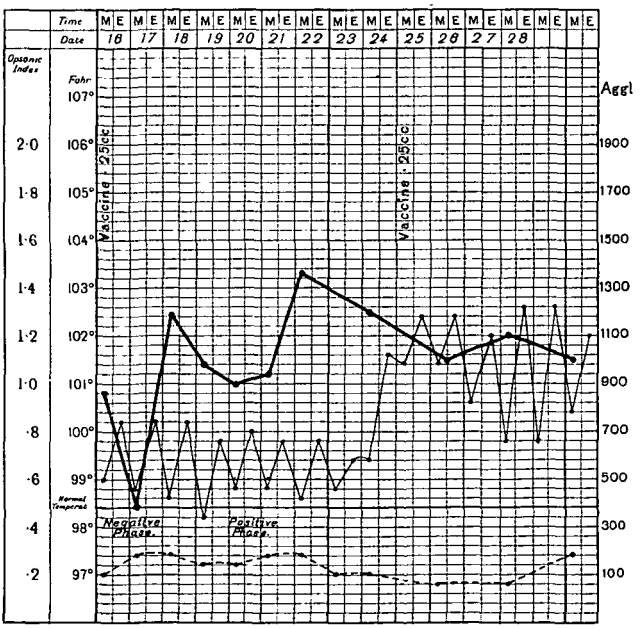
WRIGHT and DOUGLAS (1904). "Further observations on the rôle of the blood in connection with phagocytosis." *Proc. Royal Soc. London*, Vol. LXXIII. p. 128.

WRIGHT and DOUGLAS (1905). "On the general principles of the therapeutic inoculations of bacterial vaccines." *Lancet*, Vol. II. pp. 1598 and 1674.



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

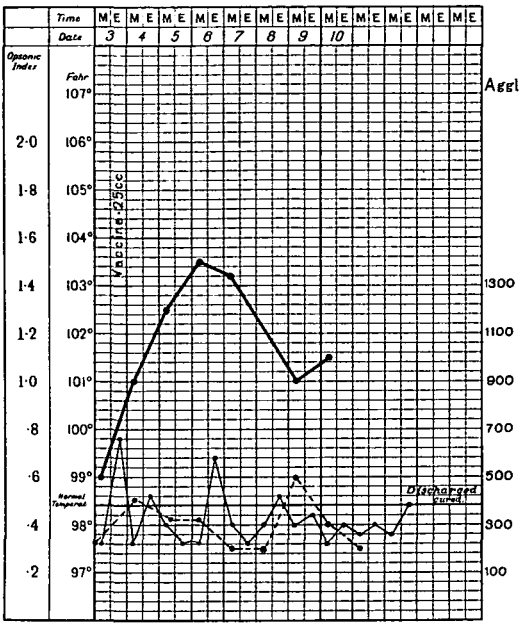
Chart No. 1. Case No. 38. 1st injection of vaccine in 11th week of the fever.



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

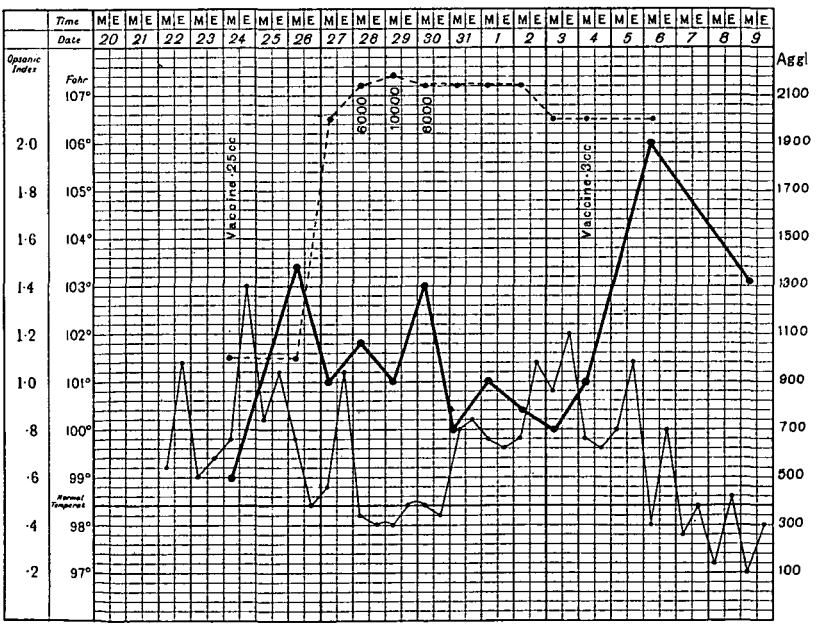
Chart No. 2. Case No. 40. Vaccine commenced end of 3rd month of fever.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

