THE DIAZO REACTION

and

BACILLURIA

in

ENTERIC FEVER.

ALEX. McCALL, M.B. Ch.B.

ProQuest LLC.
789 East Eisenhower Parkway
P.O. Box 1346
Ann Arbor, MI 48106 – 1346

I N T R O D U C T I O N.

The investigations on which these observations are based were carried out in the City of Glasgow Fever Hospital, Belvidere, during a period extending from January 1908 to January 1909.

Although hitherto much attention has been given to the Ehrlich Diazo Reaction from the point of view of diagnosis, no detailed investigation has been carried out into the duration of this reaction in individual instances, nor are there at present any data bearing on the appearance of this reaction during relapse and complications. In addition little seems to be known of the significance of the reaction.

The particular objects of the present investigation were:-

1. To consider its value as a symptom in the diagnosis of enteric fever.
2. To collate the relationship of the presence of the Diazo Reaction with the clinical course of the disease.
3. To examine as to whether the Diazo Reaction was related to typhoid bacilluria.

It was thought that this last might be possible, if the diazo reaction was in any sense a guide to the presence of Bacteriaemia, as stated by Ghenken, that it also might be a guide to the presence of Bacteriuria, even although previous experiments in vitro, by Hewlett and Dawson, had failed to discover that the diazo reaction could be produced in the urine by cultivating the bacillus typhosus in that medium.

The above mentioned would obviously have great practical importance as the examination of a urine for the bacillus typhosus, even with the resources of a laboratory arranged for the purpose, requires at least 2 days observations while the presence or absence of

the/

the diazo reaction can be decided in a few minutes.

In view of the recent discoveries with regard to "typhoid carriers" a relationship between these two phenomena would be of the greatest importance if substantiated.

The urines were consequently examined chemically for the reaction and bacteriologically for the baccillus typhosus throughout the course of the case.

In all, over 3,000 positive examinations were made.

An account of the investigations is arranged in the following order:-

i. Method of Examination employed.

ii. Classification of Cases.

iii. Appearance and clinical course of the Diazo Reaction.

iv. Its value in Diagnosis.

v. Occurrence of Bacilluria.

vi. The relation between the Diazo Reaction and Bacilluria.

1. METHOD OF EXAMINATION EMPLOYED.

A. Method employed in the examination for the diazo-reaction.

The method described by the original author, with the modification described by Hewlett was closely adhered to and will be briefly described.

The following reagents are necessary in order to carry out this reaction.

a. A saturated solution of Sulphanilic Acid in a 5% watery solution of Hydrochloric Acid.

b. A 5% freshly prepared solution of nitrite of soda in water.

c. A solution of strong ammonia.

Immediately prior to the performance of the test, solutions a and b are mixed in the proportion of one part of the former to 40 of the latter.

A few Cubic Centimetres of this mixture are placed in a test tube, an equal quantity of urine added, and the whole thoroughly mixed. I cubic centimetre of strong ammonia solution is then allowed to run slowly on to the surface of the mixture. At the junction of the two liquids a ring varying in colour from deep crimson to a delicate rose pink is produced if the reaction is present. If the mixture be shaken up a pink froth forms on the surface.

This is the reaction as commonly described and apparently accepted by the most recent authors (Vide Osler) but an important addition to this description was made by Hewlett in 1897.

If a tube of urine which has shown a well marked diazo reaction be allowed to stand upright for 12 to 24 hours a deposit forms varying in colour from dark to light green. The significance of this deposit will be/

be discussed later.

The urines of 100 cases of enteric fever were examined for this reaction during the patients' residence in hospital. In most of these cases the observations extended over 6 to 8 weeks and in some instances for several months. Fresh specimens of urine were examined.

To avoid sources of error these specimens were regularly tested as well for blood, albumin, and sugar, and in addition, as phenol and morphine give when present in the urine a similar reaction to that commonly called the diazo reaction, the urine was tested for the presence of these in any case in which the administration of Salol or Morphine was part of the treatment. None of the latter however, were found to be present in the urine of the cases under observation in sufficient amount to react to the ordinary test.

B. METHOD OF EXAMINATION EMPLOYED FOR THE BACILLUS TYPHOSUS.

The method adopted for the separation of the bacillus typhosus was the same as that used in the Glasgow Public Health Laboratory and is described under the headings -

1. Preparation of Media.
2. Method of examination for the presence of the organism.

1. Preparation of Media:-

a. Taurocholate Agar.

The basis of this medium was a solution made up according to the following:-

Taurocholate of Soda 5 grms.
Saturated Solution of Peptone in water 2 c.c's
Distilled water 100 c.c's.

About 1½% to 2% of agar was dissolved in this solution in the autoclave cleared with white of egg and filtered. After filtration 1% of lactose was added and 5% of a 1% solution of neutral red. This was sterilized at a temperature of 100°F. in the autoclave without pressure for 10 minutes on two successive days and then transferred to Petri's capsules.

b. Taurocholate Liquid Media.

These were prepared by adding respectively 5% of glucose and 1% of lactose, dulcite, mannite, saccharose and .5% of a 1% solution of neutral red, to the solution described above. These media were sterilized as described above in the previous heading.

Test tubes containing fermentation tubes were used to contain these media.

c. Broth.

This was prepared according to the following formula.

Lemco .5 grms.
Chloride of Sodium .5 grams.
Peptone 1 grm.
Water 100 c.c's.

These ingredients were dissolved in warm water and boiled for half an hour. The solution was then carefully neutralised with a 10% solution of caustic soda, litmus being used as the indicator. This solution was cleared with white of egg, filtered and sterilized in the autoclave under pressure, for half an hour on 3 consecutive days.

2. Method adopted in the examination of the urine.

The specimen of urine to be examined was centrifugalised, the supernatant fluid withdrawn by pipette and the deposit investigated by spreading a loopful of it on a taurocholate neutral red, lactose agar plate and incubating at a temperature of 37°C.

On this medium the colonies of the bacillus coli communis after 20 hours incubation are large raised, circular, opalescent and possess a red centre. Those of the bacillus typhosus on the other hand, after a like period of incubation though raised, are much smaller and appear colourless to reflected light but by transmitted light are blue. To complete the differentiation of these organisms the media containing the different sugars were employed.

The bacillus Coli Communis produces acid and gas in all the media.

This is indicated by the deepening of the red colour and the displacement of the liquid in the fermentation tube. The bacillus typhosus produces acid in glucose, and mannite and no gas in any of the media. A suspension of the organism to be examined was made in the broth above described and from this a series of tubes containing the different media inoculated.

These were incubated at a temperature of 37°C for a period of 20 hours, examined, and the nature of the fermentation recorded. For the purpose of verification a culture was made at the same time in broth which was

examined for the serum reaction after incubation for a period of 18 hours.

Standard selected sera from an undoubted case of enteric fever were used in dilutions of 1 in 50, the time allowed for agglutination to complete itself, being one hour. Such sera were tested at intervals against the laboratory strain of Bacillus Typhosus.

ii. CLASSIFICATION OF CASES:-

The cases are divided into three groups:- Severe - Medium - and Mild while those with relapse and complications have been considered more specially.

The first group, the Severe, includes those cases in which the temperature was 104°F. or more and whose t emperature of remission varied about 103°F. These numbered 18 of whom 8 died from toxaemia.

The second group, the Medium, includes those cases in which the temperature ranged about 103°F as maximum with a remission in some cases to about 100°F. These cases numbered 58 of whom only 4 died, all from perforation.

In the third group, the mild, the maximum temperature range varied from 101° to 102°F. They number 23.

TABLE I.

Table showing number of cases of enteric fever examined, for the first time, on each day of illness.

Day of Earliest observation of the Diazo Reaction.	Diazo Reaction. Positive	Diazo Reaction. Negative.	No. of cases showing Green Deposit on first examination.	Total No. of Cases.
3rd. Day of Illness.	2.	–	2.	2.
4th. " " "	5.	–	5.	5.
5th. " " "	1.	–	1.	1.
6th. " " "	4.	–	4.	4.
7th. " " "	6.	–	3.	6.
8th. " " "	13.	–	9.	13.
9th. " " "	11.	–	7.	11.
10th. " " "	3.	–	3.	3.
11th. " " "	7.	–	6.	7.
12th. " " "	4.	–	4.	4.
13th. " " "	5.	–	3.	5.
14th. " " "	3.	–	1.	3.
15th. " " "	5.	–	3.	5.
16th. " " "	2.	–	1.	2.
17th. " " "	4.	–	1.	4.
18th. " " "	2.	–	1.	2.
21st. " " "	13.	–	6.	13.
24th. " " "	1.	–	1.	1.
28th. " " "	5.	–	2.	5.
35th. " " "	3.	–	–	3.
?	–	1.	–	1.
	99.	1.	63.	100.

iii. APPEARANCE AND CLINICAL COURSE OF THE DIAZO REACTION.

a. Date of onset of the Diazo Reaction.

Apart from the type of the disease there is one point which may be taken as common to all cases of enteric fever, viz:- the date of illness at which the reaction is first observed.

There is no general statement so far as I have been able to find as to when the reaction appears. The earliest date at which it is recorded is the 5th day of illness (Dawson) but even this author does not state that in any case of undoubted enteric fever he has found it absent when he first examined for its presence. In all the cases except one as shown in Table I. the reaction was obtained from the earliest date at which the patients were seen, though on account of family infection with the disease, some of these cases were examined as early as the 3rd and 4th days of illness. In two cases it was present on the 3rd day of illness and in 5 on the 4th day. These cases were subsequently proved to be typical cases of enteric fever by their clinical course and were verified by a positive serum reaction. It would appear from this that the diazo reaction is definitely present during the early stages of the disease.

It is unfortunate that the number of cases coming under observation at this stage of the disease was so small but if experience bears out that the reaction is regularly present at this early date and present as in my cases with unmistakable distinctness, it should prove of great assistance in the early diagnosis of the disease.

As the reaction has always been present at the earliest date of examination and as some of the cases examined in the very early days belong to each of the

TABLE II.

Day on which the Diazo Reaction disappeared in 89 cases.

Number of Cases. Day of Illness.	13.	14.	15.	16.	17.	18.	19.	20.	21.	22.	23.	24.	25.	26.	27.	28.	29.	30.	32.	33.	34.	36.	39.	40.	42.	TOTALS.
Mild.	-	3.	3.	1.	1.	1.	2.	1.	-	3.	-	-	3. D											1.	1.	20.
Medium.	- D	- 2D	1. D	1. D	4.	5. D	3.	2.	3.	5. D	4.	5.	2.	2.	3.	4.	2.	1.	2.	D 1.	D 2.	-	D 1.	-	2.	55.
Severe.	1.	2.	1.	1.	-	1.				2.						1.			1.	1.	1.	1.			1.	14.
	1.	5.	5.	3.	5.	7.	5.	3.	3.	10.	4.	5.	5.	2.	3.	5.	2.	1.	3.	2.	3.	1.	1.	1.	4.	89.

D = Death.

3 groups, it is obvious that no comparison can be drawn between the early appearance of the reaction and the type of the disease.

b. Duration of the Diazo Reaction.

The duration of the reaction has a more definite relation to the type of the disease, than the date of its appearance. The days of illness on which the urine was last examined with positive result, are given in the annexed Table (Table II).

The shortest duration of the reaction observed among cases extended in the mild group to the 14th day of illness, in the medium to the 15th, and in the severe to the 28th day. In each of the three groups the longest duration was the same viz:- 42 days.

In the mild group of cases, the reaction terminated during the first three weeks of the disease in 15 of 20 instances and in the medium 44 of 55 cases, show the reaction terminating within the first 28 days of illness. There are however, 4 cases belonging to the mild and 2 to the medium group, in which the duration of the reaction was very prolonged. But as these cases came under observation late in the disease, it is possible that they might have been classified differently, had they come under observation earlier.

In the severe group of cases, the duration of the reaction in cases ending in recovery, was always very prolonged. The mean duration in the mild cases was 21 days, medium, 23.4, and severe 31.4. If the outlying two cases in the medium and mild groups be omitted, the mean duration then becomes in the former group 22.8 days and in the latter 19.6.

From these figures it would appear that there is little difference in the duration of the reaction in the medium and mild groups but that the reaction is prolonged in the severe cases.

TABLE III.

Table showing day on which the Green Deposit ceased in 54 Cases.

Number of Cases.	Day on which Deposit ceased.	6.	7.	8.	9.	10.	11.	12.	13.	14.	15.	16.	17.	18.	19.	20.	22.	23.	24.	30.	31.	TOTALS.
	Mild.		1.																	1.		2.
	Medium.	1.	1.	1.	4.	2.	3.	3. D 1.	5. D 1.	1. D 3.	1. D 1.	1. D 3.	2.	2.	3.	D 1.	D 1.	3.	D 2.		D 1.	38.
	Severe.						1.	1.	1.	3.	1.	3.		1.	2.					1.		14.
		1.	2.	1.	4.	2.	4.	4.	6.	4.	2.	4.	2.	3.	5.	1.	1.	3.	2.	2.	1.	54.

D = Death.

The above remarks apply specially to the reaction originally described but it is in addition necessary to discuss the occurrence of the green deposit already referred to, as discovered by Hewlett. If a tube of urine in which the diazo reaction has been performed with positive result be allowed to stand upright for 12 to 24 hours a sediment ranging in colour from light to dark green precipitates frequently. This deposit, however, is in general only found during the earlier portion of the fever and does not persist through the whole period during which the Diazo Reaction is obtained. It has always been observed in those cases which came under observation early.

Out of the 99 cases (Table III) examined, the deposit was present in 64. Of these 10 are described later under the section dealing with complications. The remaining 54 cases are distributed as follows - 2 belong to the mild group. 38 to the medium, and 14 to the severe, that is to say that in the severe group the deposit was present in all the cases.

Of the 2 cases in which it was observed in the mild group in one it lasted till the 7th day and in the other to the 30th day of illness. From Table IV it can be seen that if present, it had disappeared in 3 cases before the 7th day and in 3 before the 9th so that it is possible that in these cases it may be frequently absent.

The shortest duration in the medium group, was 6 days and the longest 31. Even in this group it was absent in 4 cases first examined on the 8th day of illness and in one examined on the 9th day.

IN/

TABLE IV.

Table showing day on which the Green Deposit was absent from the first examination.

Number of Cases. Day of 1st. examination.	7.	8.	9.	11.	13.	14.	15.	16.	17.	18.	21.	28.	35.	TOTALS.
Mild.	2.		3.	1.		2.	2.		3.		3.		1.	18.
Medium.		4.	1.		1.			1.		1.	4.	3.	2.	17.
Severe.														
	3.	4.	4.	1.	1.	2.	2.	1.	3.	1.	7.	3.	3.	35.

In the severe group the shortest duration was 11 days and the longest 30 days.

It is specially to be noted that it was always present in these cases terminating fatally, in both medium and severe groups.

TABLE V.

SIMPLE RELAPSE CASES.

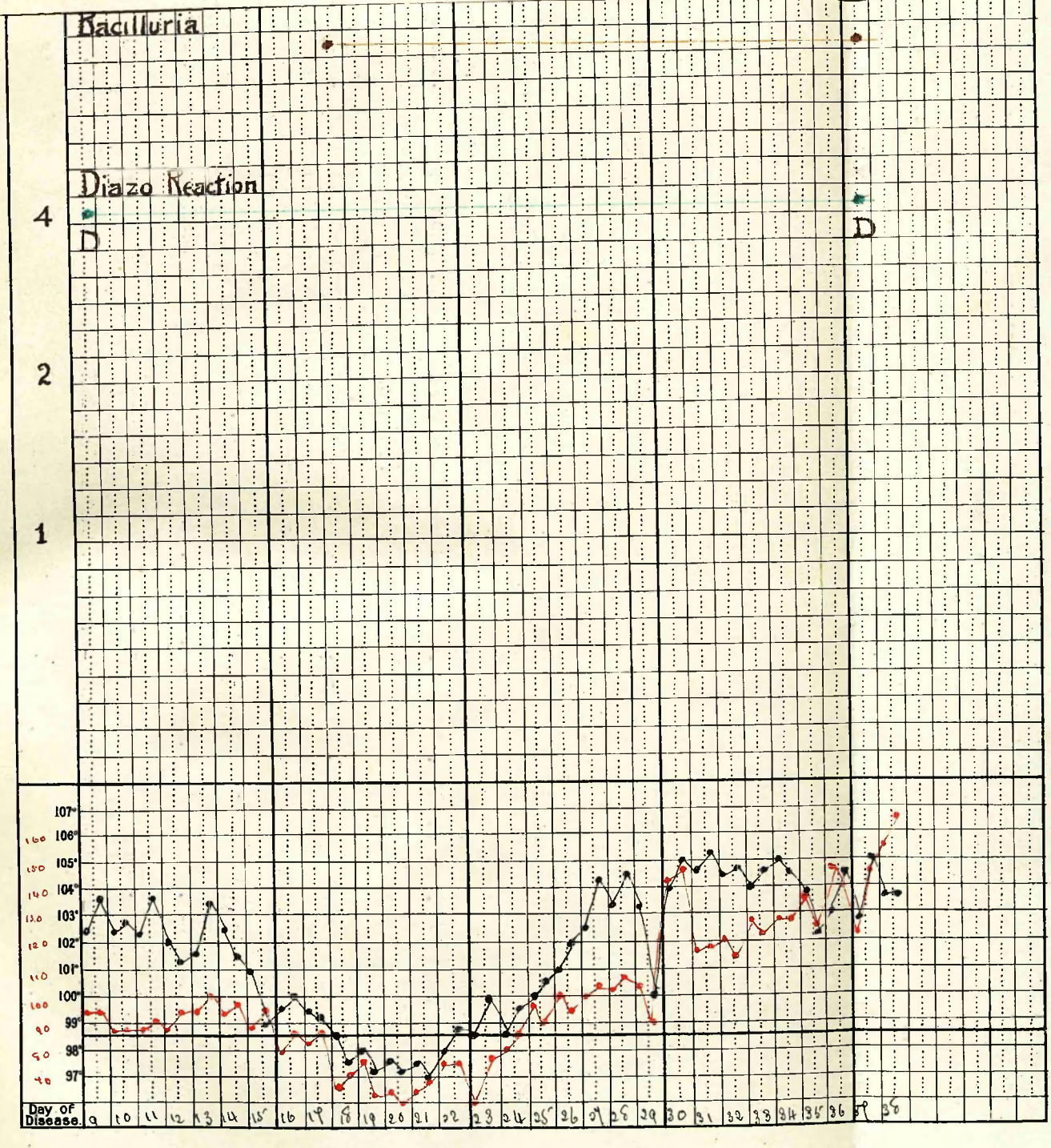
Table showing the period during which the Diazo Reaction and the sedimentation deposit were observed compared with the duration of the pyrexial periods of the fever.

Number of Cases.	Type of Disease.	Result.	Description of Pyrexial Period.	Pyrexial Period.	Days on which Diazo Reaction Was observed.	Days on which Deposit was observed.
1.	Severe.	Death.	Initial Attack.	9th. - 18th. days.	9th. - 37th. days.	9th. - 37th. days.
			Relapse.	23rd. - 38th. "		
2.	Medium.	Rec.	Initial Attack.	9th. - 17th. "	9th. - 19th. "	9th. - 13th. "
			1 st. Relapse.	31st. - 51st. "	30th. - 51st. "	30th. - 40th. "
			2nd. "	63rd. - 77th. "	58th. - 78th. "	58th. - 65th. "
3.	Medium.	Rec.	Initial Attack.	9th. - 21st. "	9th. - 21st. "	9th. - 12th. "
			Relapse.	38th. - 58th. "	38th. - 58th. "	38th. - 46th. "
4.	Mild.	Rec.	Initial Attack.	8th. - 16th. "	8th. - 16th. "	8th. - 10th. "
			Relapse.	19th. - 40th. "	20th. - 43rd. "	20th. - 28th. "

CHART Nº I

W.H. MALE AGE 24

For "METHOD ADOPTED IN THE CHARTING OF THE REACTION"
see page 23.



D - D = Duration of Deposit

c. Occurrence of the Diazo Reaction in these cases associated with Relapse or some other complication.

Owing to the great variability in the occurrence and duration of the Diazo Reaction in this group of cases, these are discussed separately. Ten cases in all are described, 4 with simple relapse, 3 with relapse complicated by furunculosis, one with relapse complicated by abscess formation, one case with simple furunculosis and one with pyuria.

1. Simple Relapse Cases.

Case 1. W. H., Male, age 24, (Chart 1)

This patient was admitted on the 9th day of illness suffering from a severe type of enteric fever. The temperature reached normal on the 18th day of illness but 5 days later on the 23rd day of illness a relapse began which caused death 15 days later, i.e. on the 38th day of illness.

The diazo reaction and the green deposit were both present ffom the 9th day till death.

Although there was an interval of 5 days between the two periods of pyrexia, the temperature remaining normal during this period., - yet the reaction was maintained without intermission and without any diminution in its intensity from the commencement till the end.