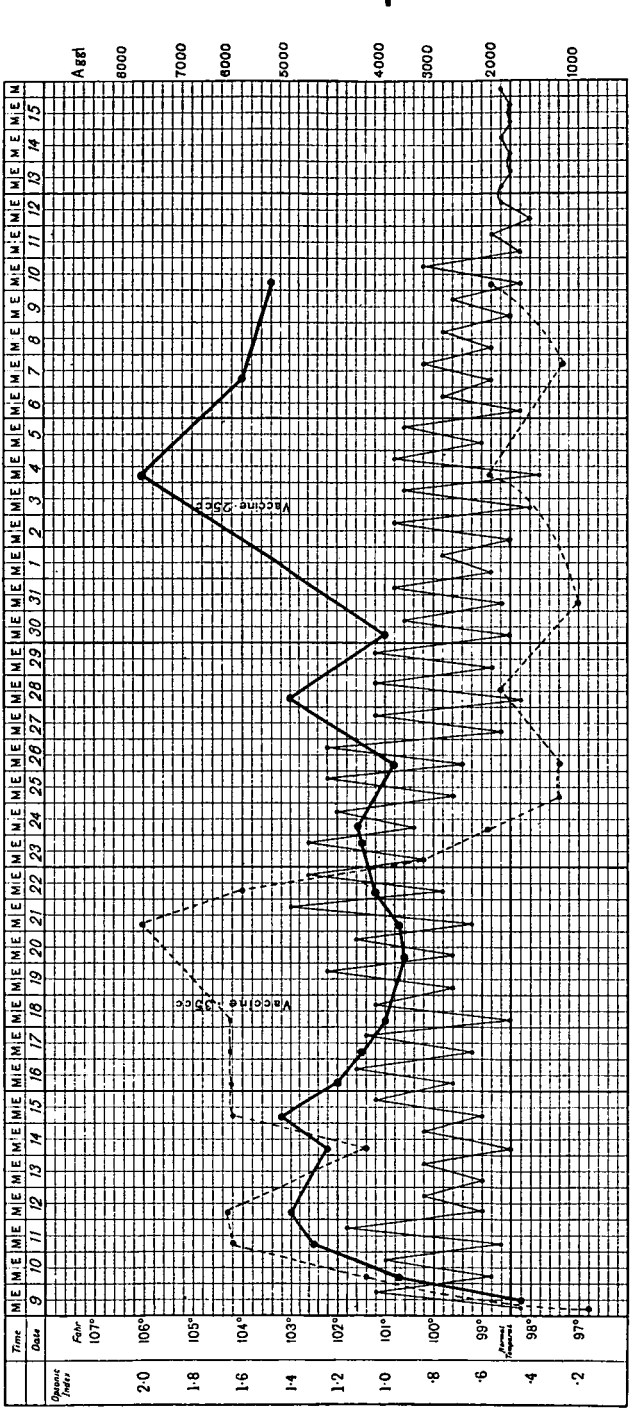
Chart No. 3. Case No. 41. 4th injection of vaccine 8th week of the disease.



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

Chart No. 4. Case No. 43. Vaccine commenced in 10th week of fever.