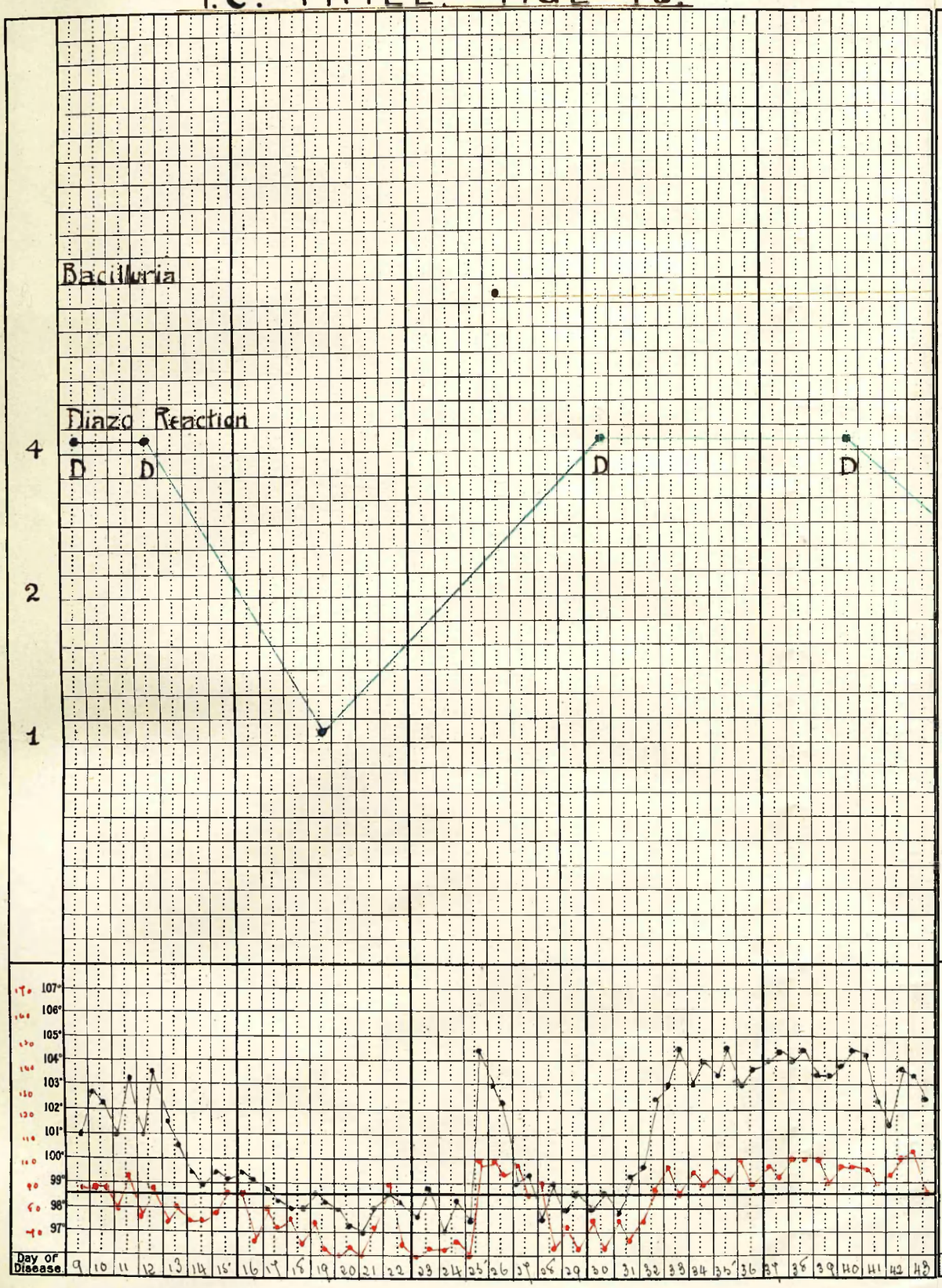
Case 2. T. C. male, age 18. (Chart 2)

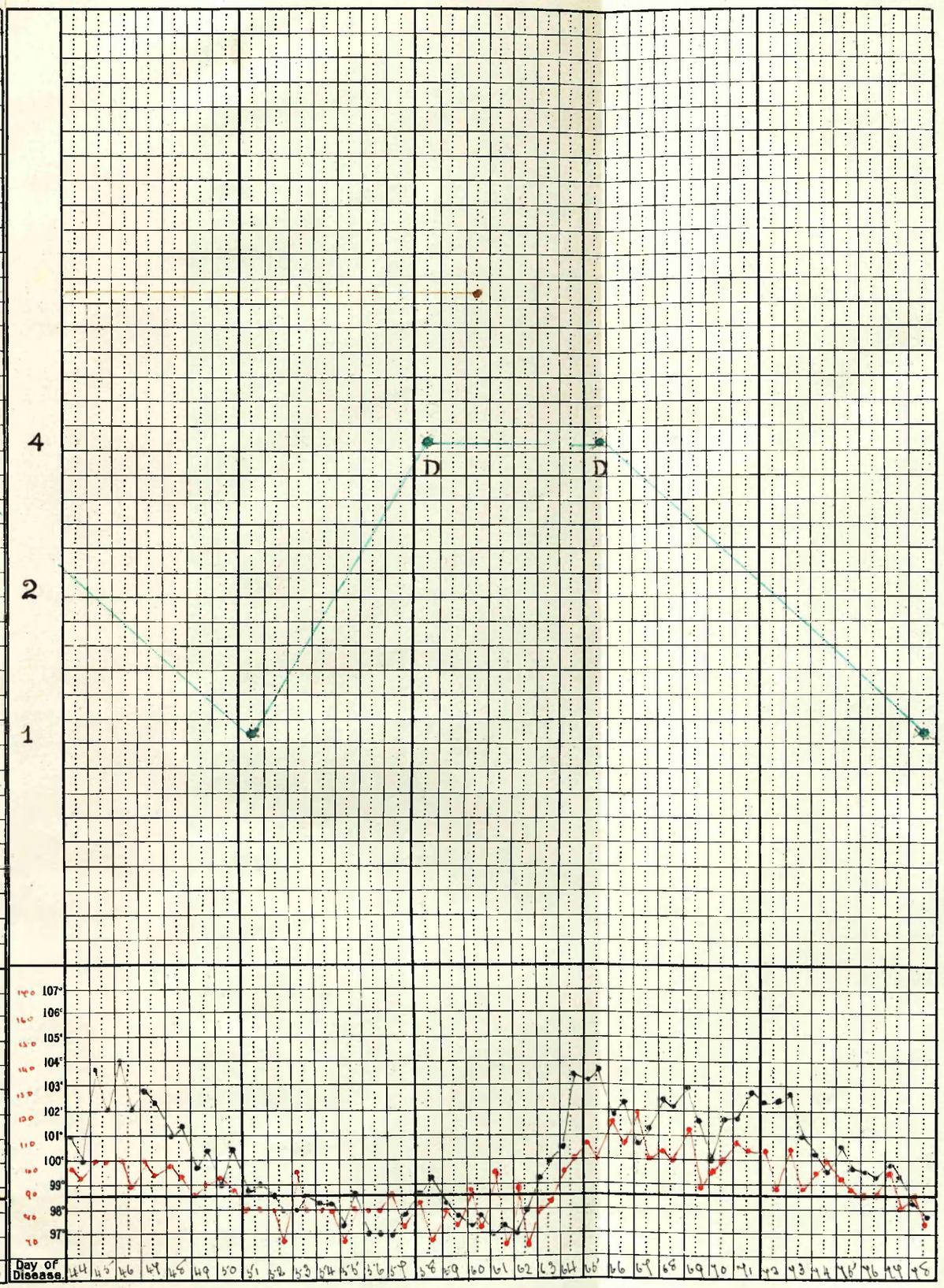
This patient was admitted in his 9th day of illness. The initial pyrexia terminated on the 17th day and from this point the temperature remained normal till the 31st day of illness (except for a period of 2 days when an elevation occurred the cause of which was not determined). A severe relapse began on this day and continued till the 51st day of illness. A second relapse began on the 63rd day and ended on the

78th/

CHART II

T.C. MALE. AGE 18.

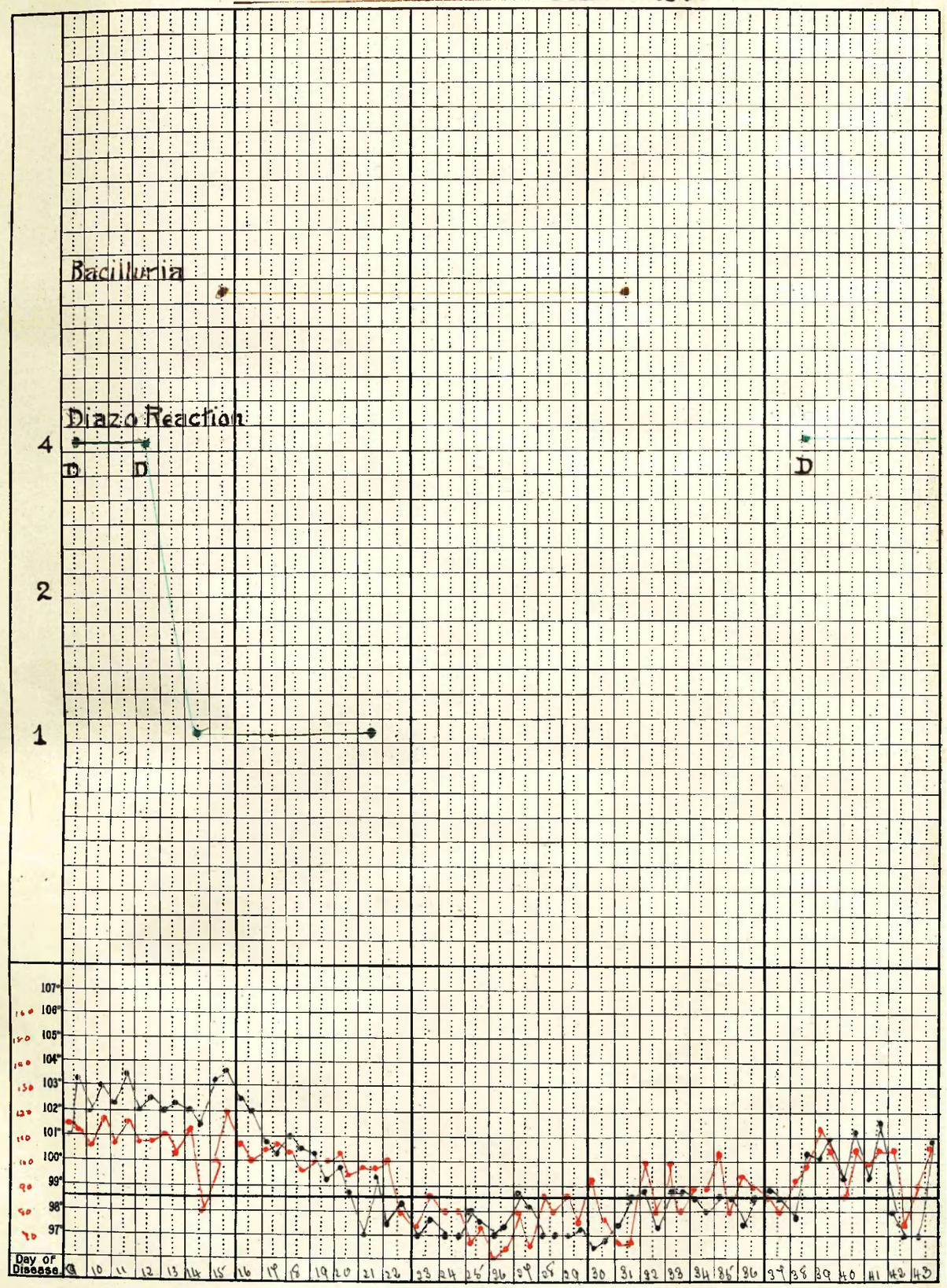


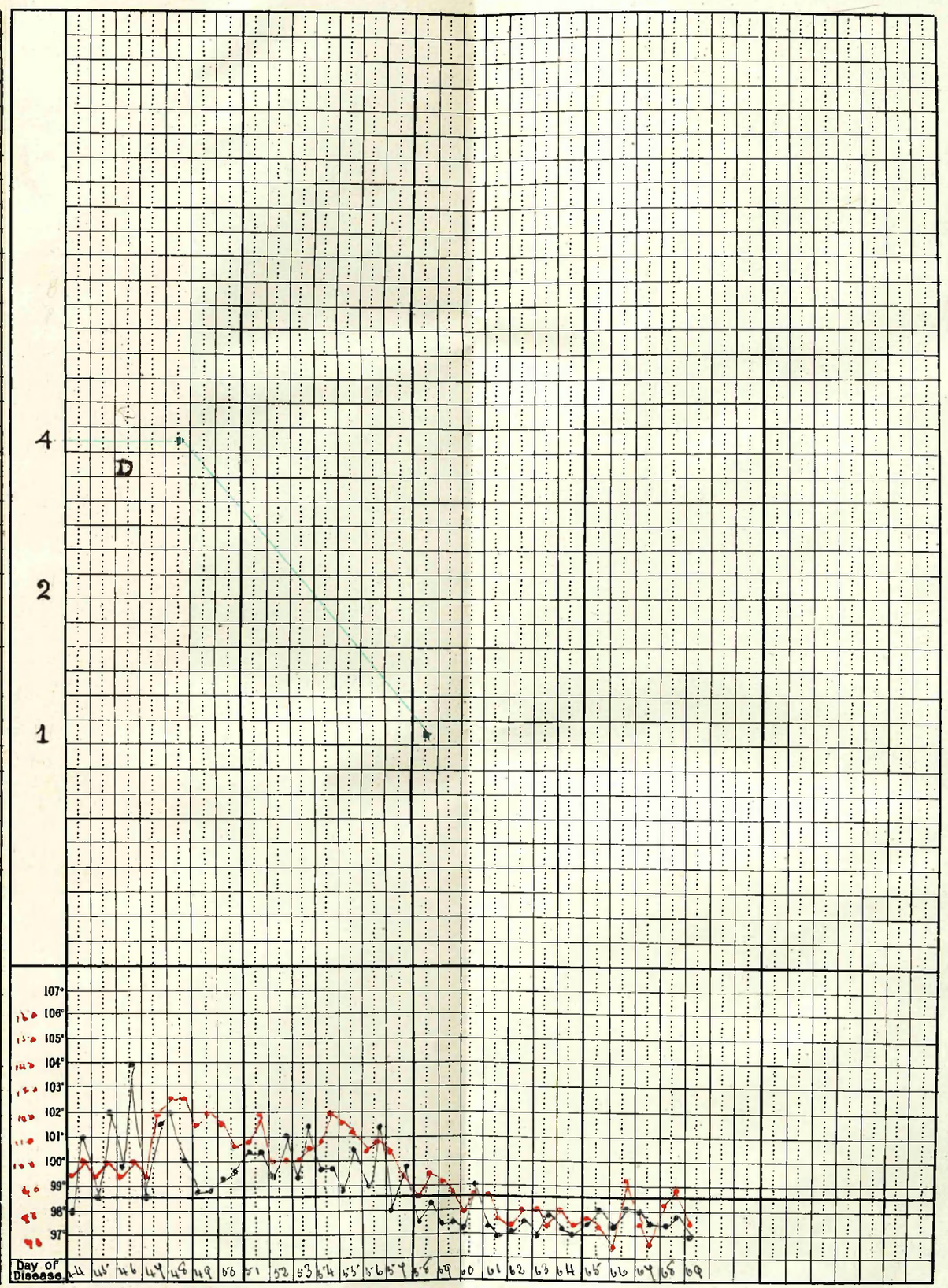


D-D = Duration of Deposit

CHART III

A. McG. MALE AGE 21





D.-D = Duration of Deposit

78th day of illness.

During the initial attack the diazo reaction lasted till the 19th day i.e. it outlasted the pyrexia by 2 days. The reaction returned on the 30th day or one day before the commencement of the relapse and persisted till the 51st day. It reappeared on the 58th day that is to say 5 days before the beginning of the 2nd relapse and terminated synchronously with it.

Case 3. A.McG. male. age 21. (Chart 3)

The patient was admitted on the 9th day of illness with an attack of Enteric Fever. The initial pyrexia lasted till the 21st day of illness and during this period the diazo reaction and green deposit were present, the former from the 9th to the 21st day of illness and the latter from the 9th to the 12th day of illness. A relapse began on the 38th day of illness lasting till the 58th and during this period the diazo reaction was present. The deposit during this attack lasted from the 38th to the 46th day of illness.

Convalescence was uninterrupted and patient was dismissed well.

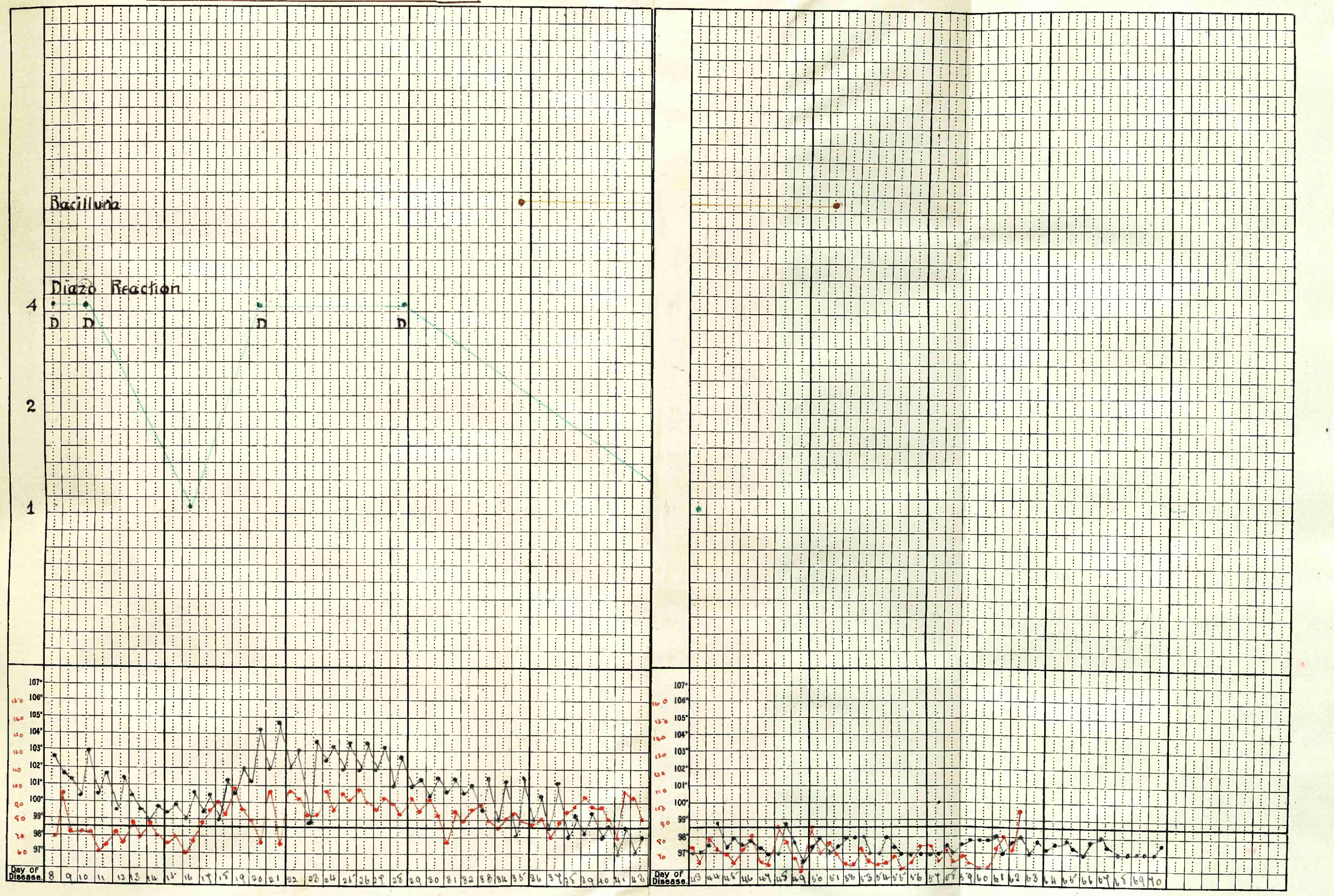
Case 4. P. W. male. age 14 (Chart 4)

This patient was admitted on the 8th day of illness, the initial pyrexia lasting till the 16th day of illness. During this period the diazo reaction and green deposit were present, the former throughout the period of fever and the latter from the 8th to the 10th day of illness. A relapse began on the 19th day of illness and lasted till the 40th day.

The diazo reaction appeared on the 20th day and lasted till the 43rd day, that is to say that it appeared one day after the rise in temperature and outlasted the period of pyrexia by 3 days. The deposit during this relapse lasted from the 20th day of illness to the/

CHART IV

P.W. MALE. AGE 14



D - D = Duration of Deposit

the 28th.

Convalescence was good and patient was dismissed well.

T A B L E VI.

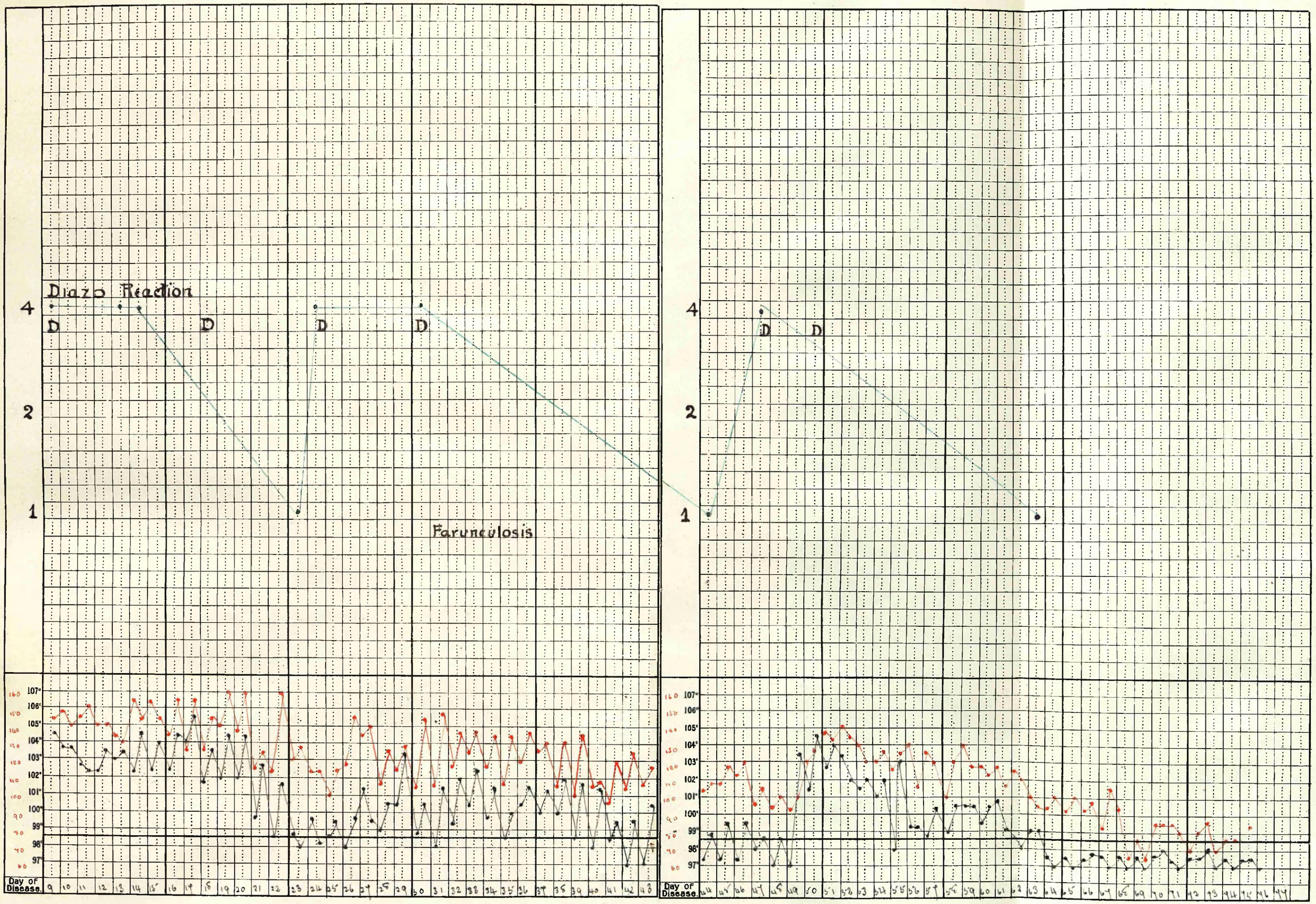
CASES WITH RELAPSE COMPLICATED WITH FARUNCULOSIS.

Table showing 3 cases in which farunculosis occurred, with the period during which the Diazo Reaction and Sedimentation Deposit were observed with the duration of the pyrexial periods of the fevers.

No. of Cases.	Type of Disease.	Result.	Description of Pyrexial Period.	Pyrexial Period.	Days on which the Diazo React. was observed.	Days on which the Sedim. Dep. was observed.
1.	Severe.	Rec.	Initial Attack.	9th. - 23rd. days.	9th. - 23rd. days.	9th. - 18th. days.
			Farunculosis.	24th. - 44th. "	24th. - 44th. "	24th. - 30th. "
			Relapse.	49th. - 62nd. "	47th. - 63rd. "	47th. - 50th. "
2.	Severe.	R.	Initial Attack.	11th. - 23rd. "	11th. - 23rd. "	11th. - 20th. "
			Relapse.	27th. - 47th. "	28th. - 47th. "	28th. - 36th. "
			Farunculosis.	49th. - 56th. "	49th. - 55th. "	49th. - 50th. "
			Renewed Farunculosis.	58th. - 61st. "	58th. - 61st. "	58th. - 59th. "
3.	Medium.	R.	Initial Attack.	7th. - 15th. "	7th. - 15th. "	7th. - 9th. "
			Relapse & Farunculosis.	18th. - 45th. "	19th. - 45th. "	19th. - 31st. "
			Mild Farunculosis.	49th. - 60th. "	49th. - 60th. "	49th. - 51st. "

CHART V

B.C. FEMALE AGE. 14.



D-D = Duration of Deposit.

Cases with Relapse complicated with Farunculosis.

Case 1. B.C. female. age 14. (Chart 5 page 16)

This patient was admitted on the 9th day of illness suffering from a severe attack of enteric fever, the initial pyrexia lasting till the 23rd day of illness. The temperature however, rose again on the 24th day of illness and continued elevated till the 44th day. This elevation of temperature was due to the occurrence of a severe attack of farunculosis. A second rise of temperature occurred on the 49th day of illness terminating on the 62nd day and due to a relapse of enteric fever. During the initial period the reaction was present while the temperature was elevated, the green deposit lasting till the 18th day of illness. Simultaneously with the commencement of farunculosis on the 24th day of illness the reaction recurred and terminated along with it on the 44th day. Two days before the commencement of the enteric relapse i.e. on the 47th day of illness the reaction was observed to be present and continued to be present till the 63rd day outlasting the relapse by one day.

Convalescence was good and the patient was dismissed well.

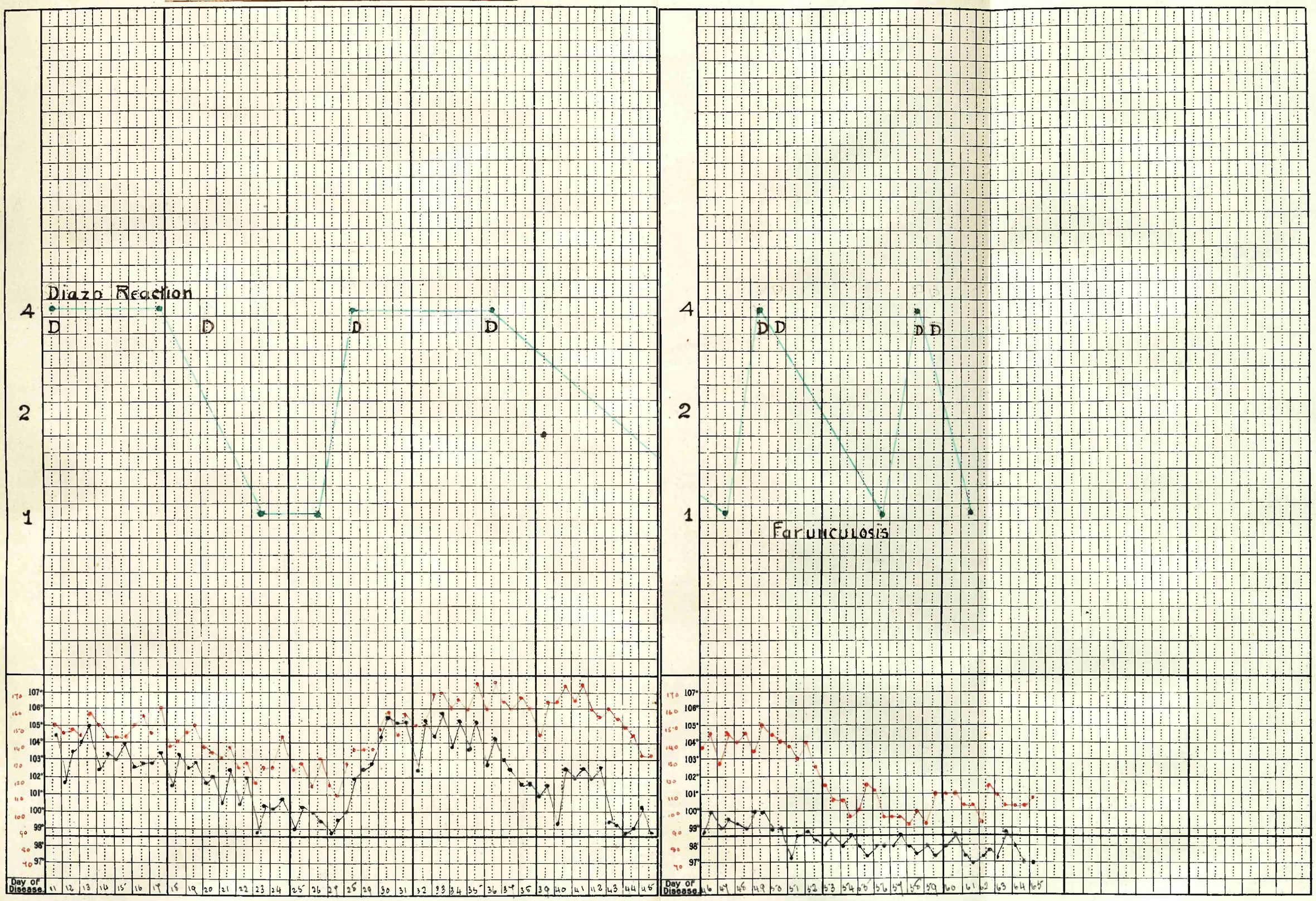
Case 2. R. B. female, age 10 (Chart 6 page 17)

This patient was admitted on the 11th day of illness suffering from a severe attack of enteric fever, the period of fever lasting till the 23rd day. The diazo reaction and the deposit were present during this period, the former throughout the whole period and the latter till the 20th day of illness. A relapse began on the 27th day of illness terminating on the 47th day.

The/

CHART VI

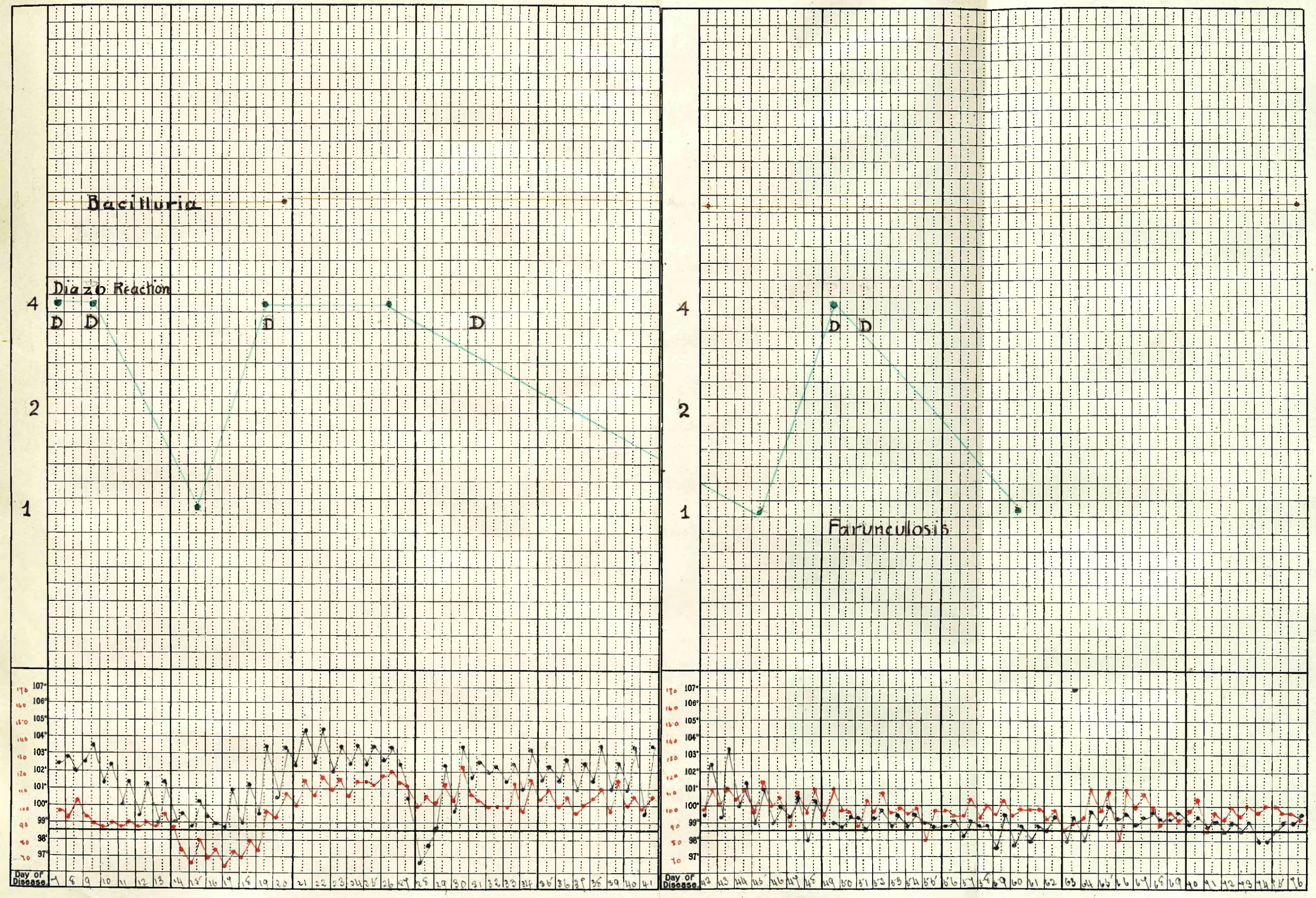
R.B. FEMALE AGE 10.



D-D = Duration of Deposit

CHART VII

J.S. MALE. AGE 14.



D-D = Duration of Deposit

The reaction reappeared at this period terminating withthe fever though the green deposit only persisted till the 36th day of illness.

An attack of farunculosis began on the 49th day of illness lasting till the 56th day and with this the reaction and the green deposit reappeared, the former lasting till the 55th day and the latter till the 50th day of illness. The reaction and the green deposit reappeared with a renewal of the farunculosis on the 58th day of illness (during which the temperature range was 98°F to 99°F.) and ceased to be present on the 61st day of illness. In this instance the green deposit lasted till the 59th day of illness. Convalescence was uninterrupted.

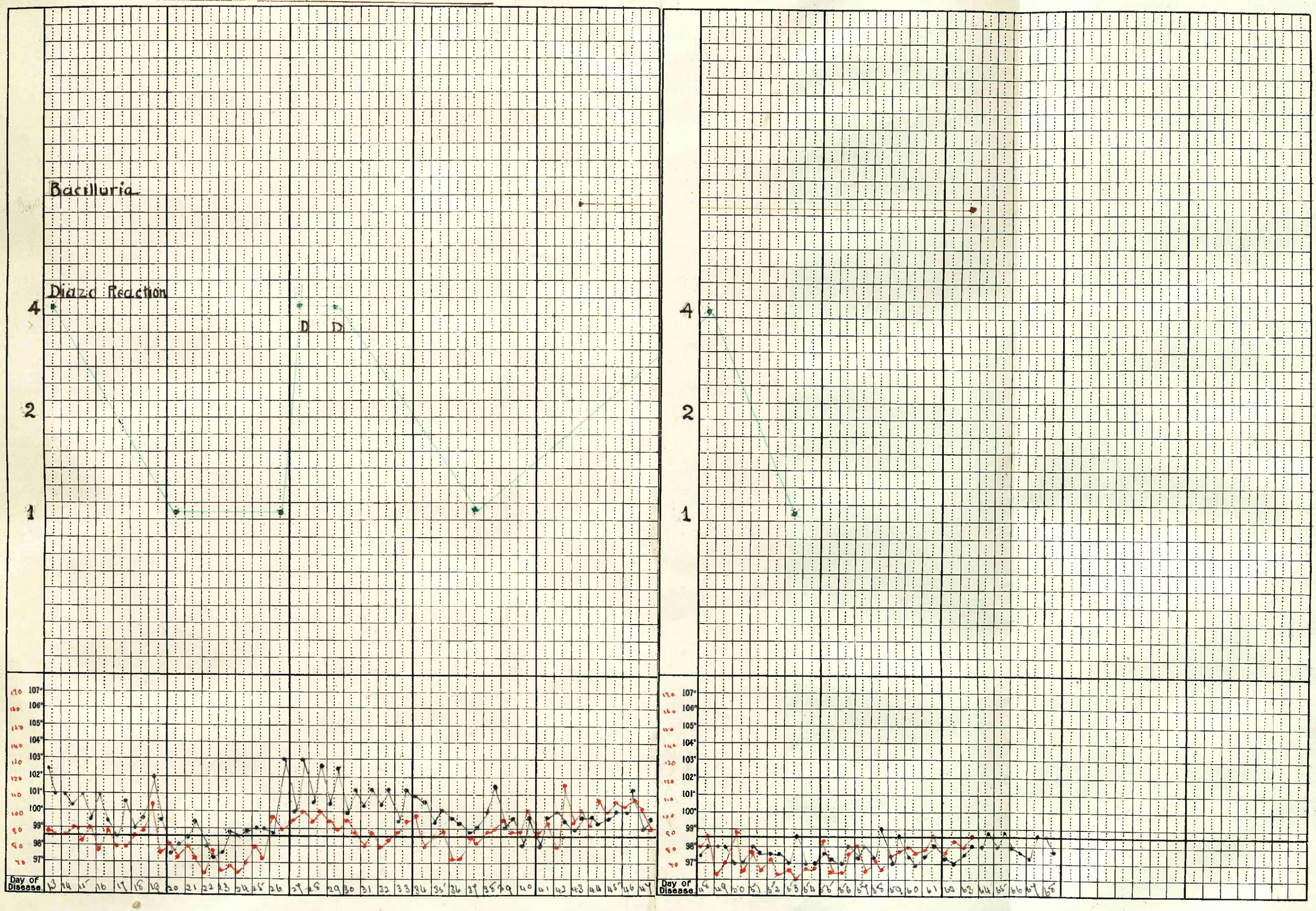
Case 3. J. S. male, age 14. (Chart 7 page 17)

In this case the duration of the pyrexia of the initial attack and of the reaction was the same both disappearing on the 15th day of illness. The sedimentation deposit was not however observed after the 9th day. A relapse complicated by farunculosis began on the 18th day of illness and lasted till the 45th day. In this instance the reaction and the green deposit commenced on the 19th day of illness, the former terminating on the 45th day and the latter on the 31st day of illness. The reaction reappeared on the 49th day of illness and lasted till the 60th due to a recurrence of the farunculosis. The green deposit during this exacerbation lasted till the 51st day of illness.

Convalescence was good and the patient was dismissed well.

CHART VIII

W.D. MALE AGE 43



D-D = Duration of Deposit

Case of Relapse complicated by Abscess formation.

W. D. male. age 43. (Chart 8 page 18)

This patient was admitted on the 13th day of illness suffering from a mild attack of enteric fever. The initial pyrexia lasted till the 20th day of illness and during this period the diazo reaction was well marked persisting to a lesser degree till the 27th day when the reaction again assumed a deep crimson colour which continued till the 37th day of illness. During this latter period there was also an elevation of the temperature due to a relapse commencing on the 26th day of illness and terminating on the 37th day. On this latter date the patient complained of pain in the ischio-rectal region without the reaction again appearing. On the 48th day of illness, however, a crimson reaction was observed and at the same time it became evident that an abscess was forming in the region above mentioned. This was incised and drained on the 51st day of illness after which the reaction became less marked finally disappearing on the 53rd day of illness.

Convalescence was uninterrupted.

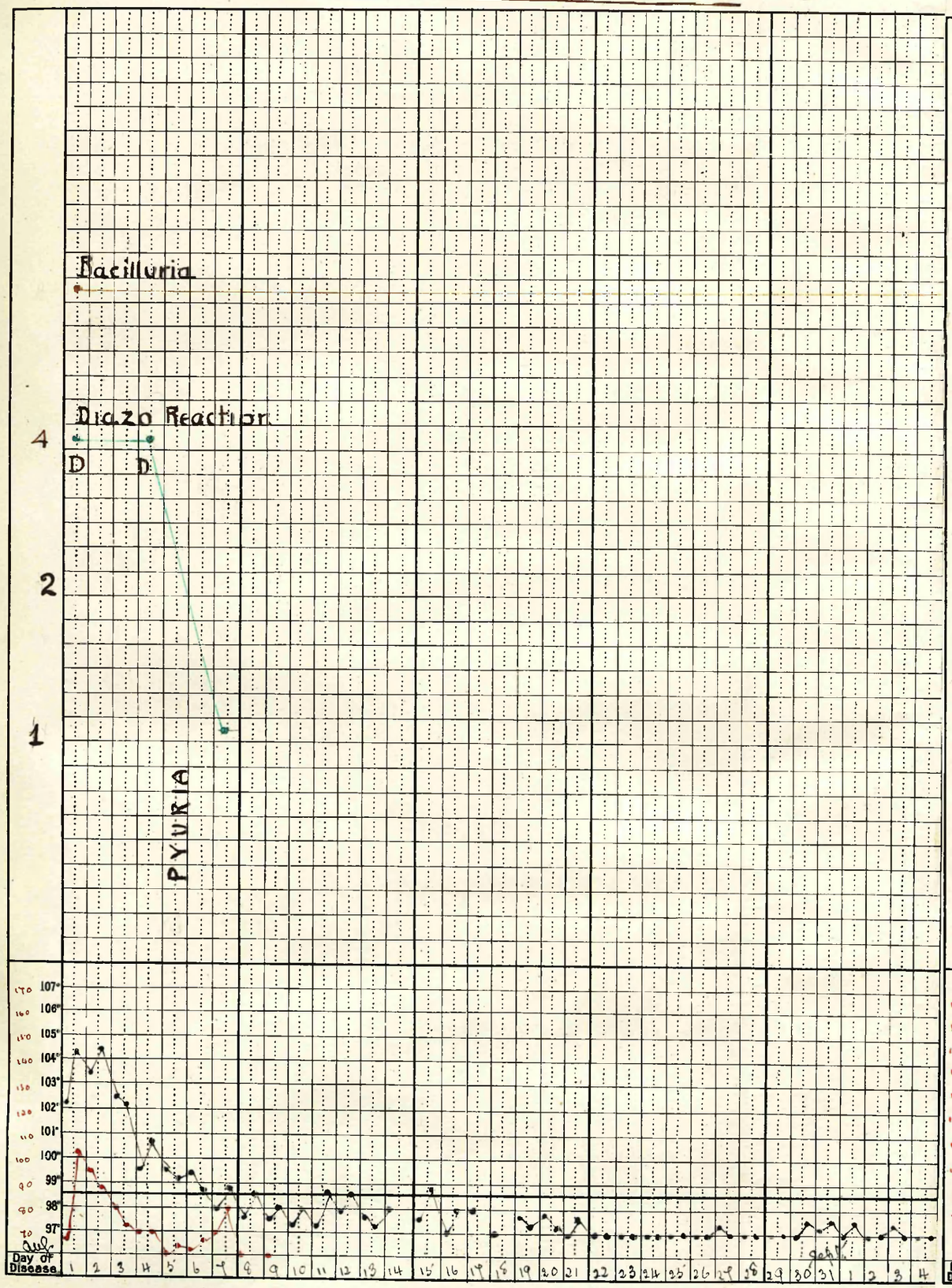
Case with Pyuria, B.D. male, age 28 (Chart 9 page 19)

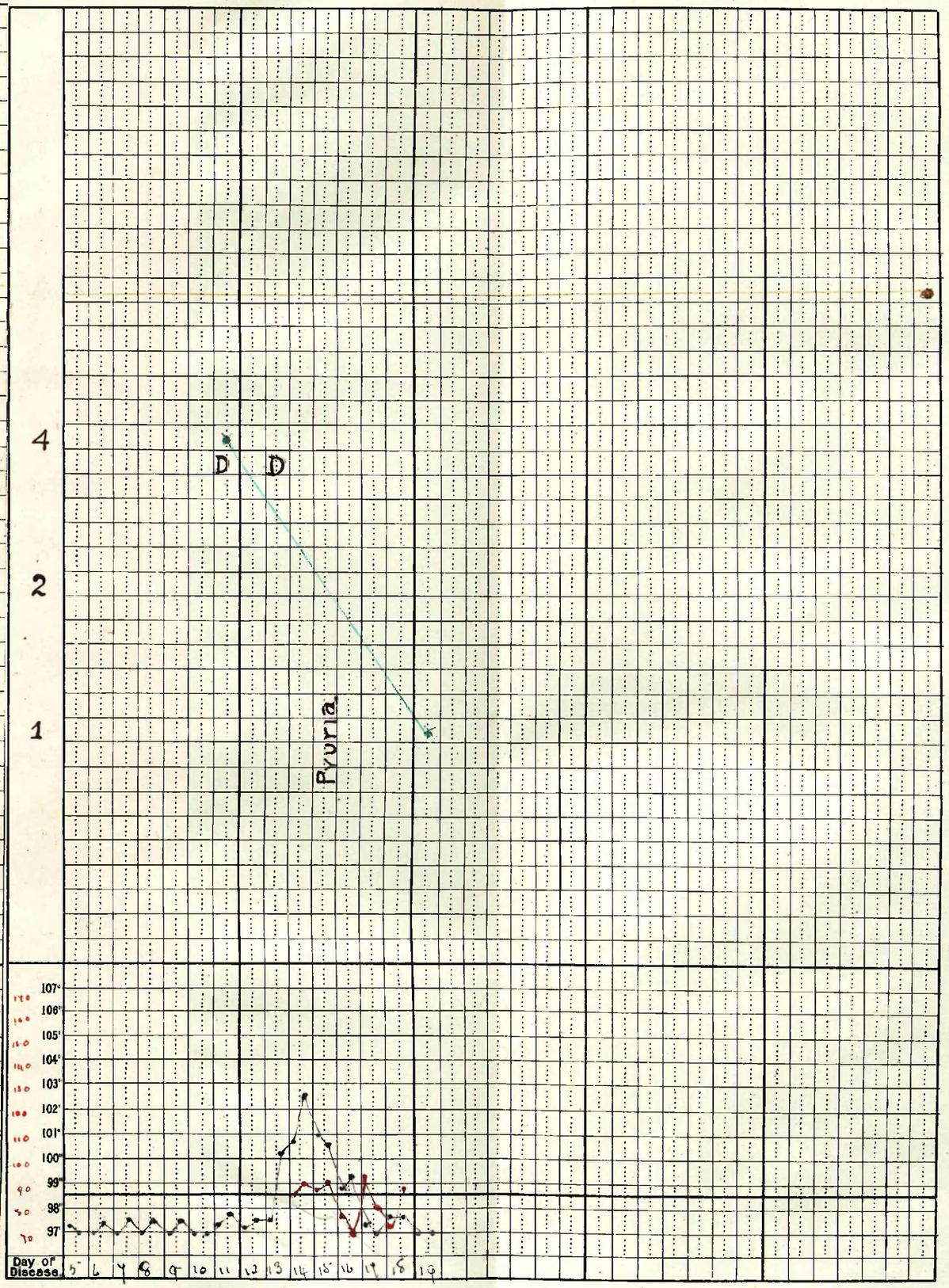
This patient was admitted to hospital in March 1908 and was dismissed in May of the same year having passed through an attack of enteric fever with nephritic complications. He was readmitted in July as he had apparently infected his wife with typhoid fever and had then been found to be suffering from bacilluria. He was treated as a convalescent patient and allowed to go about.