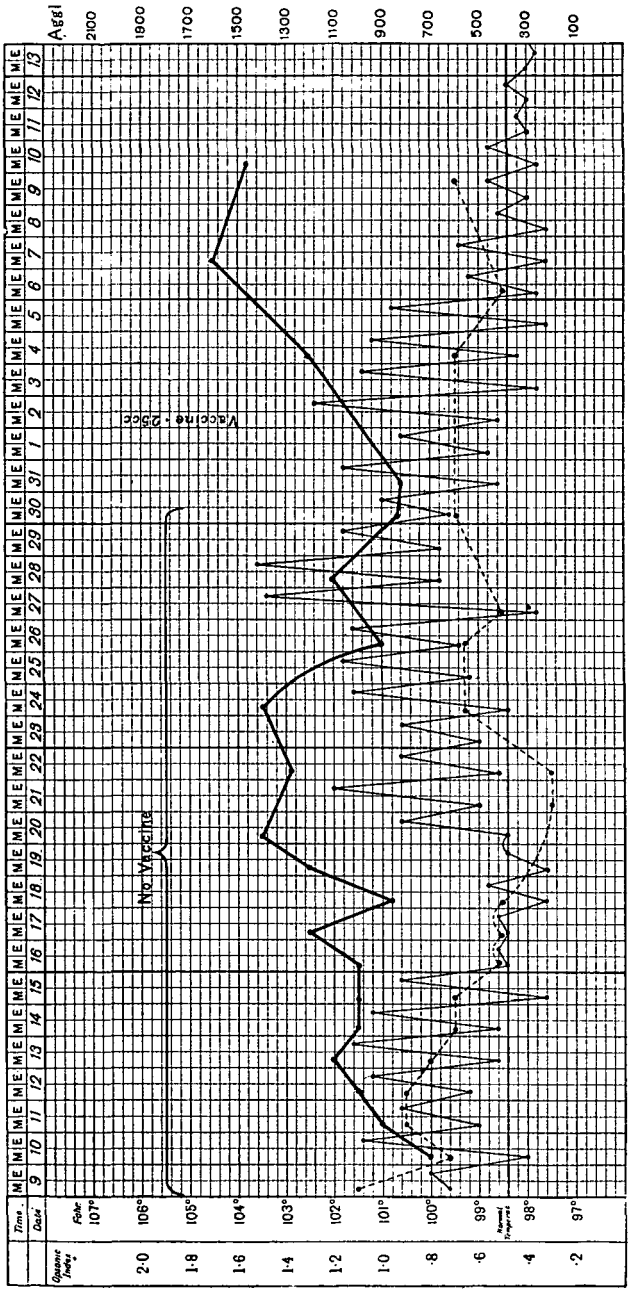
https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve. Dotted line = Agglutinin curve.

Chart No. 5. Case No. 44. Vaccine commenced middle of 3rd month of fever. N.B. Agglutinin curve is on a different scale to other charts.

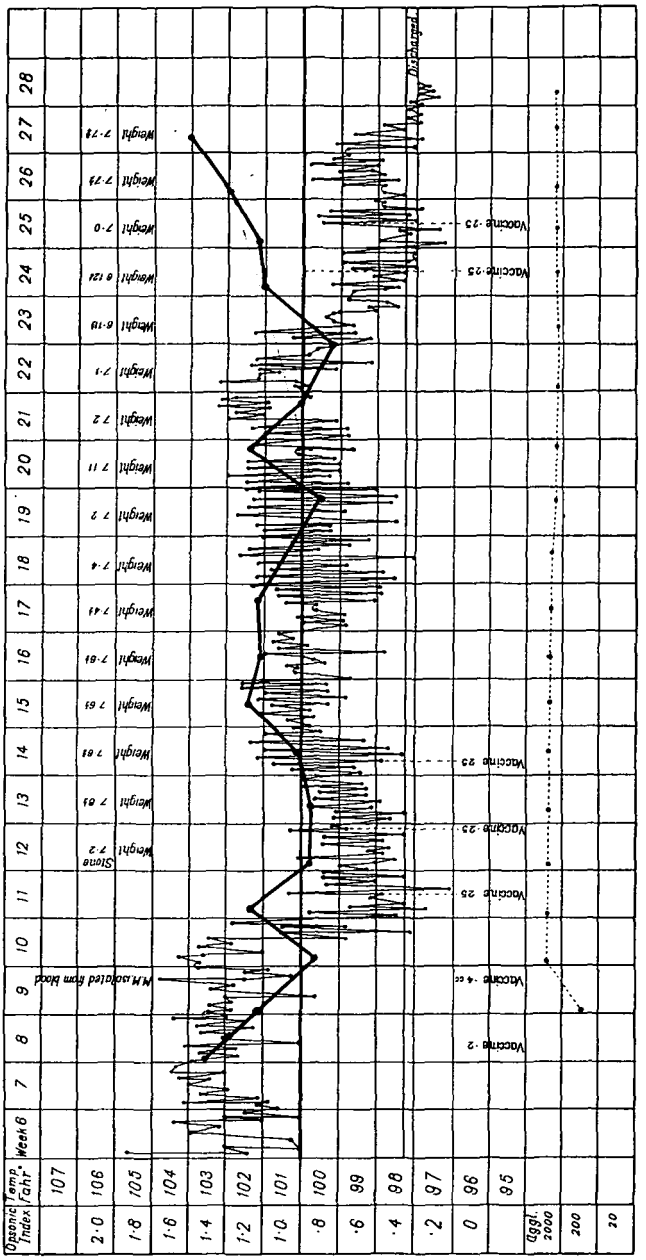
https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve. Dotted line = Agglutinin curve.