Three weeks after readmission, on August 1st he complained of severe pain in the left renal region accompanied by pyrexia lasting 7 days. On this date the diazo reaction was markedly present associated with the green deposit. Four days later, on August 5th pus appeared in/

CHART IX

B.D. MALE AGE 28





D-D = Duration of Deposit.

in the urine and with its appearance the diazo reaction became less marked and disappeared finally on the 7th day of this attack. The green deposit lasted till August 5th. Six weeks later a similar attack occurred when the reaction again appeared after having been absent in the interval. Typhoid bacilluria was present throughout the whole period and still existed when the patient was dismissed under sanitary supervision.

Case with Furunculosis. A. McI. male. age 18 (Chart 10 p. 21)

This patient was admitted on the 21st day of illness the initial pyrexia lasting till the 45th day. During this period the reaction and the green deposit were well marked, the former persisting till the 44th day of illness and the latter till the 31st. On the 50th day of illness an attack of furunculosis began which persisted without intermission till the 253rd day.

Throughout this period the diazo reaction and the green deposit were marked to a greater or less extent. The reaction and the green deposit were pronounced from the 50th day of illness till the 56th. From the 56th to the 90th day of illness there was a marked diminution in their intensity. On the 91st day of illness a fresh attack of furunculosis occurred which lasted till the 99th day. During this period the reaction became more pronounced and the deposit more abundant.

After evacuation of these abscesses the reaction became less pronounced. With each subsequent attack of furunculosis the same phenomena were repeated.

On the 175th day of illness a large abscess formed on the right buttock and coincidently with this the reaction again became marked. Natural resolution occurred without interference and with resolutionthe reaction disappeared. From this convalescence was uninterrupted and the patient was dismissed well.

In all those cases in which abscess formation occurred, a bacteriological examination was made.

The materials obtained from the abscesses were first tested for the presence of the bacillus typhosus according to the method previously described. In no case was a positive result obtained.

The only organism which grew in any media tried, agar, serum etc., was the staphylococcus pyogenes aureus this being always obtained in pure culture.

THE RELATION OF THE DIAZO REACTION TO PYREXIA.

The effect of the fever on the intensity of the diazo reaction will now be considered.

From the Table on page39. it will be noted that the day on which the diazo reaction was first observed is the same as that on which the patient was admitted to hospital so that the day on which it ceased can alone be discussed, for the primary attack.

Out of the 89 uncomplicated cases of enteric fever recorded in this paper, the diazo reaction terminated on the same day as that in which the pyrexia ceased in 65 instances.

In 23 cases the reaction was present for periods of 1 to 5 days beyond the termination of the febrile period. In only one instance did this occur among the mild group of cases when the period of prolongation was 5 days. In the medium group, the diazo reaction outlasted the period of fever by 1 day in 12 instances, by 2 days in 5 cases, and by 3 days once.

In the severe group the reaction outlasted the pyrexia by 1 day in one instance, by 2 days in two cases, and by 4 days once.

One case remains to be specially noted.

M. N. female, age 28. (Chart 11 page 21)

This patient was admitted on the 15th day of illness, the temperature reaching normal on the 28th day. From this/

4

2

1

D

D

Renewed
Furunculosis

Day of Disease

4

2

1

Day of Disease

4

2

1

Day of Disease

4

2

1

107° 106° 105° 104° 103° 102° 101° 100° 99° 98° 97°

Day of Disease 224 225 226 227 228 229 230 231 232 233 234 235 236 237 238 239 240 241 242 243 244 245 246 247 248 249 250 251 252 253 254 255 256 257 258 259 260 261

4

2

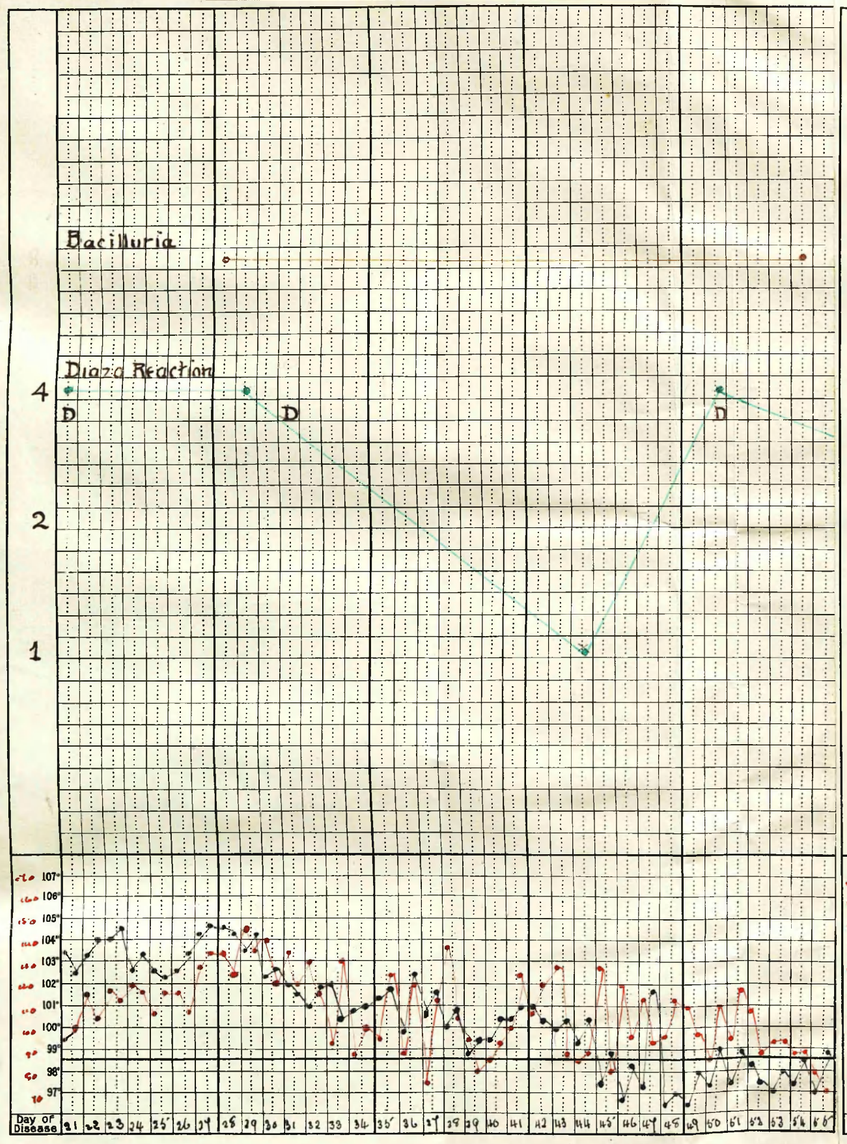
1

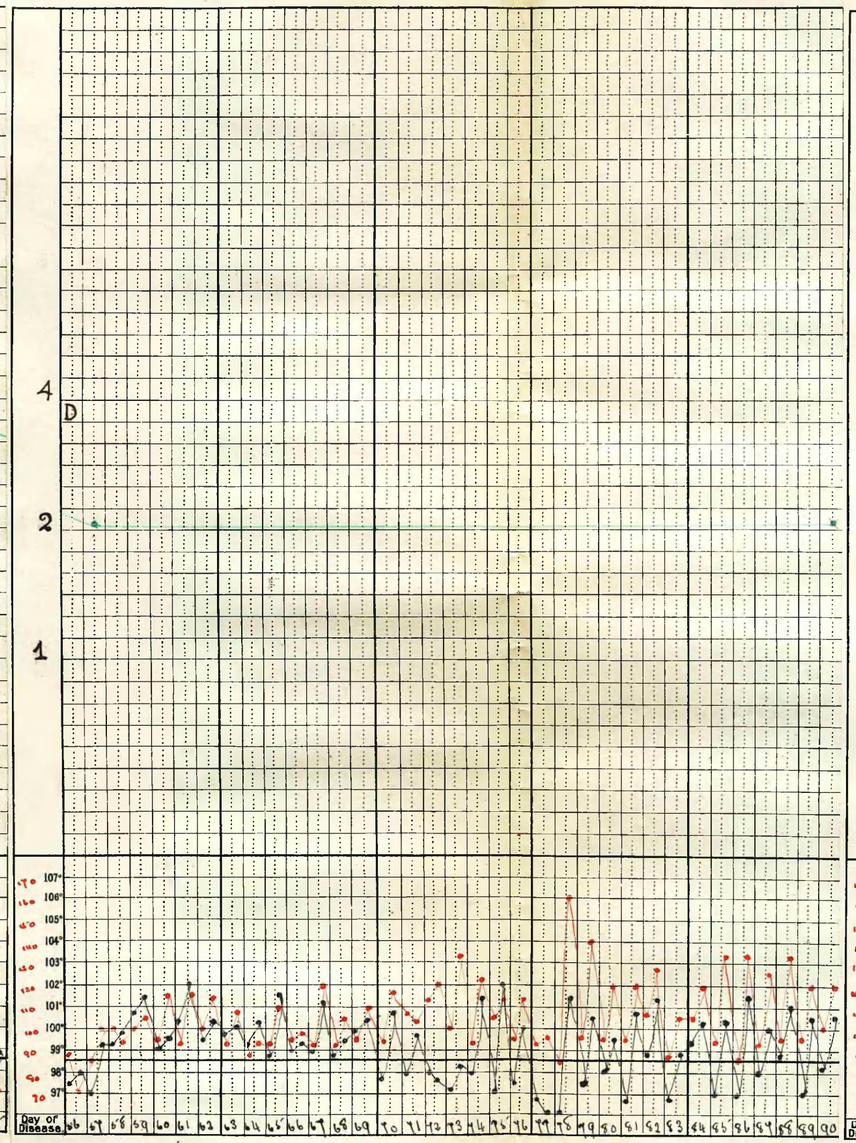
107° 106° 105° 104° 103° 102° 101° 100° 99° 98° 97°

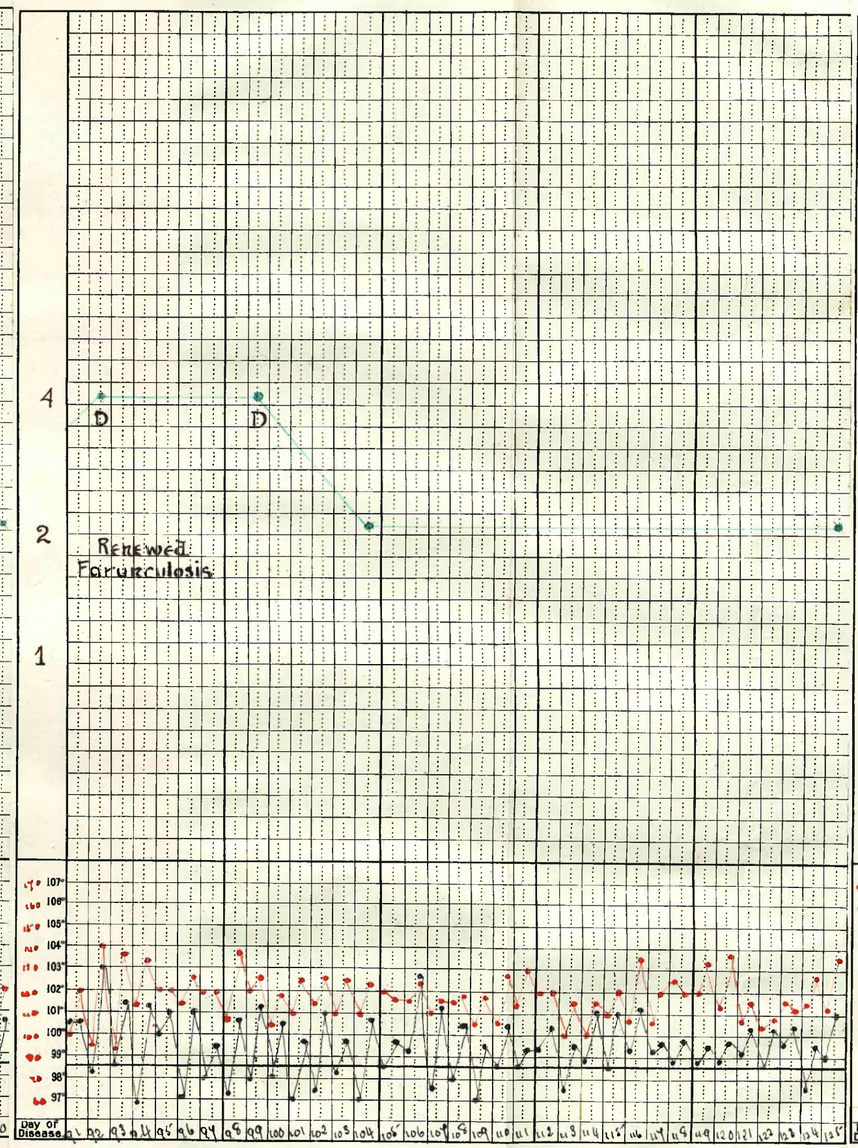
Day of Disease 262 263 264 265 266 267 268 269 270 271 272 273 274 275 276 277 278 279 280 281 282 283 284 285 286 287 288 289 290 291 292 293 294 295 296

CHART X

A. McI. MALE AGE. 18



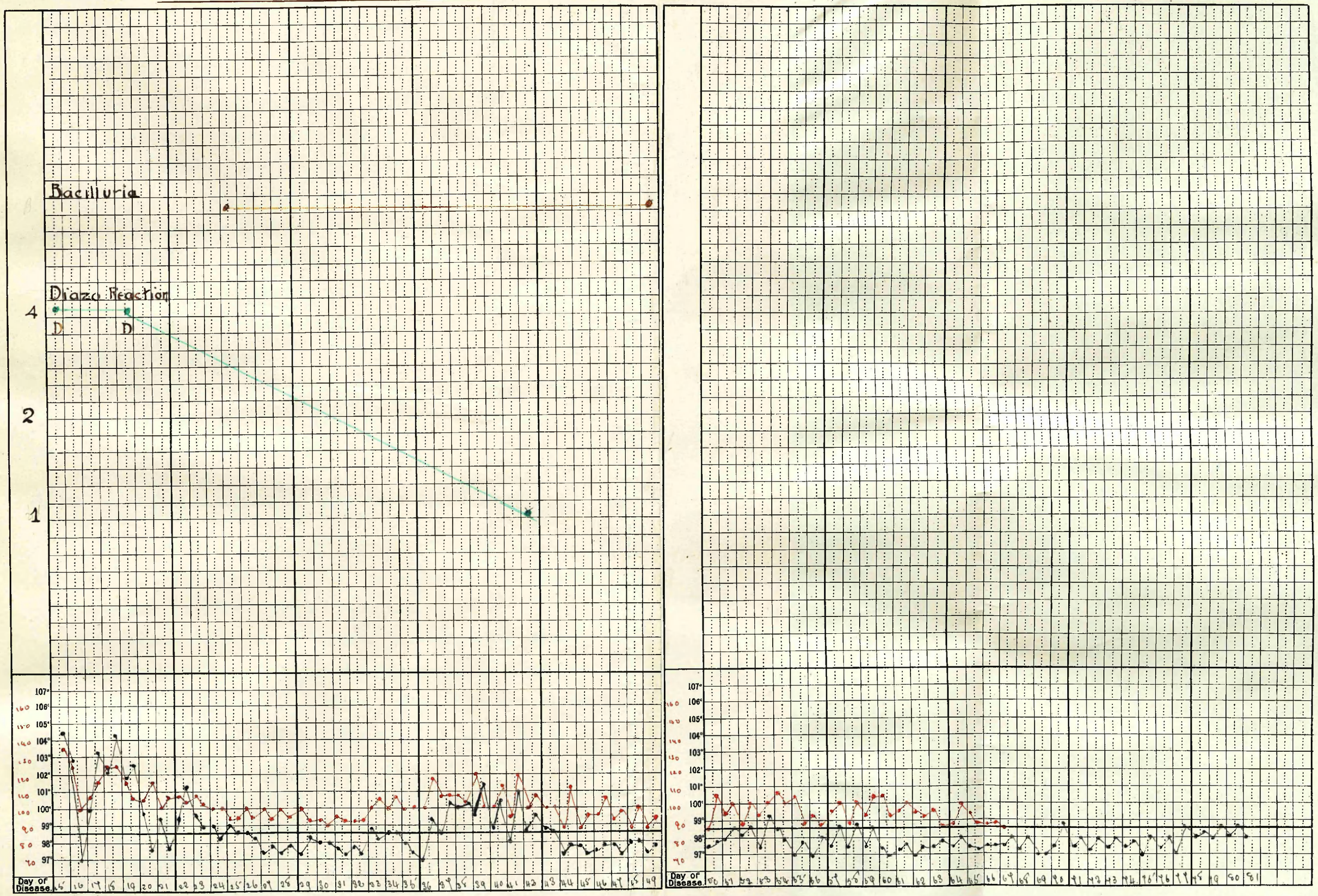




D-D = Duration of Deposit.

CHART XI

M.N. FEMALE AGE 28.



D-D = Duration of Deposit

this date till the 42nd day of illness, the temperature remained unstable finally settling to normal on the 42nd day. The reaction was maintained in this case from the commencement of the attack till the latter date.

The temperature during relapse.

The above remarks refer to the pyrexia during the primary attack but one additional point is learned from those cases in which relapse occurs viz:- that the presence of the diazo reaction frequently precedes the onset of the pyrexia in some cases by as much as 5 days. In one instance alone did the reaction fail to appear before the rise of the temperature, in this case appearing the day after.

In all other points the statements with regard to its behaviour during the primary attack apply equally to its behaviour during relapse.

Relation of the Intensity of the Color in the Diazo Reaction to the Temperature.

The previous remarks have described merely the presence or absence of the diazo reaction and no indication of its degree of intensity has been made.

In general the diazo reaction is much more marked during periods of high pyrexia and decreases in intensity as the lysis of the fever proceeds. This relation to the temperature though marked from the acme of the fever onwards does not hold in the onset. The reaction commences during the initial pyrexia. It is best marked at the commencement of the illness when the temperature is high and at this period consists of a deep crimson ring at the junction of the two liquids. As the temperature settles the intensity of the reaction alters, changing from the deep crimson to a light crimson and finally assuming a rose pink colour. The rose pink tint is as a rule obtained when the temperature approaches normal and not infrequently persists for a few days longer.

The final colour reaction obtained, a reddish brown is not considered characteristic of enteric fever. With regard to the green deposit described by Hewlett it has been previously stated that it only occurs during the first few days of pyrexia.

iv. THE VALUE OF THE DIAZO REACTION IN THE DIAGNOSIS OF ENTERIC FEVER.

It has been shown that the diazo reaction has been present at the time of first observation in 99 cases out of 100 and its presence has been demonstrated in cases of enteric fever coming under observation as early as the third and fourth days of illness.

From this it would appear that the presence of the diazo reaction is of some importance from the point of view of diagnosis.

It is also present invariably at the beginning of a relapse and in some instances precedes the elevation in temperature.

In the appendix, observations on its presence in other conditions, are discussed and from the remarks made there it will be evident that the value of the abscence, as a negative sign, in any case coming early under observation, is much greater than any positive value that it may have.

There seems to be a considerable field for its use in public health investigation. Family infections are common and in children especially, it is almost beyond medical skill to make a diagnosis in the early stages of enteric fever and it is from children that the chief risk of general infection in a family arises. It would seem that the systematic use of the diazo reaction in such cases had a distinct future. It would also seem that in the case of such outbreaks as those due to the infection of milk, that considerable assistance might be given in the separation between those illnesses, among the/

the consumers of the milk, due specially to the bacillus typhosus or to other causes, and in thus preventing further risk of the spread of infection.