Chart No. 6. Case No. 45. Vaccine commenced 4th month of fever.

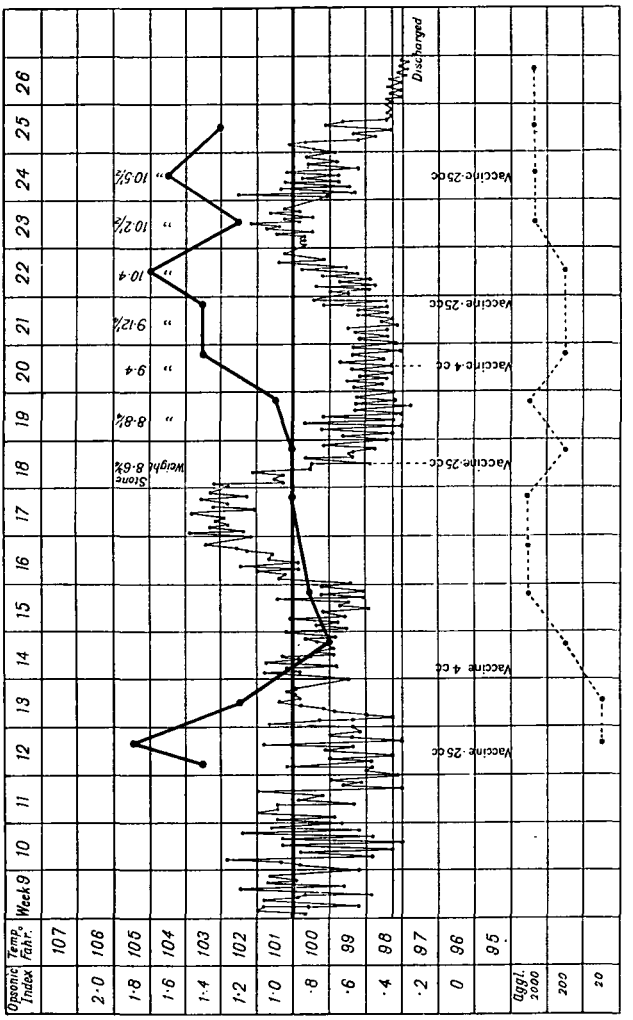
https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve. Dotted line = Agglutinin curve.

Chart No. 7. Case No. 1. W. E. G., 24, Stoker. Onset 3 March 1906. Discharged 14 September 1906. Invalided for deafness.

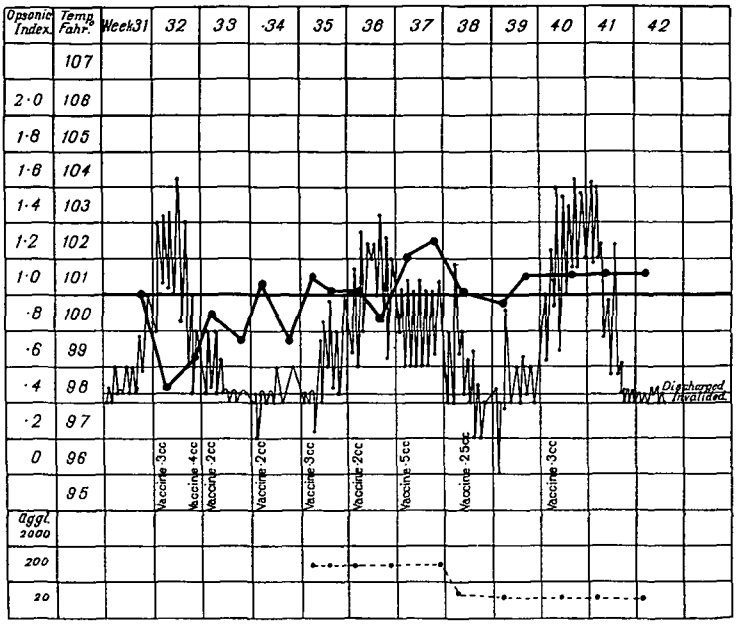
https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve. Dotted line = Agglutinin curve.

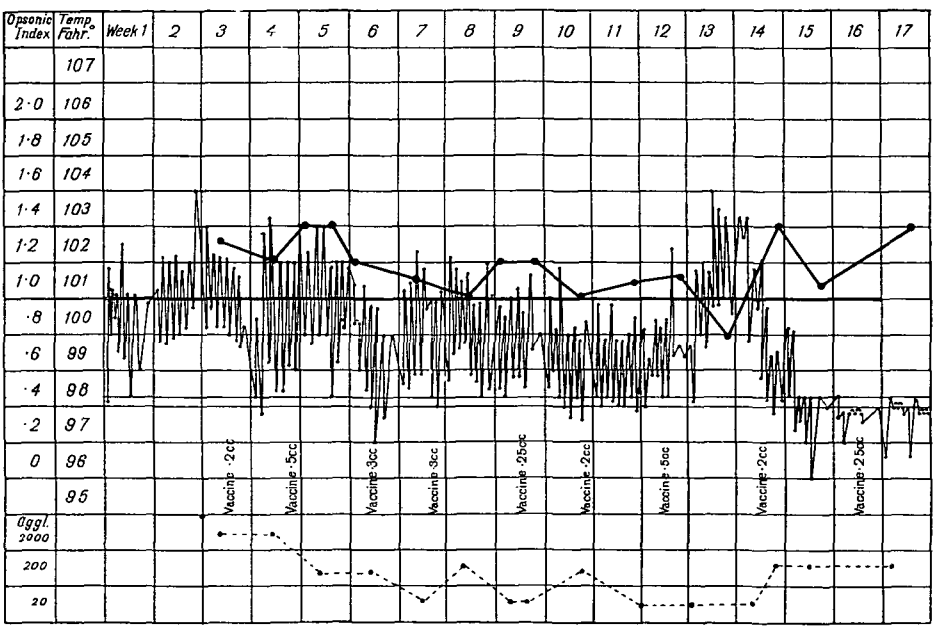
Chart No. 8. Case No. 3. V. H., 38, Ch. Cook. Onset 3 February 1906. Discharged 11 August 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

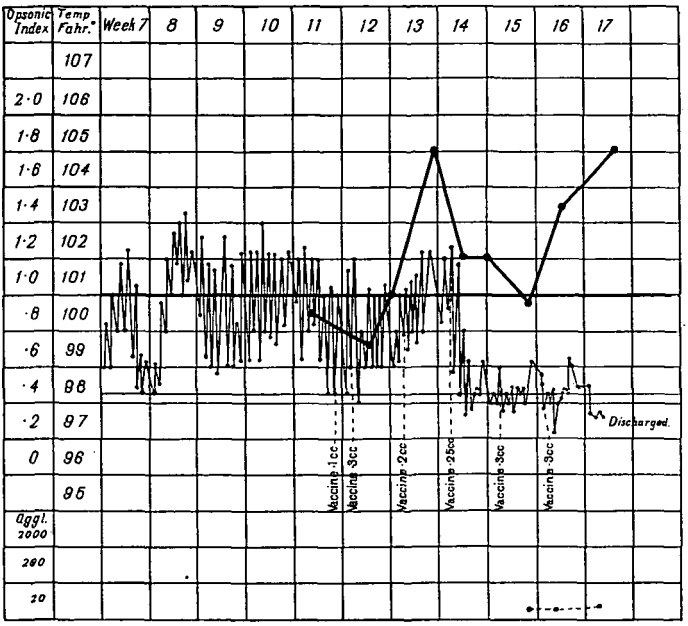
Chart No. 9. Case No. 5. C. N., 17, Boy. Leviathan. Onset 5 July 1905.
Discharged 17 April 1906. Invalided.



Thin line = Temperature curve. Thick line = Opsonic curve. Dotted line = Agglutinin curve.