With regard to the diagnostic significance of the green deposit described by Hewlett, its presence has been demonstrated in 54 uncomplicated cases of enteric fever and in 10 complicated cases, i.e. it occurs in about 64% of cases. It is not however peculiar to enteric fever as its presence has been demonstrated in typhus fever and pneumonia as will be shown in the appendix.

A report of the case in which the diazo reaction did not appear will be given in the section in Bacilluria.

Method adopted in the charting of the reaction.

In order that the Reaction might be quantitatively charted and compared with the course of the temperature, experiments were carried out with urine which when undiluted gave a very marked crimson reaction. Various dilutions in water of such specimens were made and the effect of dilution upon the colour reaction noted. This dilution was carried on till a faint reaction was produced, which was comparable with like reactions of an undiluted urine observed during the course of enteric fever.

It was found that dilution in the proportion of 1 in 1 of water generally gave a reaction of a light crimson colour and that further dilution in the proportion of 1 in 4 gave a faint rose pink reaction, comparable with a similar reaction occurring in an undiluted urine during the latter part of the febrile period in enteric fever. Beyond this point of dilution no characteristic reaction was obtained.

This point, (viz., the rose pink) is charted as unity/

unity and the different degress in ascending order of intensity by the numbers 1 to 4. The reaction charted as 4 must contain observations of greater range of variation than those below as after a certain degree of intensity is produced, it is probable that the darkness of the colour does not permit of further differentiation by the naked eye.

v. THE OCCURRENCE OF TYPHOID BACILLURIA.

Within recent years much attention has been given to the subject of "typhoid carriers" in enteric fever. That the bacillus typhosus is present in the urine of p patients suffering from enteric fever is not an new fact.

In 1894 Wright and Semple stated that the bacillus typhosus was present in the urine at a very early stage of the disease and that it occurred very frequently. They even went the length of suggesting that a bacteriological examination of the urine might be made for purposes of diagnosis. Further they were of opinion that it was by the urine and not by the faeces that infection was spread. Some remarks upon this point will be made later. Further they stated that in those urines in which no typhoid bacilli were found that bacilli coli were regularly present. They explained this phenomenon by stating that the typhoid organism cannot maintain itself in a urine to which the bacillus coli has gained access.

Within the last few years this subject has attracted much attention and it has been considered by many that the urine is a common source in the spread of the infection. How far this is so, and to what extent the disease is actually disseminated by this means, is still a matter of doubt.

The etiology of typhoid bacilluria is still a matter of dispute. By some authorities it is considered to be associated with an infection of the kidney, by others it is thought that it occurs in cases in which the bacillus typhosus is present in the blood and that the occurrence of bacilluria is due to a filtration of the organism from the blood into the excretory apparatus. It has also been suggested/

suggested that bacilluria is the result of a stray organism finding its way into the bladder and multiplying there.

These three suggestions will be discussed later in the light of my own observations.

Specimens of urine from the same cases as those examined for the diazo reaction were investigated for the presence of the organism of typhoid fever, by the method already described. In all 100 cases were examined bacteriologically on every second day from the date of admission till the date of dismissal of the patients.

The account of the observations is arranged in the following order:-

1. Date of appearance and frequency of bacilluria.
2. Duration.
3. Relation of Bacilluria to Pyrexia.
4. Associated abnormal condition of the urine.
5. Importance to be accorded to the occurrence of bacilluria as a means of spreading infection.

1. Date of appearance and frequency of Bacilluria.

Horton Smith records the 14th day of illness as the earliest day on which he found the bacillus typhosus in the urine. In general, however, it is considered that bacilluria does not occur until the third week of illness.

With regard to the frequency it has been stated by Horton Smith that bacilluria occurred in 17 cases out of 45 or 38%

Other observers have given different estimates but the number of their cases has been too small to render their results of special value. In fact any results can only be considered to apply to the epidemic examined, as it is highly probable that the relative frequency will vary from epidemic to epidemic.

In the series of cases under consideration the

TABLE VII.

Table showing the day of onset and the duration of Bacilluria, etc.

	Number of Case.	Day on which Temp. Reached Normal.	Day of first Examination.	Day of Appearance of Bacillus Typh.	Condition of the Urine.	Nature of Case.	Result.
	1.	18th. day.	4th. day.	15th. - 22nd. day.	Normal.	Severe.	R.
	2.	- "	10th. "	16th. - 22nd. "	Trace of Albumin.	"	D.
	3.	- "	9th. "	18th. - 37th. "	Trace of Albumin.	"	D.
	4.	24th. "	9th. "	22nd. - 52nd. "	Normal.	"	R.
	5.	28th. "	15th. "	25th. - 49th. "	Trace of Albumin.	"	R.
	6.	45th. "	21st. "	28th. - 54th. "	" " "	"	R.
	7.	20th. "	9th. "	15th. - 31st. "	Normal.	Medium.	R.
	8.	17th. "	6th.	16th. - 40th. "	Trace of Albumin.	"	R.
	9.	- "	17th. "	17th. - 25th. "	Normal.	"	R.
	10.	14th. "	3rd. "	18th. - 36th. "	"	"	D.
	11.	16th. "	7th. "	20th. - 76th. "	"	"	R.
	12.	23rd. "	7th. "	21st. - 28th. "	"	"	R.
	13.	- "	18th. "	21st. - 33rd. "	"	"	R.
	14.	- "	21st. "	21st. - 34th. "	"	"	D.
	15.	30th. "	21st. "	23rd. - 35th. "	"	"	D.
	16.	17th. "	9th. "	26th. - 60th. "	Trace of Albumin.	"	R.
	17.	-	28th. "	28th. - 39th. "	Normal.	"	R.
	18.	20th. "	7th. "	29th. - 39th. "	"	"	D.
x	19.	14th. "	8th. "	35th. - 51st. "	"	"	R.
	20.	28th. "	13th. "	35th. - 66th. "	"	"	R.
x	21.	20th. "	13th. "	?	Purulent.	Mild.	R.
	22.	18th. "	4th. "	18th. - 42nd. "	Normal.	"	R.
	23.	35th. "	28th. "	28th. - 55th. "	"	"	R.
	24.	42nd. "	35th. "	35th. - 69th. "	"	"	R.
x	25.	20th.	13th. "	43rd. - 63rd. "	"	"	R.
x	26.	?	?	?	Purulent.	"	R.

X Cases 19, 21, 25 and 26 will be described more specially later.

presence of the bacillus typhosus was demonstrated in 26 instances (Table 7). Of these 26 cases, 6 belong to the severe group, 14 to the medium, and 5 to the mild: that is to say that bacilluria occurred in $33^1/_3\%$ of severe cases, 24% of the medium, and 21.9% of the mild.

The search for the organism in the urine was begun in every case on the day of the patient's admission to hospital.

The earliest day on which the bacillus typhosus was found to be present was the 15th and in this case the patient came under observation on the 4th day of illness. The latest date at which the organism made its first appearance was the 43rd day of illness and in this instance examination for the organism had been carried out from the 13th day of illness, a period antedating the earliest appearance of the organism in my series of cases. In 11 cases the presence of the organism was not demonstrated till after the 23rd day of illness though in 7 of these cases the first examination had been made on or before the 21st day of illness and in the remaining 4 bacilluria was present at the time of admission to hospital.

Two of the 7 cases in which bacilluria occurred late in the disease are related to relapse and will be briefly described.

Case 1. W.D. age 43. (Case No.25 table 7. chart No.8 page 18)

(This case has already been referred to on page 18. and its relation to the Diazo Reaction discussed.)

This patient was admitted on the 13th day of illness and the pyrexia lasted till the 20th day. Relapse began 7 days later i.e. on the 27th day of illness and terminated on the 37th. The presence of the bacillus typhosus was not however demonstrated till the 43rd day of illness/

illness, that is to say on the 16th day of relapse. The bacilluria persisted till the 63rd day of illness outlasting the relapse by 26 days.

Before convalescence was completed bacilluria had disappeared.

Case 2. P.W. male, age 14. (Case No. 19 Table 7, Chart No. 4 Page 15.)

(This case has already been referred to in the Section on the Diazo Reaction.)

This patient was admitted on the 8th day of illness. The temperature did not settle and a relapse began on the 19th day of illness persisting till the 40th day. Bacilluria developed on the 35th day of illness i.e. on the 15th day of the relapse and persisted till the 51st day i.e. for 8 days after the temperature had reached normal. Frequent examinations of the urine after this date failed to reveal a recurrence of this condition and patient was dismissed well.

From the consideration of the foregoing it would appear that bacilluria does not occur in the early stages of the disease. It is not as a rule found till after the second week. Further it occurs more frequently in the severer forms of the disease.

Relation between Severity of the Attack and the Early occurrence of Bacilluria.

It might be thought that Bacilluria would appear earlier in severe cases than in mild. This however is not so as may be easily seen from an examination of Table No. 7, a fact that rather tells against the theory that an accidental contamination of the urine in the bladder takes place, and is rather in favour of the suggestion that the occurrence of bacilluria, at least in the beginning, is an eliminative process.

TABLE VIII.

Table showing duration (in days) of Bacilluria in 24 Cases.

Number of Cases. Duration in Days.	6.	7.	8.	10.	11.	12.	13.	16.	18.	19.	20.	24.	26.	27.	30.	31.	34.	56.	TOTALS.
Mild.											1.	1.		1.			1.		4.
Medium.		1.	D 1.	1.	D 1.	D 2.	D 1.	2.	1.			1.				1.	1.	1.	14.
Severe.	D 1.	1.								D 1.		1.	1.		1.				6.
	1.	2.	1.	1.	1.	2.	1.	2.	1.	1.	1.	3.	1.	1.	1.	1.	2.	1.	24.

D = Death.

Duration of Bacilluria. (Table 8.)

Horton Smith has stated that the shortest duration of bacilluria in his cases was 8 days and the longest 70. Gwyn, however, mentions a case in which bacilluria was present for years.

In the cases under consideration the shortest period during which bacilluria was observed to persist, in cases ending in recovery, was 7 days and the longest 56. None of these cases received any treatment which was intended, or could be expected, to interfere with the natural termination of the condition. Two cases are excepted from the above statement (Cases Nos. 21 and 26 Table 7) as in these cases there was a definite pyogenic condition of the kidney.

With regard to the character of the case the shortest duration of bacilluria was, in the severe group 7 days, medium 7 days and mild 20 days while the longest period of duration was in the severe cases 30 days, medium 56, and mild 34 days.

From these facts it would appear that bacilluria is of very variable duration and that its duration bears no relationship to the type of the disease, is borne out by the fact that the same periods of duration during which bacilluria is present appear in all three groups. The days on which bacilluria was observed to cease, are given in the annexed table (Table 9).

The earliest day on which bacilluria ceased in cases ending in recovery was, in the severe group the 22nd day of illness, in the medium the 28th and in the mild the 42nd day of illness, while that latest day of illness on which it was observed was in the severe group, the 54th day of illness, in the medium the 76th and in the mild the 69th day.

TABLE IX.

Table showing the day in which Bacilluria ceased in 24 Cases.

Day on which Bacill. ceased.		22.	25.	28.	31.	33.	34.	35.	36.	37.	39.	40.	42.	49.	51.	52.	54.	55.	60.	63.	66.	69.	76.	TOTALS.
Number of Cases.	Mild.												1.					1.		1.		1.		4.
	Medium.		D 1.	1.	1.	D 1.	D 1.	1.	1.		D 2.	1.			1.				1.		1.		1.	14.
	Severe.	D 2.								D 1.				1.		1.	1.							6.
		2.	1.	1.	1.	1.	1.	1.	1.	1.	2.	1.	1.	1.	1.	1.	1.	1.	1.	1.	1.	1.	1.	24.

D = Death.

The Relation of the Onset of Bacilluria to its Duration.

From the consideration of Table No. 7 page 27. it will be seen that the day of the onset of bacilluria is no guide to its duration as cases with the same days of onset of bacilluria have widely different periods of duration..

3. The Relation of Bacilluria to the Pyrexia.

It has been stated earlier in the paper that bacilluria occurs more frequently in the severer forms of the disease, that is to say, more frequently in casds with a high temperature.

With regard to the duration of the pyrexia and the duration of bacilluria, a glance at Table 7 page 27. will show that no association exists between them, as, among cases in which the period of pyrexia is short, instances of prolonged bacilluria are just as frequent as among those in which the period of pyrexia is prolonged. Further, that cases, in which bacilluria did not occur till after the cessation of the temperature, have as long periods of duration as those commencing during the pyrexial period. One point yet remains to be emphasised in connection with the temperature, viz:- that at the time of the first appearance of the bacilluria no distinct elevation of the temperature was seen (see Charts) except in two cases in which it was associated with pyrexia. These cases are summarised in the following notes.

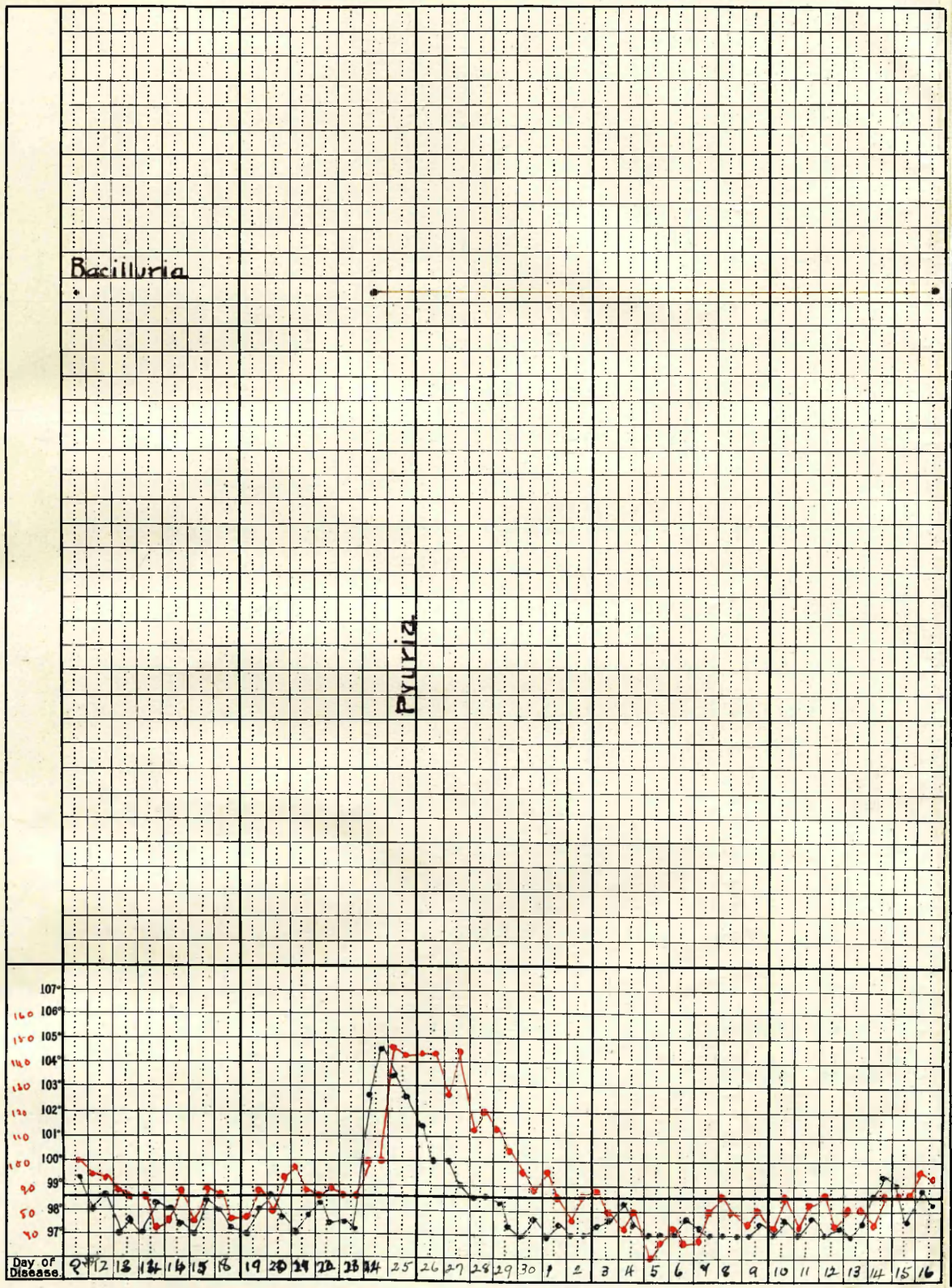
Case 1. B.D. male. age 28 (Chart 9 page 19.)

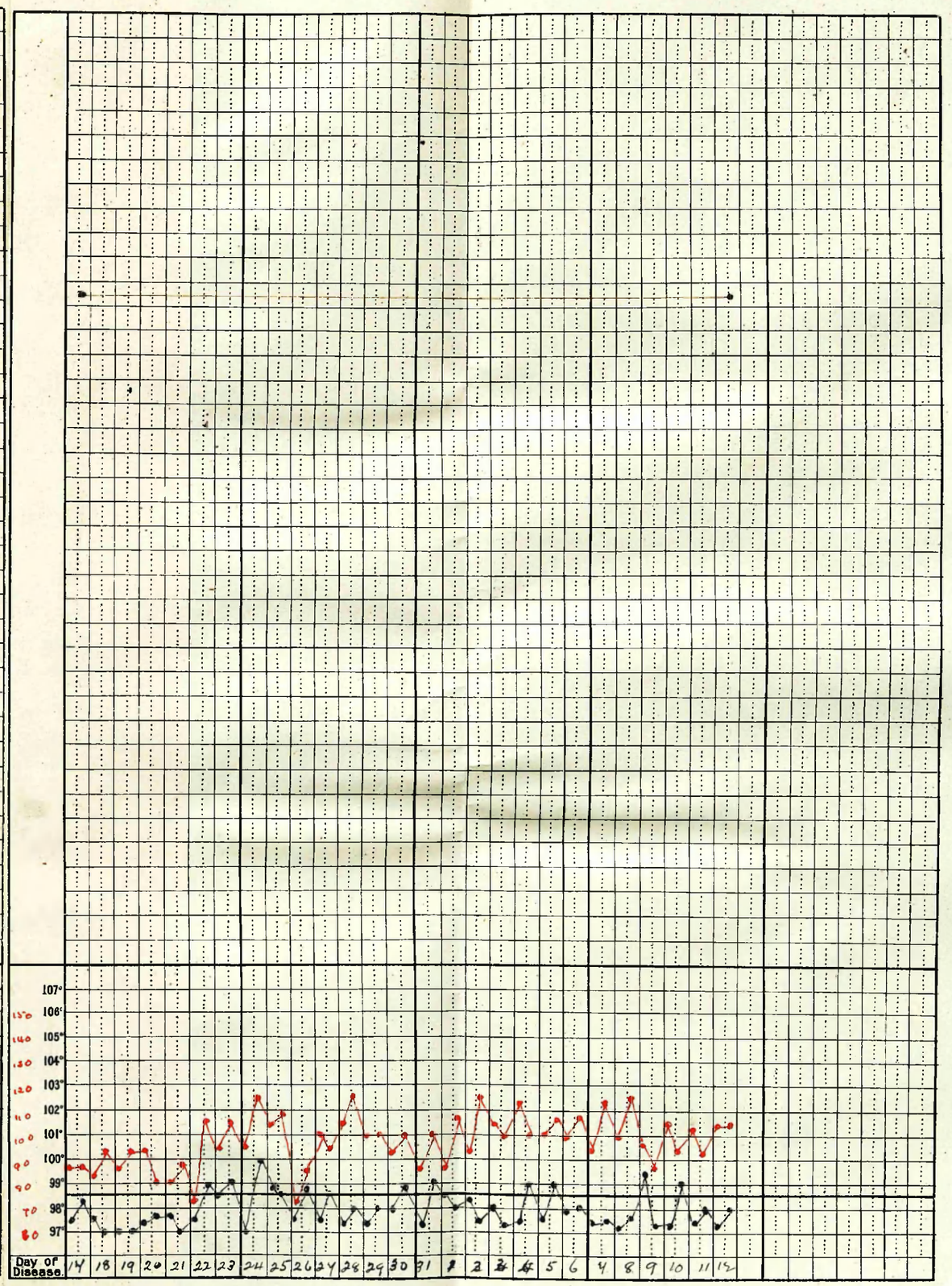
(This case has already been referred to on page 18.)

The patient was admitted in March 1908 and was dismissed in May of the same year. During his residence in Hospital he had a nephritic complication, large quantities/

CHART XII

M. A. FEMALE AGE 14.





quantities of albumin being present in the urine. After his dismissal his wife was admitted with enteric fever.

The faeces and urine were then subjected to examinationnby the Sanitary Department, as a result of which he was readmitted, with Bacilluria, on July 12th 1908. At the time of readmission, with a normal temperature, slight traces of albumin were present in the urine. Three weeks later, on August 1st, he was confined to bed in consequence of an elevation in the temperature and at this time he complained of pain in the left renal region. On August 5th pus appeared in the urine lasting four days.

On Sept. 11th a similar attack occurred the pyrexia, on this occasion, persisting for two days. Bacilluria continued without intermission throughout the patient's residence in hospital and on dismissal, under sanitary supervision, the organisms were still present in the urine.

Case 2 M. A. female, age 14 (Chart No. 12 page 31)

In this case the day of illness was not ascertained as the patient came under observation late in the disease.

She was admitted to hospital on 12th November 1908 with a normal temperature. An examination of the urine at this time was negative.

Twelve days after admission, on November 24th, an elevation of the temperature occurred, the highest register being 104.6°F and on the following day, November 25th, pus appeared in the urine. An examination of the urine at this stage revealed the presence of large numbers of the bacillus typhosus. The purulent condition persisted for 3 weeks while bacilluria persisted for 7 weeks from the date of onset.

The convalescence of the patient was satisfactory

and in spite of bacilluria she was dismissed well on January 29th 1909, two weeks after bacilluria had ceased.

In these two cases a secondary typhoid abscess of the kidney probably occurred as typhoid pyelitis pure and simple does not seem to be associated with a temperature.

The occurrence of Bacilluria in these cases seems to have an analogy with cases of typhoid abscess elsewhere rather than with the typical bacilluria.