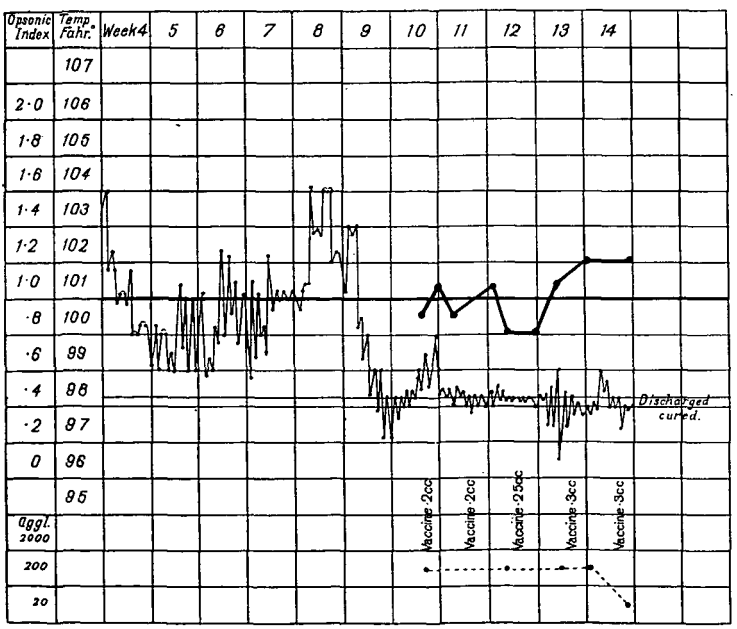
Chart No. 10. Case No. 7. P. A., 23, Pte R.M.L.I., Egmont. Onset 28 November 1905.
Discharged cured 12 June 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

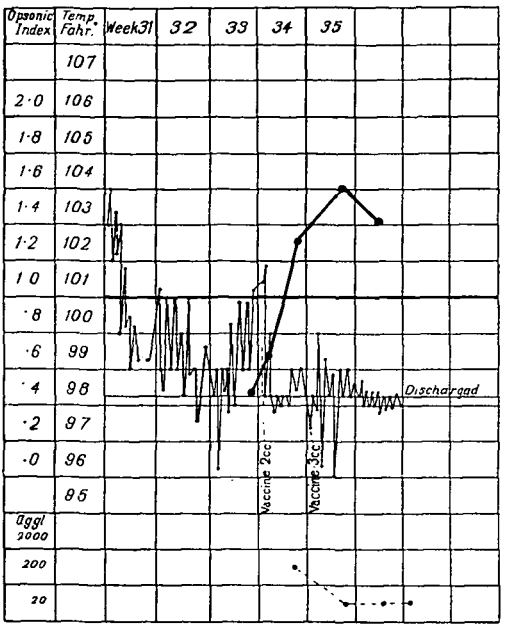
Chart No. 11. Case No. 10. J. W., 28, 2nd S.B.S., Malta Hospital.
Onset 14 November 1905. Discharged 11 March 1906.



Thin line = Temperature curve. Thick line = Opsonic curve
Dotted line = Agglutinin curve.

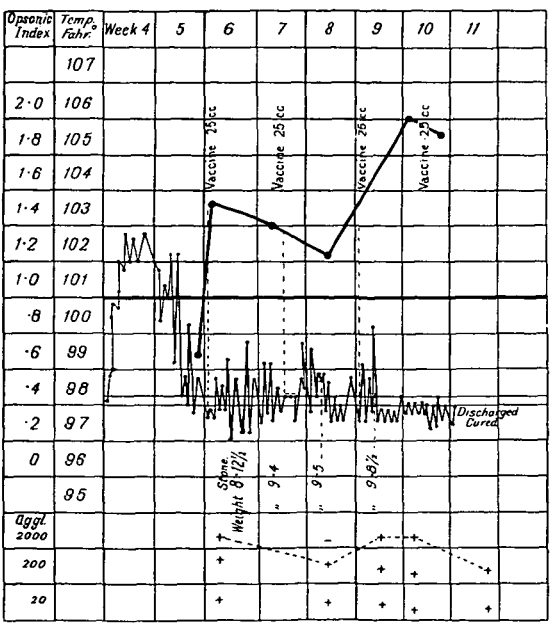
Chart No. 12. Case No. 11. T. L., 23, G.M.A. Onset 30 November 1905.
Discharged 14 March 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