4. Associated abnormal Conditions of the Urine.

With regard to the character of the urine in cases of bacilluria much has been written. Many authors state that bacilluria is most commonly associated with an infection of the kidney, and that, in consequence, albumin or pus is present in the urine. In Horton Smith's cases 9 out of 17 showed pyuria. In the series of cases just considered in only 2 of 26 patients did pyuria occur. At the onset of bacilluria, in only 6 of the remaining 24 cases was albumin present while blood was never found. Of these, 4 cases belong to the severe group and 2 to the medium.

In these cases, however in which albuminuria was present at the onset of bacilluria a certain amount of febrile nephritis might be inferred.

Another feature of the urine described by most authors is the presence of a characteristic "shimmer", on holding the test tube up to the light. This is considered characteristic of a urine containing typhoid organisms in large numbers.

In my series of cases this "shimmer" was present. in the urine of 16 patients and it was impossible to say whether the number of organisms present in these urines was greater than in urines showing no "shimmer".

Further, I noticed that this "shimmer" was occasionally present in urines which, on examination, were found to contain the bacillus coli.

It is certain, however, that the bacillus typhosus is frequently present in such large numbers as to produce turbidity of the urine.

Etiology of Bacilluria.

The etiology of typhoid bacilluria is still a matter of dispute. As previously stated some authorities consider it to be due to an infection of the kidney, others, that it is due to bacteriaemia and that the presence of the bacillus typhosus in the urine is due to a filtration from the blood into the excretory apparatus. A third view accepted is that it is caused by a stray organism finding its way into the bladder and multiplying there.

From my own cases it is evident that kidney infection plays only a small part in explaining the occurrence of bacilluria, as in only two cases was there any evidence of kidney infection.

As before remarked the absence of any special pyrexia during the onset of bacilluria, and the independence which the date of onset of bacilluria shows with regard to the type of the fever, rather favours the idea that it is in part an eliminative process. Of clinical evidence of bacteriaemia the occurrence of "rose spots", in the light of present knowledge, is probably conclusive as it is well known that these not infrequently occur after the temperature is normal. The presence of the bacillus typhosus in the blood does therefore, not necessitate the presence of pyrexia. In view of this, the general numerical distribution of the date/

date of onset suggests that some power is acquired by the kidney of actively eliminating the organism of typhoid fever.

With regard to the third hypothesis viz., that the typhoid organisms accidentally finds its way into the bladder and there multiplies, one difficulty was raised by the older writers viz:- that the bacillus typhosus was not capable of multiplying in the urine. This, however, is a complete mistake (vide appendix). On the whole it would seem probable that in the bulk of the cases the original infection of the urine is eliminative but the continuance of the infection is possibly largely a matter of the organism multiplying in the bladder.

The importance to be accorded to Bacilluria as a means of spreading infection.

Typhoid bacilluria has been long stated to be an important factor in the dissemination of Enteric Fever.

Although it is a matter of dispute as to the percentage of cases in which bacilluria occurs, the fact that its presence is demonstrated might be sufficient to account for many cases of enteric fever the cause of which has been unexplained. In the series of cases just considered bacilluria occurred in 26% and it has been shown that it is not during the acute period that risk of infection occurs but when the patient is approaching convalescence.

With the exception of a single instance, all the cases were dismissed from their clinical appearances and it is important to note that even in the most prolonged bases of bacilluria, with the exception of B.D. Case21) the bacilluria had ceased for two weeks before dismissal from hospital.

It/

It would then seem that there is in general no great risk of the dissemination of enteric fever by this means. What, however, has been specially learned is, that when there is a definite kidney lesion the bacillus of enteric fever may be present for many months.

This is in accordance with previous experience in the hospital where a case of recurrent pyelitis of the kidney was found on each occasion of the recurrence to be associated with a great increase of the typhoid organisms in the urine. Apart from recurrent pyelitis however, it has, in the experience of the hospital, been observed that some cases of pyelitis were exceptionally chronic and although the urine, in these cases, was not specially examined for the typhoid organism it is probable that it would easily have been recognised had it been specially looked for. In such cases, however, the urine has appearances which are sufficiently obvious to the naked eye, and which in future will receive more detailed examination. They constitute, but a very minute fraction, considerably less than 1%, of the total cases admitted.

vi. THE RELATION BETWEEN THE DIAZO REACTION AND BACILLURIA.

As has been previously stated it was thought that if the presence of the diazo reaction in enteric fever was an indication that the bacillus typhosus was present in the blood it might also indicate its presence in the urine.

From the previous sections it will have been observed that the diazo reaction always occurs in cases of enteric fever and early in the disease whereas bacilluria is present in only 26% of cases and never occurs earlier than the third week of illness. Since the Diazo Reaction is present as a rule, till the third week of the disease and frequently much later, it follows that bacilluria and the diazo reaction occur together. The reaction, however, in these cases, is related to the period of the disease and not to the bacilluria. In some instances, for example, with relapse cases, the diazo reaction occurs simultaneously with the bacilluria, but in these cases it is evident that the occurrence of the reaction is due to the relapse.

It may be stated generally that in these cases in which the diazo reaction is present when bacilluria begins it ceases as a rule considerably before the termination of bacilluria.

In fact, it has been clearly demonstrated that no conclusion regarding the infectivity of the urine can be drawn from the presence or absence of the diazo reaction.

C O N C L U S I O N S.

1. The diazo reaction is always present as early in the course of the fever as the case comes under observation and if the evidence of the relationship to relapse can be taken as sound, it is probably present even before the first rise of the temperature i.e. during the incubation period of the disease.

   It is much more prolonged in severe attacks of the fever and less prolonged in cases of medium and mild severity.

2. Its intensity continues marked during the whole period of high pyrexia and gradually diminishes with the lysis of the fever. That this has some relation at least to the temperature, is borne out by the fact that in typhus fever and pneumonia the disappearance is sudden and absolute with the onset of crisis.

3. That an intense reaction is in general associated with a green deposit when the tube of urine in which the reaction has been performed with marked positive result is allowed to stand for 12 to 24 hours. This reaction, however, is also obtained in the urines of cases of Typhus Fever and Pneumonia though curiously enough not in cases of measles even when the intensity of the reaction should produce it.

   In cases of furunculosis in which the diazo reaction occurred there was no evidence that the reaction was due to a new typhoid infective process but was produced by the staphylococcus pyogenes aureus just as it is by the preumococcus in pneumonia.

TABLE X.

Table showing Day of Commencement and Duration of :-

1. Pyrexia. 2. Diazo Reaction. 3. Green Deposit. 4. Bacilluria, in 89 Cases.

Type of Case.	Result.	Day of Commencement & Duration of Pyrexia.	Day of Commencement & Duration of Diazo Reaction.	Day of Commencement & Duration of Green Deposit.	Day of Commencement & Duration of Bacilluria.
Medium.	R.	3rd. - 18th. day.	3rd. - 18th. day.	3rd. to 9th. day.	-
"	R.	3rd. - 14th. "	3rd. - 17th. "	3rd. - 6th. "	18th. - 36th. day.
Severe.	R.	4th. - 18th. "	4th. - 22nd. "	4th. - 11th. "	15th. - 22nd. "
Medium.	R.	4th. - 16th. "	4th. - 18th. "	4th. - 8th. "	-
"	R.	4th. - 15th. "	4th. - 16th. "	4th. - 7th. "	-
"	R.	4th. - 18th.	4th. - 19th. "	4th. - 10th. "	-
Mild.	R.	4th. - 18th. "	4th. - 18th. "	4th. - 7th. "	18th. - 42nd. "
Medium.	R.	5th. - 19th. "	5th. - 19th. "	5th. - 9th. "	-
Severe.	D.	6th. - 18th. "	6th. - 18th. "	6th. - 12th. "	-
Medium.	R.	6th. - 17th. "	6th. - 17th. "	6th. - 9th. "	16th. - 40th. "
"	R.	6th. - 19th. "	6th. - 20th. "	6th. - 11th. "	-
"	R.	6th. - 17th. "	6th. - 17th. "	6th. - 9th. "	-
"	R.	7th. - 20th. "	7th. - 21st. "	7th. - 13th. "	29th. - 39th. "
"	R.	7th. - 23rd. "	7th. - 25th. "	7th. - 13th. "	21st. - 28th. "
Mild.	R.	7th. - 15th. "	7th. - 15th. "	-	-
"	R.	7th. - 14th. "	7th. - 14th. "	-	-
"	R.	7th. - 14th. "	7th. - 14th. "	-	-
Severe.	R.	8th. - 26th. "	8th. - 28th. "	8th. - 16th. "	-
Medium.	R.	8th. - 17th. "	8th. - 18th. "	8th. - 12th. "	-
"	R.	8th. - 18th. "	8th. - 18th. "	8th. - 11th. "	-
"	R.	8th. - 15th. "	8th. - 15th. "	-	-
"	R.	8th. - 19th. "	8th. - 20th. "	8th. - 12th. "	-
"	R.	8th. - 18th. "	8th. - 18th. "	8th. - 10th. "	-
"	R.	8th. - 17th. "	8th. - 17th. "	-	-
"	R.	8th. - 20th. "	8th. - 22nd. "	-	-
"	R.	8th. - 20th. "	8th. - 21st. "	-	-
"	R.	8th. - 21st. "	8th. - 23rd. "	8th. - 12th. "	-
"	R.	8th. - 23rd. "	8th. - 24th. "	8th. - 13th. "	-

TABLE X Contd.

Type of Case.	Result.	Day of Commencement & Duration of Pyrexia.	Day of Commencement & Duration of Diazo Reaction.	Day of Commencement & Duration of Green Deposit.	Day of Commencement & Duration of Bacilluria.
Medium.	R.	8th. - 24th. day.	8th. - 26th. day.	8th. - 11th. day.	-
Severe.	R.	9th. - 24th. "	9th. - 34th. "	9th. - 16th. "	22nd. - 52nd. day.
"	D.	9th. - 13th. "	9th. - 13th. "	9th. - 13th. "	-
Medium.	R.	9th. - 22nd. "	9th. - 23rd. "	9th. - 13th. "	-
"	R.	9th. - 19th. "	9th. - 19th. "	-	-
Mild.	R.	9th. - 16th. "	9th. - 16th. "	-	-
"	R.	9th. - 14th. "	9th. - 14th. "	-	-
"	R.	9th. - 15th. "	9th. - 15th. "	-	-
Severe.	D.	10th. - 22nd. "	10th. - 22nd. "	10th. - 14th. "	16th. - 32nd. "
"	D.	10th. - 14th. "	10th. - 14th. "	10th. - 14th. "	-
Medium.	R.	10th. - 22nd. "	10th. - 22nd. "	10th. - 13th. "	-
Severe.	D.	11th. - 15th. "	11th. - 15th. "	11th. - 15th. "	-
"	R.	11th. - 32nd. "	11th. - 33rd. "	11th. - 18th. "	-
"	R.	11th. - 30th. "	11th. - 32nd. "	11th. - 19th. "	-
Medium.	R.	11th. - 21st. "	11th. - 21st. "	11th. - 14th. "	-
"	R.	11th. - 24th. "	11th. - 24th. "	11th. - 17th. "	-
Mild.	R.	11th. - 15th. "	11th. - 15th. "	-	-
Severe.	D.	12th. - 14th. "	12th. - 14th. "	12th. - 14th. "	-
"	D.	12th. - 16th. "	12th. - 16th. "	12th. - 16th. "	-
Medium.	R.	12th. - 24th. "	12th. - 24th. "	12th. - 18th. "	-
"	R.	12th. - 22nd. "	12th. - 22nd. "	12th. - 15th. "	-
"	R.	13th. - 28th. "	13th. - 28th. "	13th. - 19th. "	35th. - 65th. "
"	R.	13th. - 23rd. "	13th. - 23rd. "	13th. - 16th. "	-
"	R.	13th. - 22nd. "	13th. - 22nd. "	-	-
"	R.	14th. - 23rd. "	14th. - 24th. "	14th. - 17th. "	-
Mild.	R.	14th. - 19th. "	14th. - 19th. "	-	-
"	R.	14th. - 17th. "	14th. - 17th. "	-	-
Severe.	R.	15th. - 28th. "	15th. - 42nd. "	15th. - 19th. "	25th. - 49th. "

TABLE. X. Contd.

Type of Case.	Result.	Day of Commencement & Duration of Pyrexia.	Day of Commencement & Duration of Diazo Reaction.	Day of Commencement & Duration of Green Deposit.	Day of Commencement & Duration of Bacilluria.
Medium.	R.	15th. - 24th. day.	15th. - 24th. day.	15th. - 19th. day.	-
"	R.	15th. - 26th. "	15th. - 27th. "	15th. - 18th. "	-
Mild.	R.	15th. - 20th. "	15th. - 20th. "	-	-
"	R.	15th. - 19th. "	15th. - 19th. "	-	-
Medium.	R.	16th. - 23rd. "	16th. - 23rd. "	16th. - 19th. "	-
"	R.	16th. - 26th. "	16th. - 27th. "	-	-
"	D.	17th. - 25th. "	17th. - 25th. "	17th. - 20th. "	17th. - 25th. day.
Mild.	R.	17th. - 22nd. "	17th. - 22nd. "	-	-
"	R.	17th. - 22nd. "	17th. - 22nd. "	-	-
"	R.	17th. - 22nd. "	17th. - 22nd. "	-	-
Medium.	D.	18th. - 33rd. "	18th. - 33rd. "	18th. - 22nd. "	21st. - 33rd. "
"	R.	18th. - 22nd. "	18th. - 22nd. "	-	-
"	R.	21st. - 30th. "	21st. - 30th. "	21st. - 23rd. "	23rd. - 35th. "
"	D.	21st. - 34th. "	21st. - 34th. "	21st. - 24th. "	21st. - 34th. "
"	R.	21st. - 26th. "	21st. - 26th. "	21st. - 23rd. "	-
"	R.	21st. - 28th. "	21st. - 28th. "	-	-
"	R.	21st. - 29th. "	21st. - 29th. "	-	-
"	R.	21st. - 29th. "	21st. - 29th. "	-	-
"	R.	21st. - 27th. "	21st. - 27th. "	21st. - 23rd. "	-
"	R.	21st. - 28th. "	21st. - 28th. "	21st. - 24th. "	-
"	R.	21st. - 28th. "	21st. - 28th. "	-	-
Mild.	R.	21st. - 25th. "	21st. - 25th. "	-	-
"	R.	21st. - 25th. "	21st. - 25th. "	-	-
"	R.	21st. - 25th. "	21st. - 25th. "	-	-
Severe.	R.	24th. - 36th. "	24th. - 36th. "	24th. - 30th. "	-
Medium.	D.	28th. - 39th. "	28th. - 39th. "	28th. - 31st. "	28th. - 39th. "
"	R.	28th. - 34th. "	28th. - 34th. "	-	-
"	R.	28th. - 32nd. "	28th. - 32nd. "	-	-
"	R.	28th. - 32nd. "	28th. - 32nd. "	-	-

TABLE. X. Contd.

Type of Case.	Result.	Day of Commencement & Duration of Pyrexia.	Day of Commencement & Duration of Diazo Reaction.	Day of Commencement & Duration of Green Deposit.	Day of Commencement & Duration of Bacilluria.
Mild.	R.	28th. - 35th. day.	28th. - 40th. day.	28th. - 30th. day.	28th. - 55th. day.
Medium.	R.	35th. - 42nd. "	35th. - 42nd. "	-	-
"	R.	35th. - 42nd. "	35th. - 42nd. "	-	-
Mild.	R.	35th. - 42nd. "	35th. - 42nd. "	-	35th. - 69th. "

B I B L I O G R A P H Y.

DIAZO REACTION.

Ehrlich Charité Annalen 1883:
Von Jacksch, Clinical Diagnosis:
Johns Hopkins Hospital Bulletins 1890:
Dawson, Dublin Med. Sc. 1893:
Edward, Medical News, 1892:
Hewlett, "The Lancet" 1895.
Ghenken Rousski Wratch.,1907.

MEDIA.

Thompson Yates, Laboratory Reports 1900-1901.
Macconkey, Journal of Hygiene, 1905.

BACILLURIA AND "TYPHOID CARRIERS".

Wright and Semple, The Lancet 1894:
Gwyn, Johns Hopkins Hosp. Bullet. 1899.
Horton Smith, The Lancet, 1900.
Vincent, C.R.Soc. Biologie, 1903.
Herbert, Münch. Mediz.,Woch.,t, LI 1904.
Buchan, "Bacteriuria in Enteric Fever" Thesis 1905.
Forster & Kayser, Münch. mediz.,Woch.,1905.
Minelli Centralblatt für Bakteriologie, 1906:
Remlinger, Presse Medicale 1907:
Tzeider Rousski Wratch.1907.
Bauman, Gesundh, 1908.

A P P E N D I X.

1. THE OCCURRENCE OF THE DIAZO REACTION IN OTHER DISEASES BESIDES ENTERIC FEVER.

While investigating the course of the diazo reaction and its relation to Enteric Fever, it was considered advisable to carry out a series of experiments as to the presence of the reaction in other diseases. Observations were consequently made upon cases of typhus fever, pneumonia, cerebrospinal meningitis, scarlet fever, measles, German measles, chicken pox, and diphtheria.

Five cases of typhus fever were examined throughout, 3 of pneumonia and 7 of cerebrospinal fever. The behaviour of the reaction in these three diseases is specially important as they constitute a large portion of the cases between which and enteric fever, diagnosis is difficult. The result of this part of the investigation has been to show that there is no essential difference between the behaviour of the diazo reaction during the course of pneumonia and typhus fever, and that observed in enteric fever. In both of these the reaction is present during the course of the pyrexia, so far as observed, and in both it disappears when the temperature falls to normal.

It has been stated by Hewlett that the green deposit so often seen in enteric fever was confined to that disease but, in my experience, it is equally marked in typhus fever and pneumonia and in both persists till the crisis. There is, therefore, no aid in differential diagnosis to be obtained by the application of this method of examination.

In cerebrospinal fever, on the contrary, the reaction was never once observed. Out of seven cases examined throughout their course, the reaction was not once observed to be present even in the faint-

est degree.

Of the other diseases examined 25 consecutive cases were taken, in each case, with the exception of measles, which, being non epidemic in Glasgow at the time, was only admitted to hospital in sporadic cases three of which were examined from the commencement of the illness. These cases of all these diseases include all grades of severity with a considerable variety of the different complications. Only in the cases of measles was the reaction observed to occur. In these cases, during the acute stage it was very distinctly present and disappeared when the temperature reached normal, as seen in enteric fever, pneumonia and typhus fever.

The point of peculiarity in this disease, however, is the absence of the green deposit, which was not observed although the reaction was as intense as in many cases of enteric fever, where the sediment was considerable.

With regard to the other diseases nothing need be said beyond the fact that it was never present, although about 800 observations, in all, were made.

One point, however, is worthy of mention. Since the diazo reaction is present in measles and absent in German measles, it might be used as an aid in the differential diagnosis between these two conditions.

II. NOTE ON THE BACILLUS COLI COMMUNIS AND THE BACILLUS TYPHOSUS IN THE URINE.

It was stated by Wright and Semple that the bacillus typhosus was frequently absent from the faeces of patients suffering from typhoid fever. This has not been borne out by recent investigation. In fact, it is now practically certain that the infection of carrier cases is, in the majority of instances, at least, derived from the bowel and probably from the gall bladder.

Further, they have stated that when the bacillus typhosus is absent from the urine, the bacillus coli is, as a rule, present.

This statement which seems to be the solitary one in the literature so far as my survey of it has gone, is borne out by the results of my own investigations.

Of the 74 cases in which no typhoid bacilluria occurred, the bacillus coli was demonstrated to be regularly present. In the bulk of those cases it appeared much earlier in the disease than the bacillus typhosus and continued to be present in the urine for considerably longer periods.

It is also to be noted that these two organisms were never found associated together in a urine, the growth of either being always pure and, as a rule, luxuriant. These facts are certainly very curious but they suggest points in the theory of immunity and in the relationship of the growth of the organism upon which the investigations recorded in the paper do not throw much light and on which it seems unnecessary at present to theorise.

THE RELATION BETWEEN THE DIAZO REACTION AND BACILLURIA EXPERIMENTALLY DEMONSTRATED.

In order to further examine the relationship between the diazo reaction and bacilluria, experiments were carried out, in vitro, with specimens of normal urine. These were taken from 25 cases of enteric fever at different stages of the disease, sterilized by passing through a Pasteur filter and then inoculated with typhoid bacilli.

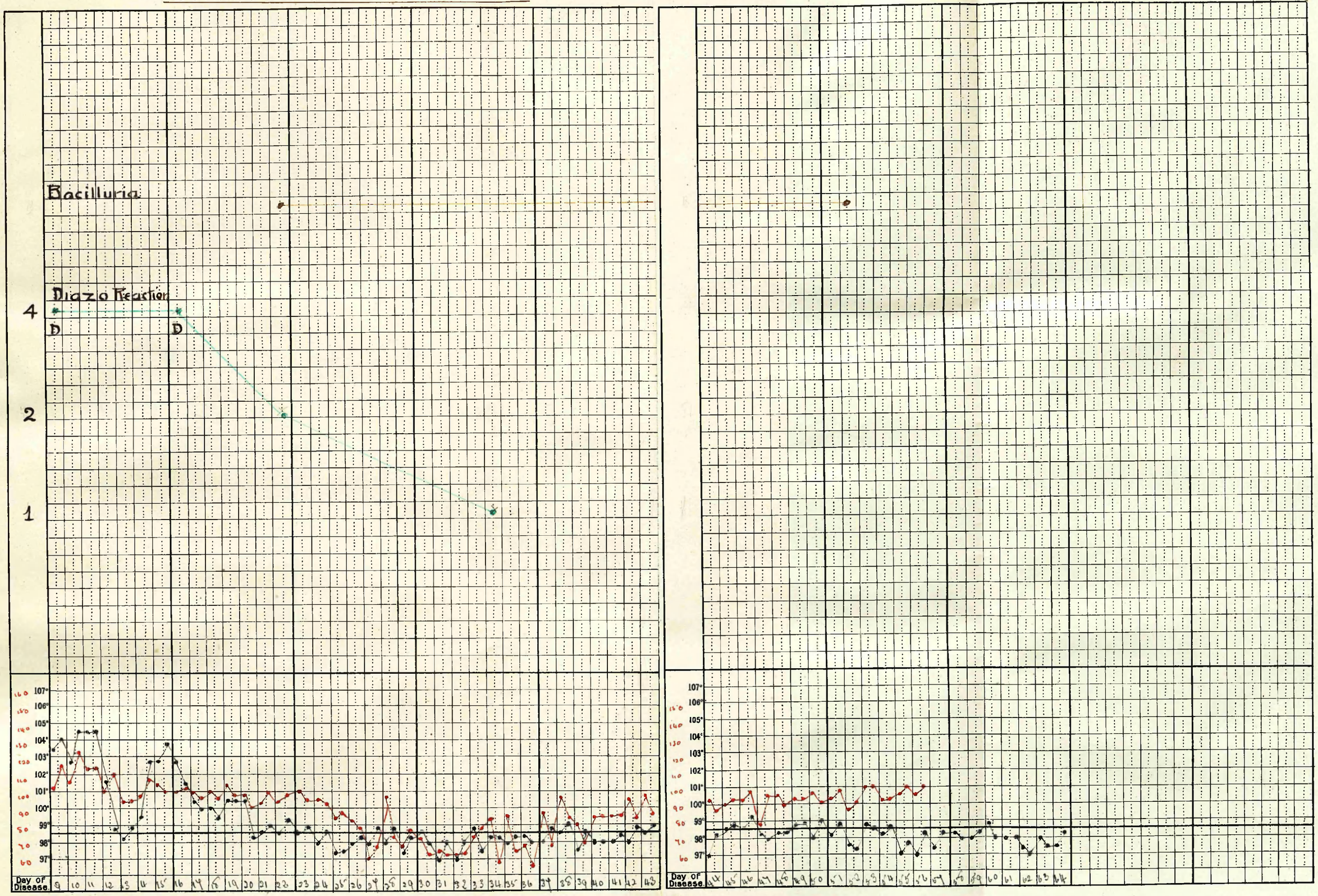
The specimens of urine were afterwards incubated at a temperature of 37°C, for periods varying from 1 to 3 days at the end of which they were tested for the diazo reaction.

In not a single instance was there any evidence of the presence of the reaction.

It may be mentioned here that in the majority of cases the urine was rendered distinctly turbid from the growth of the organism, a condition referred to earlier in the paper.

CHART XIII

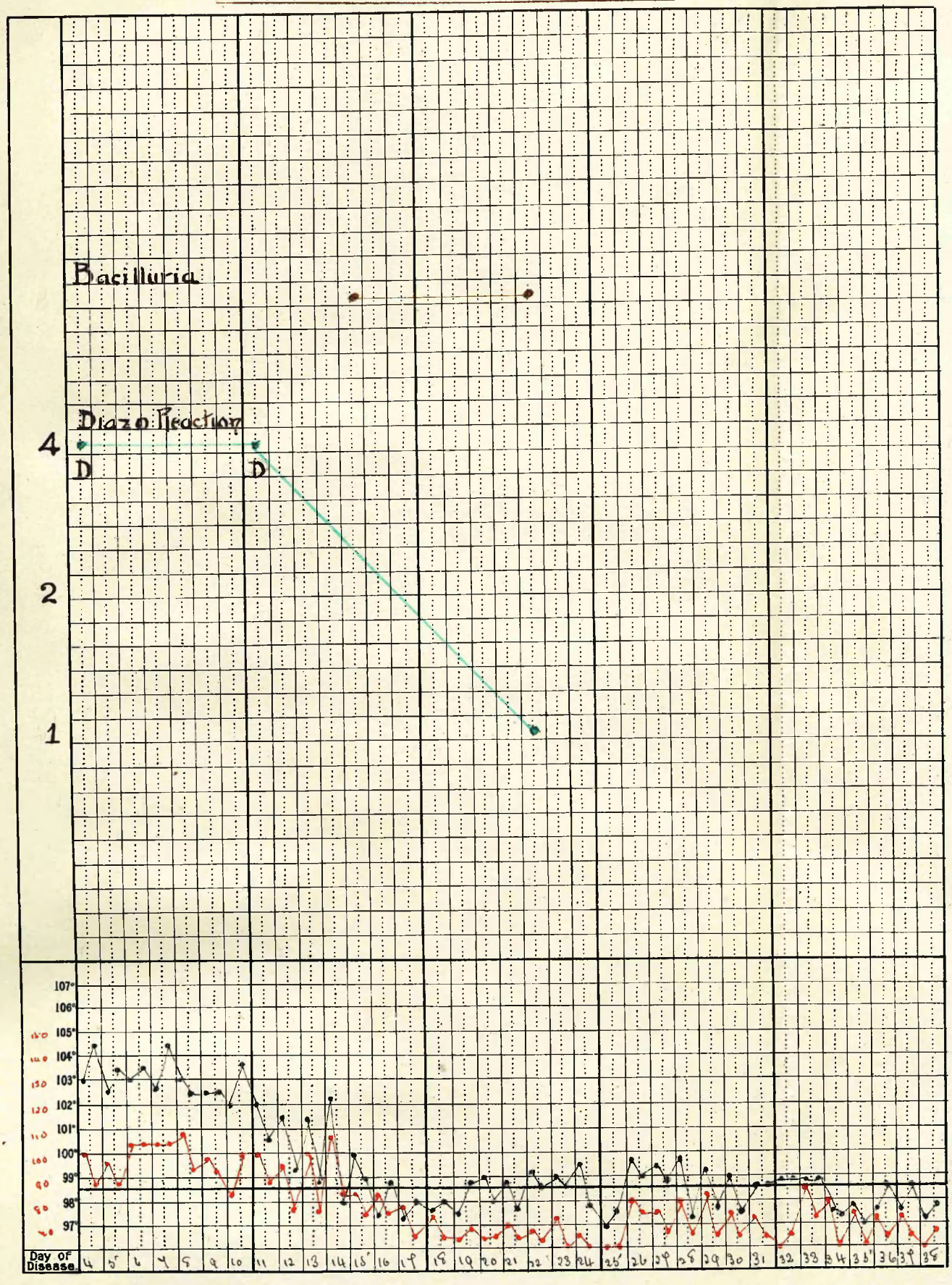
J. McN MALE AGE 12.



D - D = Duration of Deposit

CHART XIV

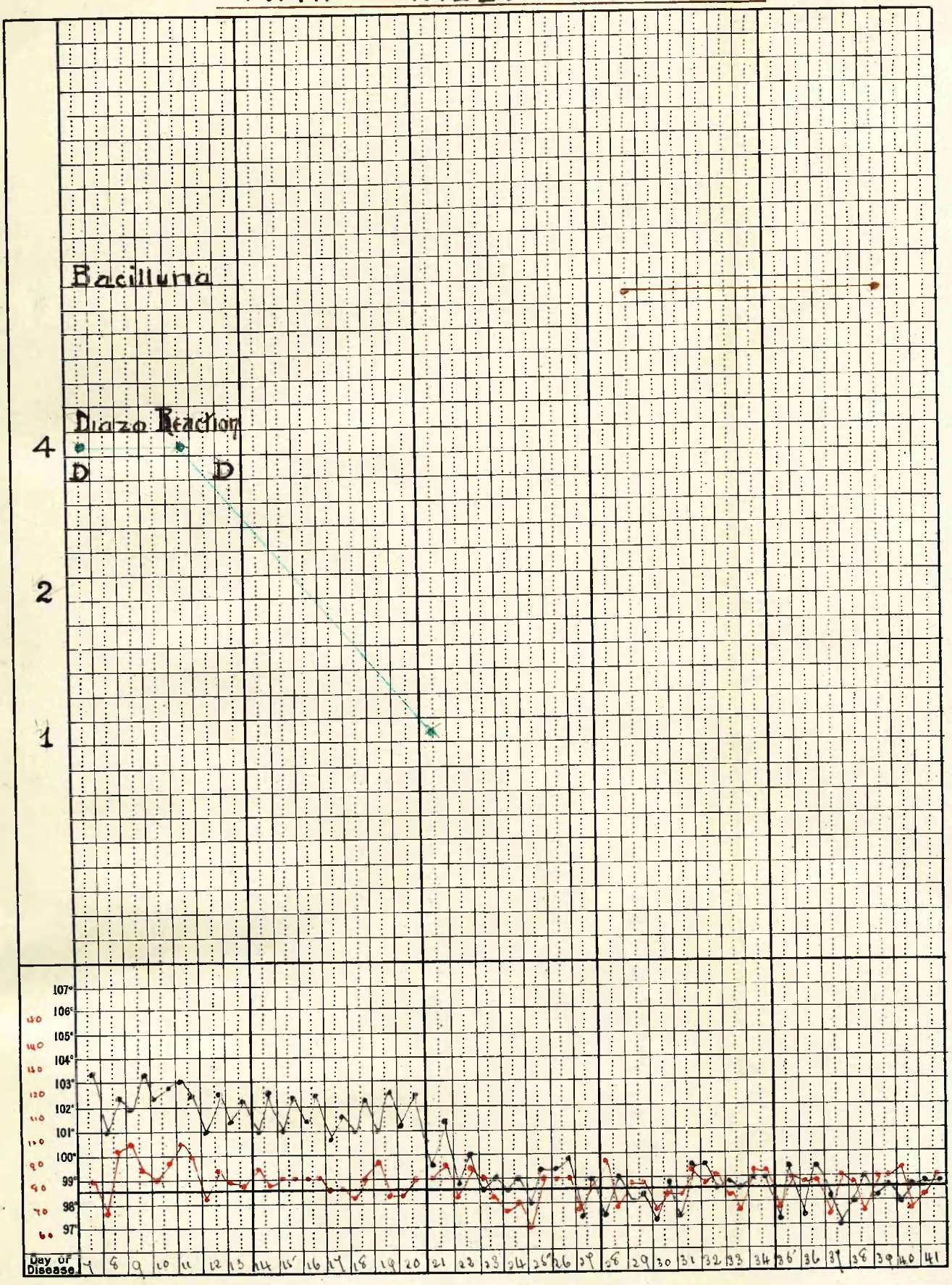
T.P. MALE. AGE. 30.



D-D = Duration of Deposit

CHART XV

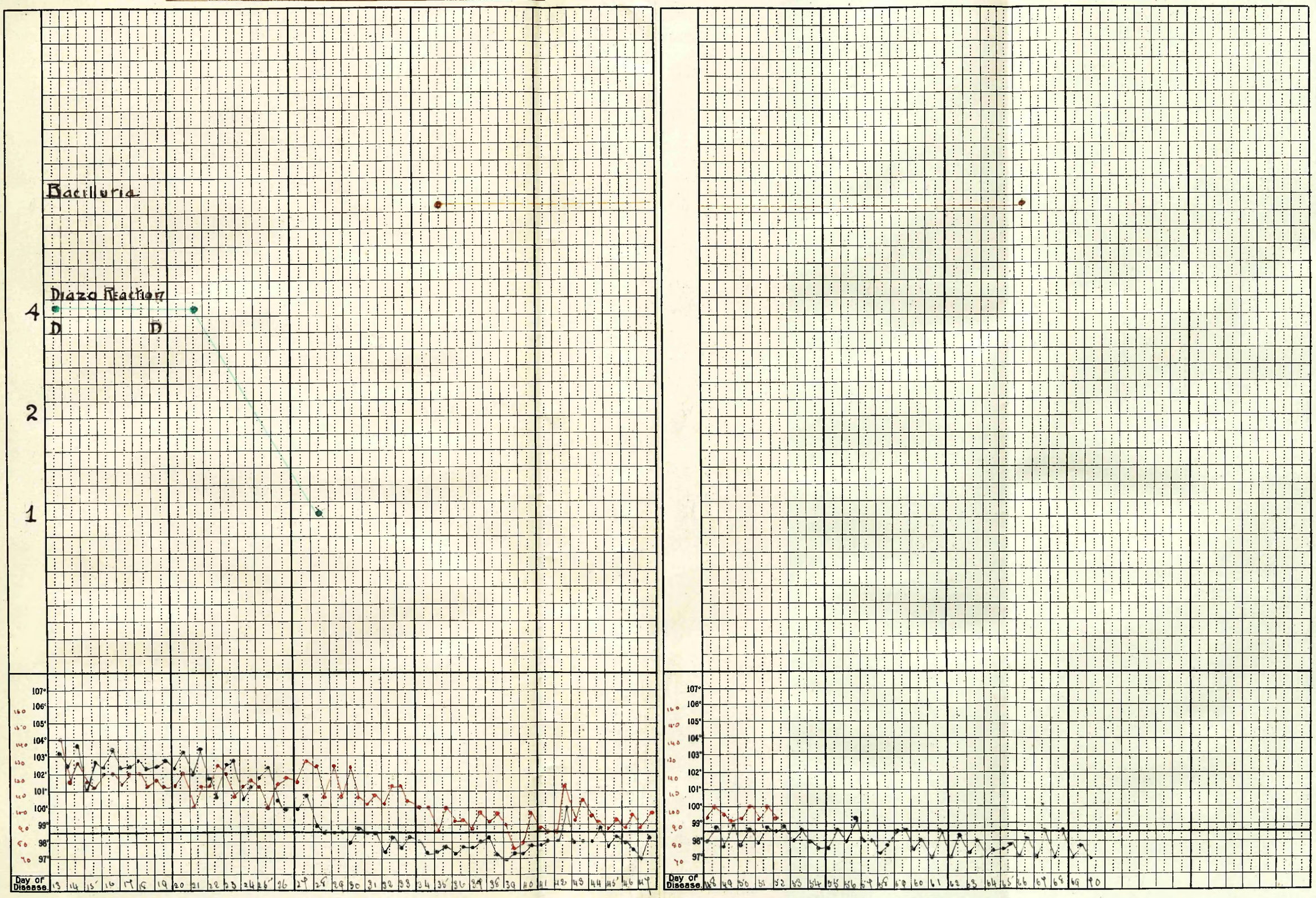
W. M. MALE. AGE 21.



D-D = Duration of Deposit

CHART XVI

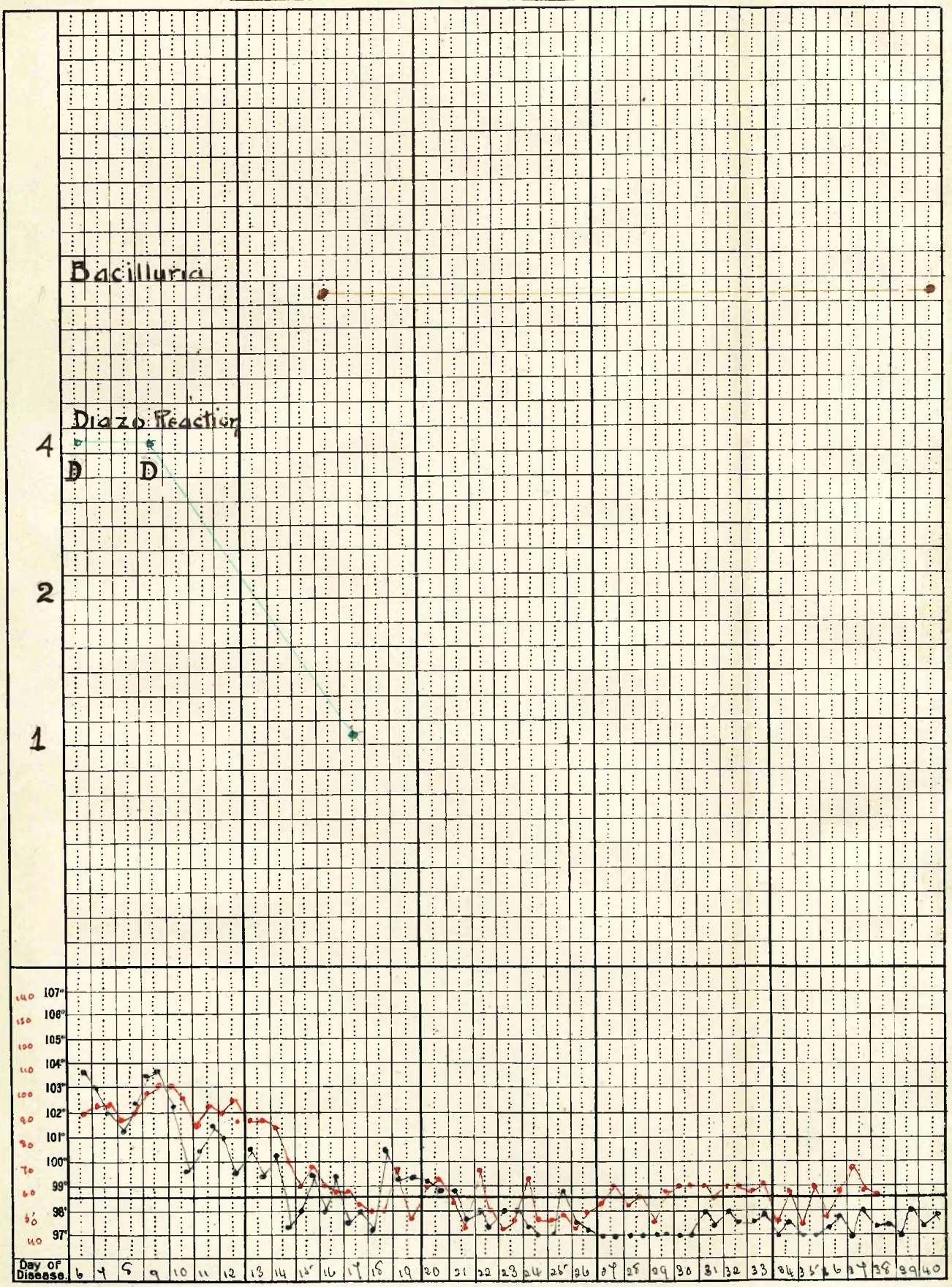
R. McN. FEMALE. AGE. 18.



D-D = Duration of Deposit.

CHART XVII

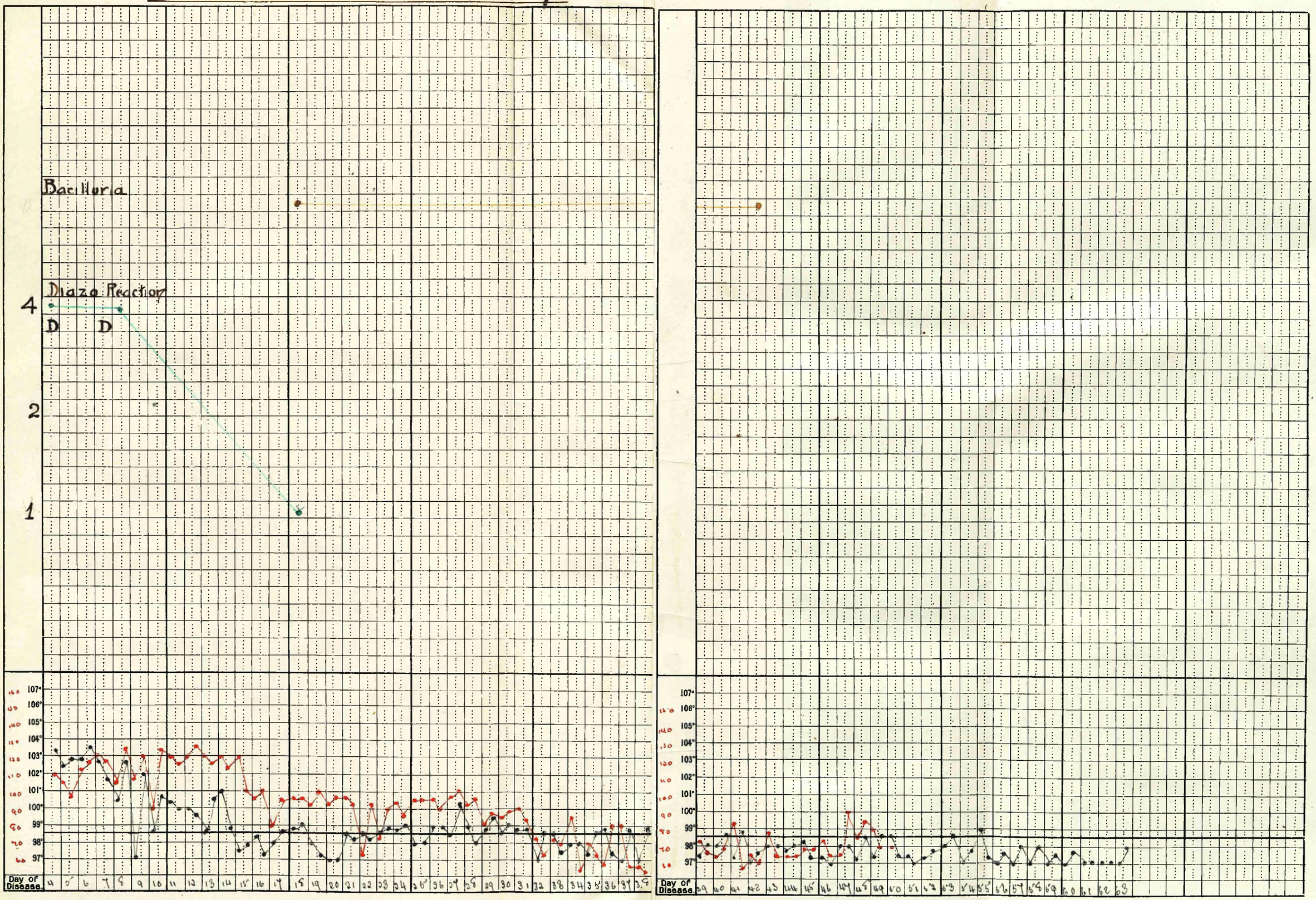
W. McA. MALE. AGE 29.



D-D = Duration of Deposit.