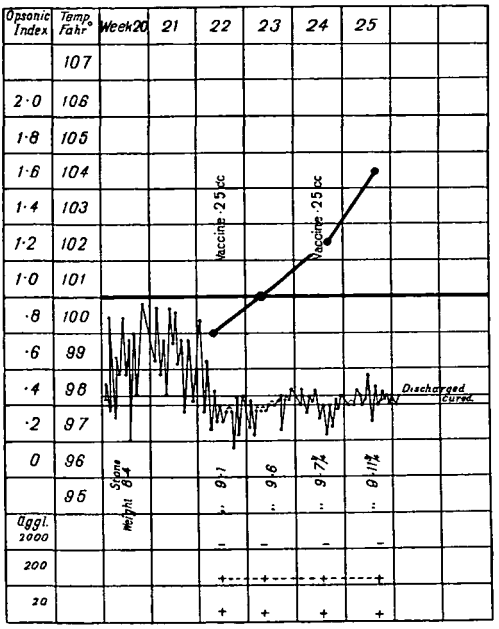
Chart No. 13. Case No. 12. J. M., 25, Sto. Vernon. Onset 29 May 1905. 31. 1. 06. Discharged 12 March 1906.



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

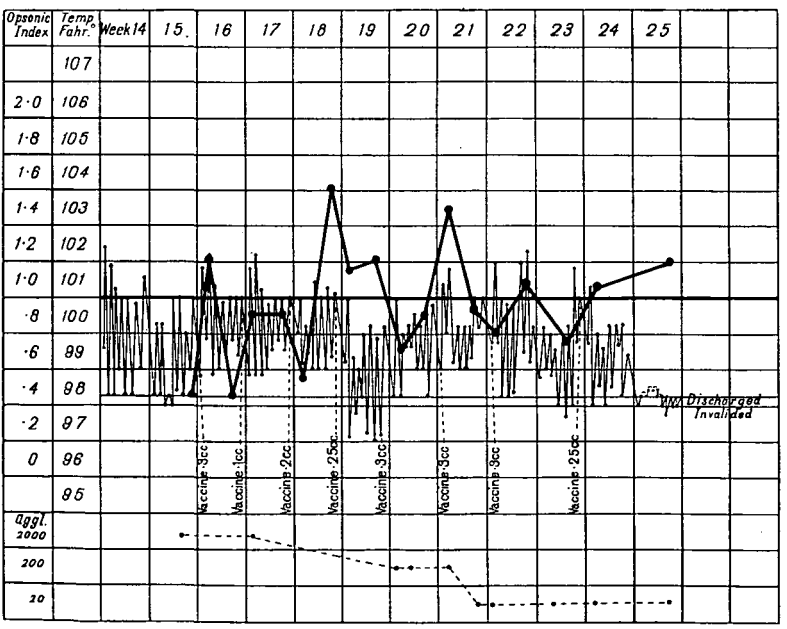
Chart No. 14. Case No. 21. B.C., 28, Stoker. Onset 13 April 1906. Discharged 23 June 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

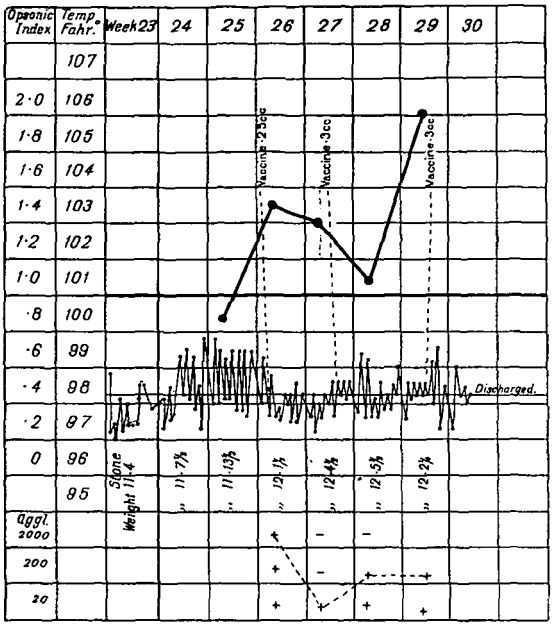
Chart No. 15. Case No. 22. T. B., 25, Bandsman. Onset 19 December 1905. Discharged 7 June 1906. Invalided.



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

Chart No. 16. Case No. 46. H. H. A.B., Vulcan. Onset 7 October 1905. Discharged invalided 9 April 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press



Thin line = Temperature curve. Thick line = Opsonic curve.
Dotted line = Agglutinin curve.

Chart No. 17. Case No. 47. G. G., 25, Leading Seaman. Onset December 1905. Discharged July 1906.

https://doi.org/10.1017/S0022172400033179 Published online by Cambridge University Press