CHART XVIII

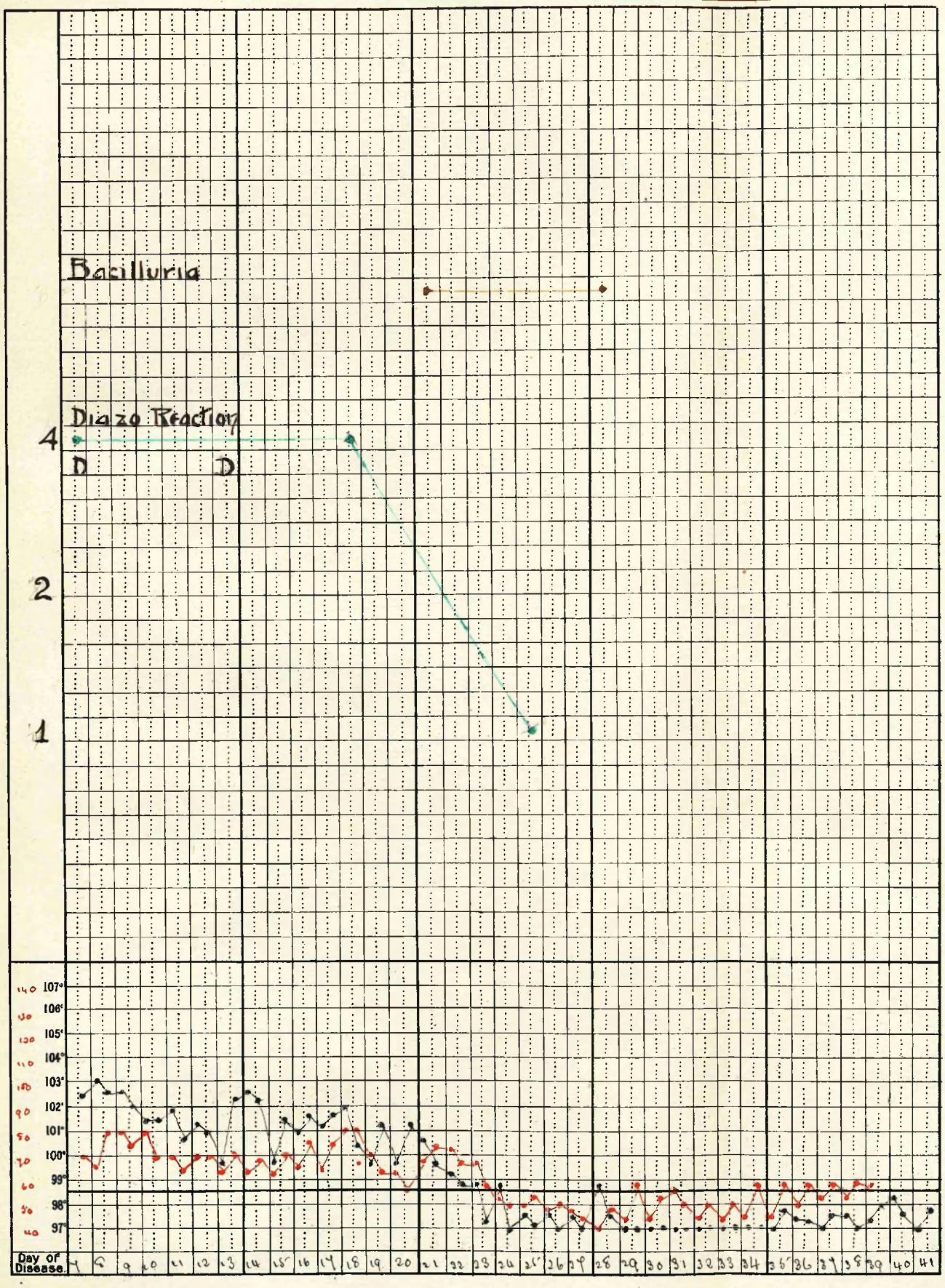
J.W. MALE. AGE. 17.



D-D = Duration of Deposit.

CHART XIX

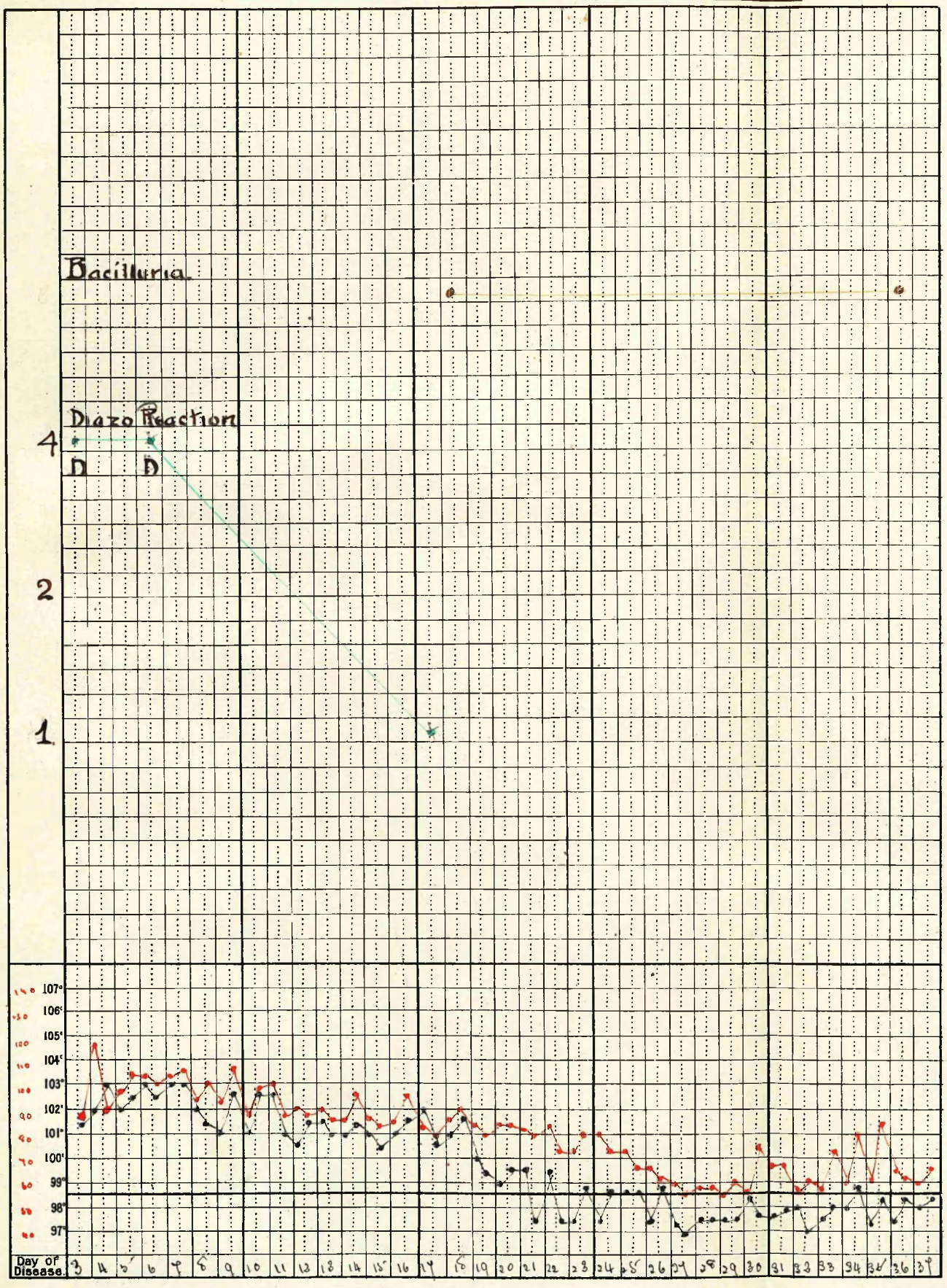
W McC. MALE AGE 22.



D-D= Duration of Deposit.

CHART XX

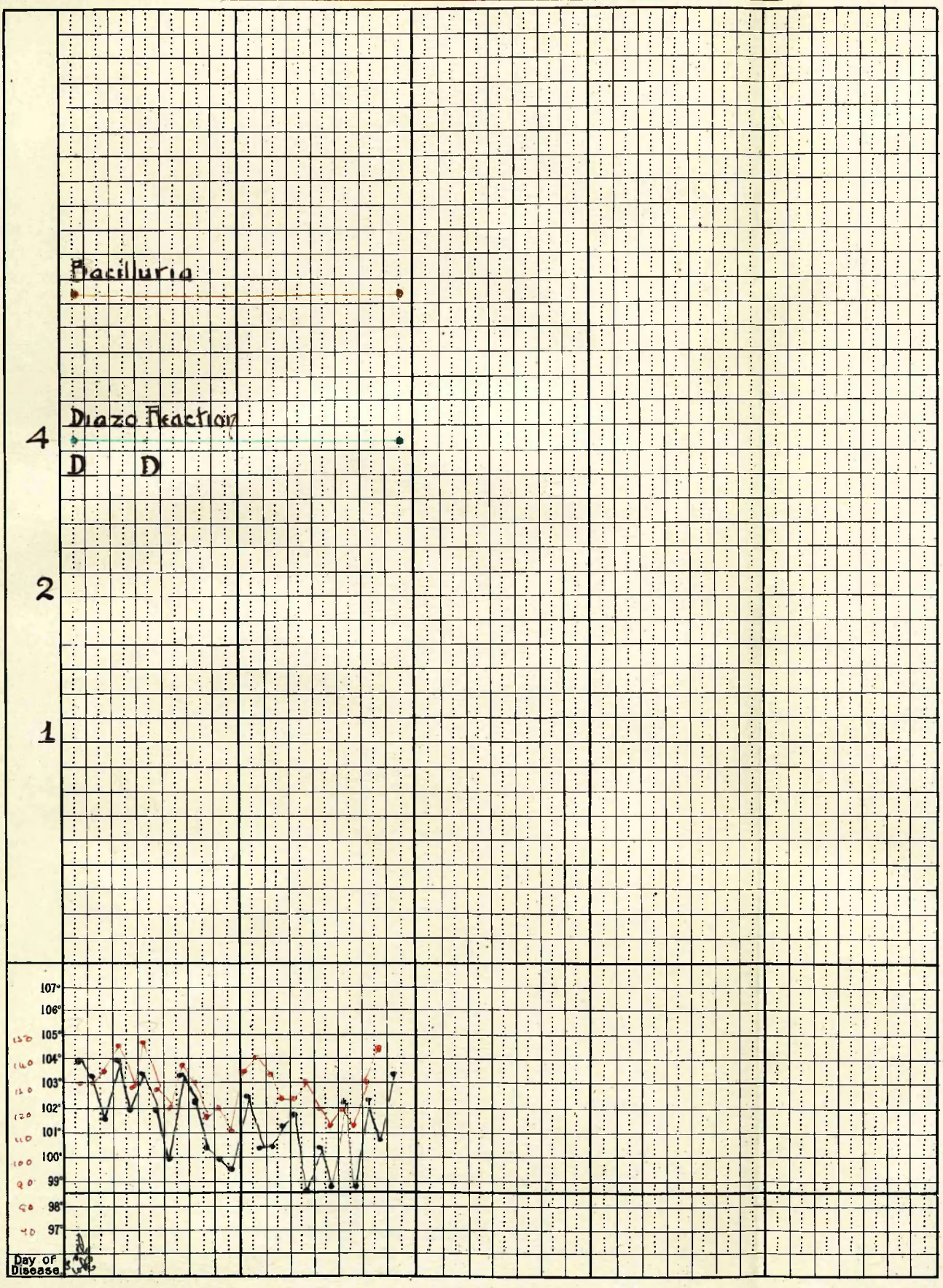
T.C. MALE. AGE. 21.



D-D = Duration of Deposit

CHART XXI

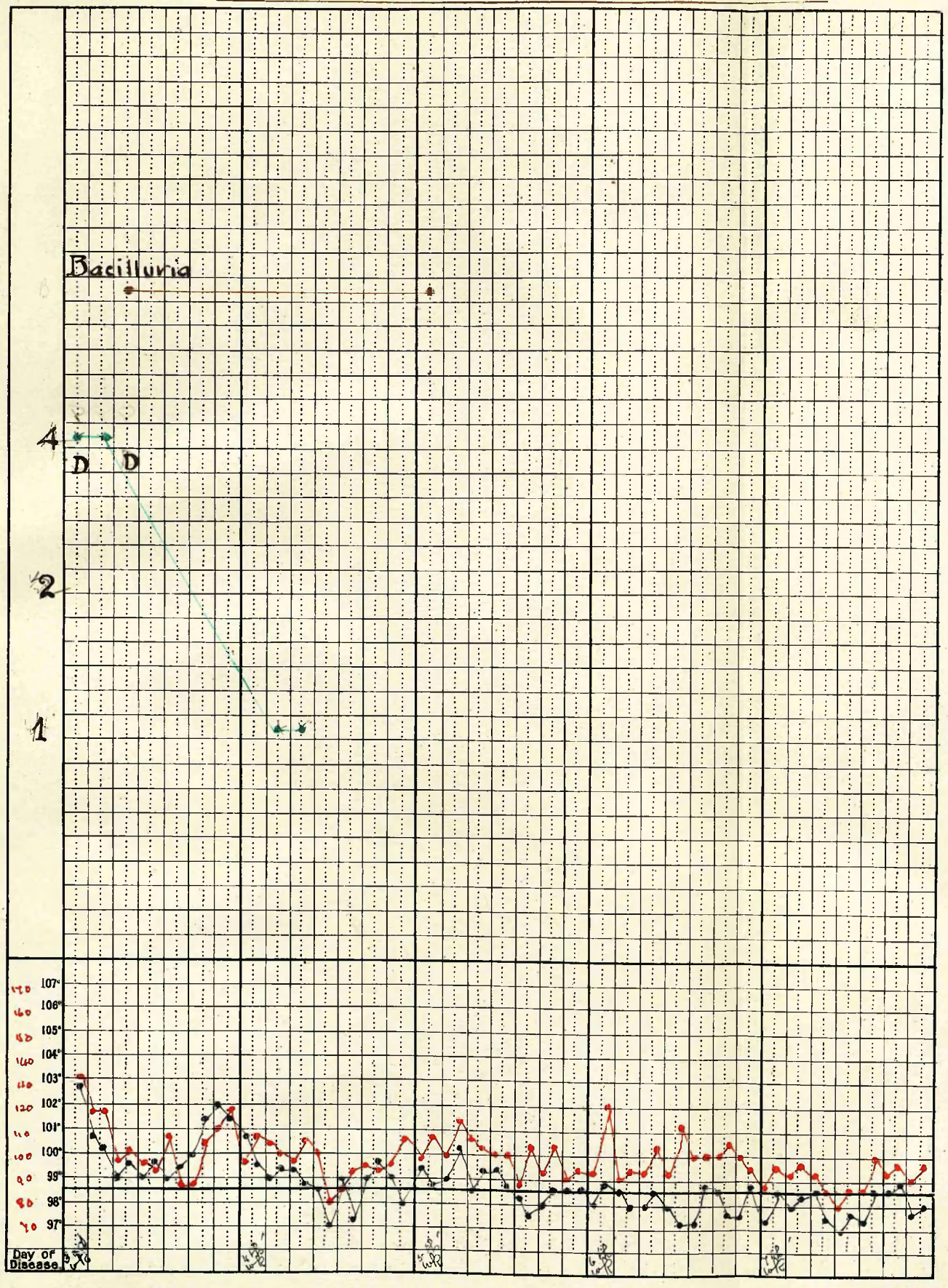
M.M.C. FEMALE AGE 26



D-D = Duration of Deposit.

CHART XXII

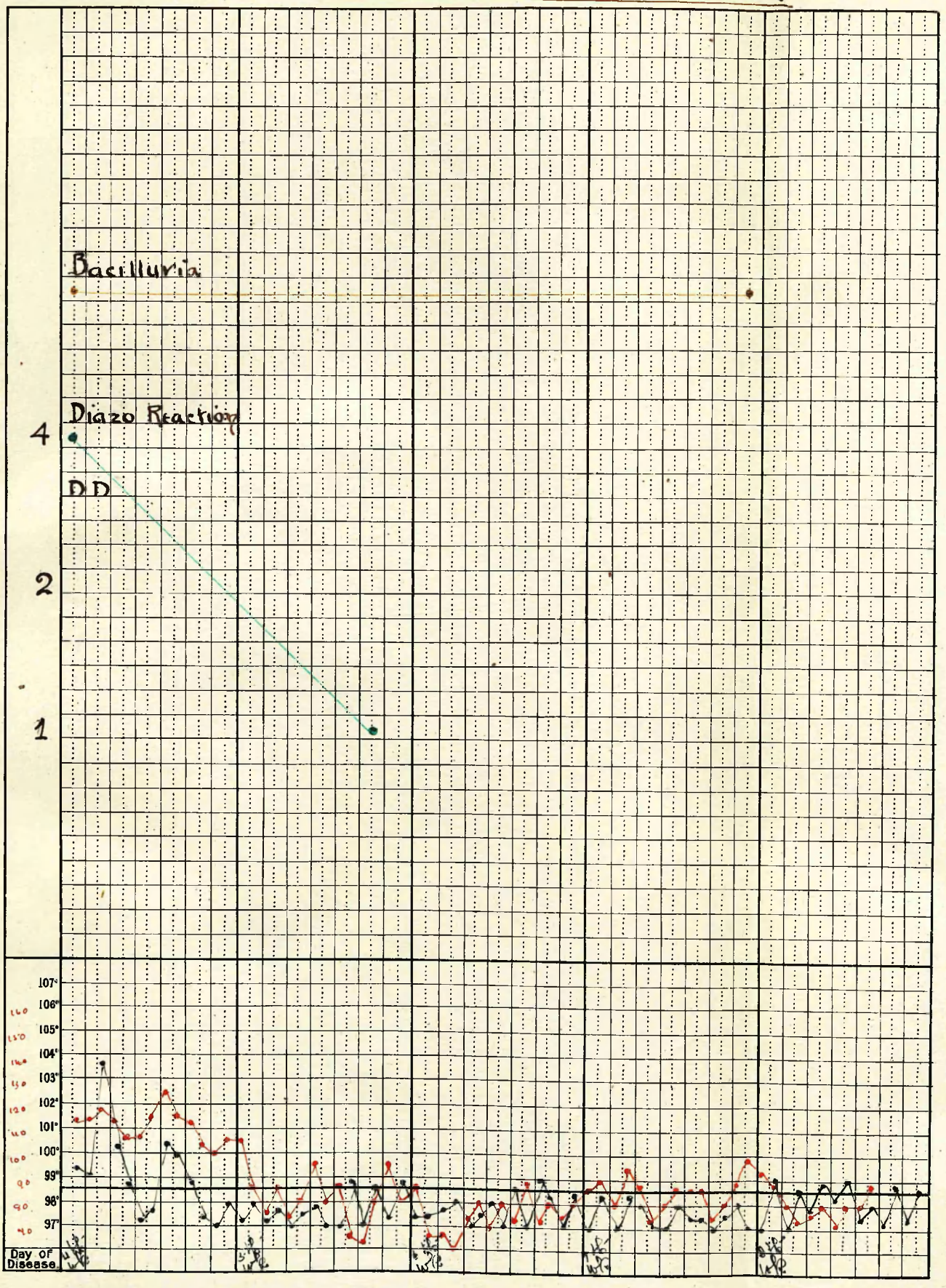
A.S. FEMALE AGE 18.



D-D=Duration of Deposit.

CHART XXIII

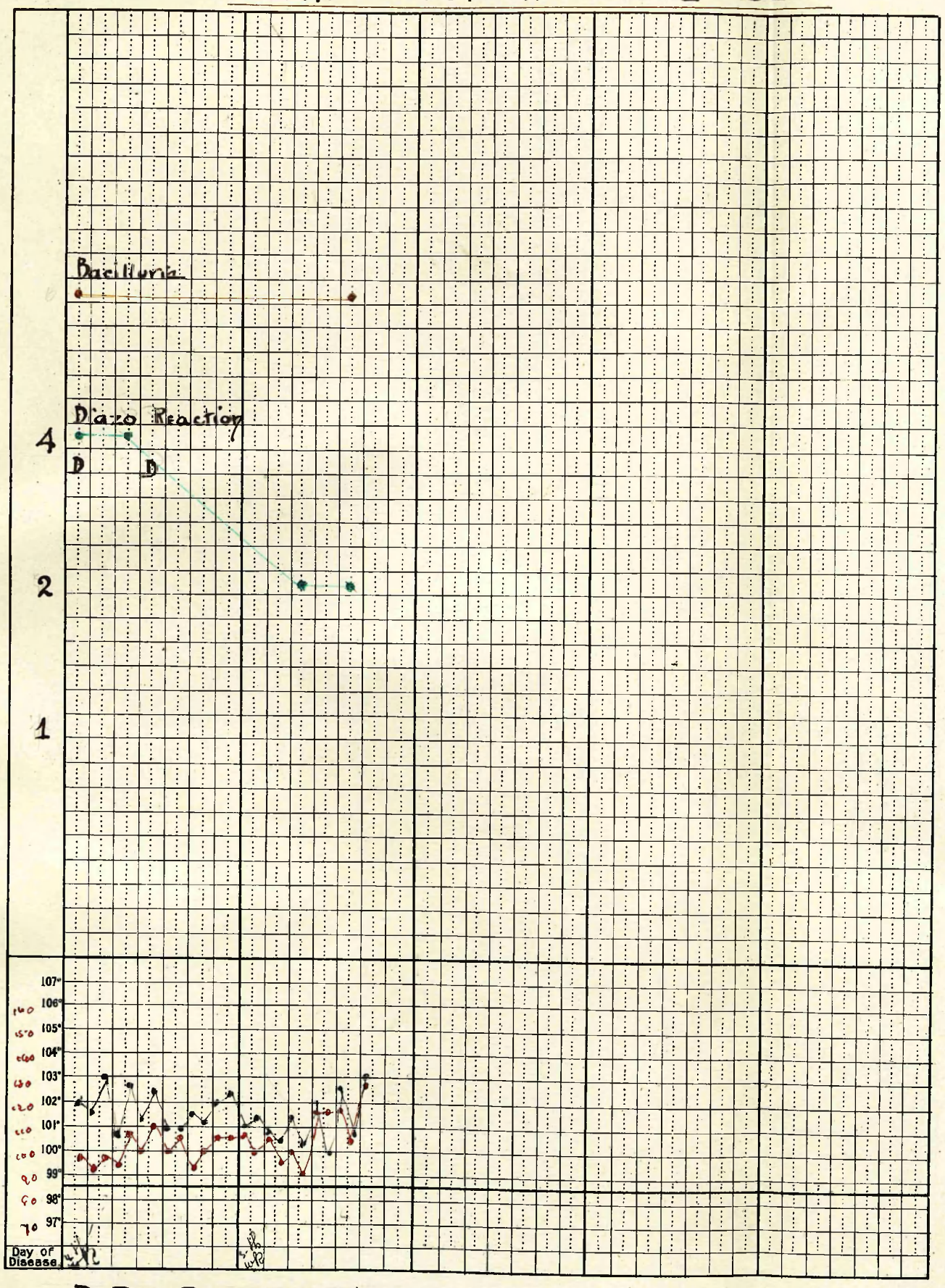
M. K. FEMALE. AGE. 30.



D-D = Duration of Deposit.

CHART XXIV

A. M. FEMALE AGE 35



D-D = Duration of Deposit.