THE DIAZO REACTION IN TUBERCULOSIS.

By CHAS. W. BUDDEN, M.B., CH.B. VICT.,

Late Assistant Resident Medical Officer, Walton Workhouse, West Derby Union, Liverpool.

THE urine test, based upon the chemical fact that if sulphanilic acid be acted upon by nitrous acid, diazo-sulpho-benzol is formed which unites with certain aromatic compounds occasionally present in urine to yield aniline colours, and known as Ehrlich's diazo reaction, was first brought forward as a valuable test for the recognition of typhoid fever. It was soon, however, found to be of very variable occurrence, and to be present in other conditions of which tuberculosis stood pre-eminent, and an effort was made to prove that the diazo reaction was of assistance in the diagnosis of tuberculous affections, and in the prognosis of that disease. At present the most diverse opinions prevail as to the behaviour and clinical value of this test, and the most conflicting statements are published from time to time of the results obtained by various observers. I believe that this is due in great part to the paucity of cases recorded by each writer, and to the many unorthodox ways of performing the reaction, since it is common knowledge that experimentalists of every age have failed because they had not discovered laws which govern variations. I have watched the reaction in three thousand persons, and performed it many times in each, endeavouring to follow each case to its conclusion before allowing myself to be influenced by the results. The record of this work, of necessity greatly epitomized, is the subject of the present paper.

It is necessary to first detail the manner of carrying out the test, since want of strict attention to this point has been the cause of many erroneous conclusions. Ehrlich prescribed the following reagents:

1. A saturated solution of sulphanilic acid (crystals) in 5 per cent. HCl.
2. A $\frac{1}{2}$ per cent. solution of pure nitrite of sodium.
3. Liquor ammon. fort.

To be employed thus: A mixture containing the sulphanilic acid solution and the solution of sodium nitrite, in the proportion of 40 c.cm. of the former to 1 c.cm. of the latter, is measured in a graduated tube, and 10 c.cm. of it added to an equal quantity of urine in a test tube. After shaking, so as to effect a thorough admixture, 1 drop of ammonia is allowed to trickle down the side of the test tube. If the reaction occurs, the following phenomena are to be observed:

1. A red ring forms at the junction of the ammonia with the rest of the fluid.
2. On inverting the test tube, so as to mix the ammonia with the urine, the whole will assume a red colour.
3. On violently shaking the tube, with the thumb held over the mouth, so as to cause a froth to form, this red colour, varying from strawberry pink to deep crimson, will be communicated to the froth.
4. After allowing the specimen to stand for twenty-four hours a sediment of a green or violet colour will have settled to the bottom of the test tube.

Ehrlich specially emphasized the point that the reaction is not positive unless the pink or crimson colour is communicated to the froth, and further stated that the presence or absence of the green precipitate was of little moment.

The test practically resolves itself, therefore, into the accurate observance of the froth, so that I have adopted the method of shaking the specimen before the addition of the ammonia, and allowing the latter to trickle directly on to the foam, when the colour struck is readily noticed, since it shows up against the white of the froth. I have found also that one or two precautions are required to obtain accurate and uniform results. They may be briefly stated:

1. The test cannot be performed except during the day, or by the use of pure white light.
2. The urine should not be more than twenty-four hours old, and should be kept covered.
3. Both the sulphanilic acid and the sodium nitrite solutions are very unstable, and deteriorate rapidly. Fresh solutions must be employed. The nitrite should be renewed weekly, the acid once a month. We do not mean to say that the reaction will not occur with older chemicals, but we positively affirm that its appearance will be variable.
4. The proportions of sulphanilic acid and sodium nitrite require accurate measurement. An excess of the latter, even in so slight an amount as 2 c.cm. in 40 c.cm., will annul a delicate test.

Having thus stated the method of performing the reaction, I may pass on to the review of the work, of which the end has been to ascertain the value of Ehrlich's test in the diagnosis and prognosis of tuberculous disease. The 3,000 cases in which the behaviour of the reaction has been followed comprised 600 cases of persons in health and 2,400 of diseased states. Of those in health not one yielded a positive reaction,

from which I at once conclude it never to be present in normal urine. Of the remaining 2,400 cases I found the following number of positive reactions:

1. Various forms of tuberculosis ...	96	positive cases out of 672
2. Measles ...	18	" " 25
3. Enteric ...	17	" " 21
4. Typhus ...	5	" " 5
5. Acute lobar pneumonia ...	4	" " 41
6. Acute bronchopneumonia ...	1	" " 16
7. Acute bronchitis ...	1	" " 96
8. Puerperal septicaemia ...	4	" " 4

The remaining 564 cases, comprising almost every acute malady of common occurrence in this country, continuously yielded a negative test. Other writers have declared its presence in erysipelas, intermittent fever, surgical septicaemia, and some other conditions, but their reports are very variable. Sacquépée, of Paris, observed it in puerperal septicaemia, and other authorities have found it more or less frequent in measles and typhus fever. We may suppose, then, that its presence is fairly constant in these three conditions, leaving its behaviour in typhoid fever out of account, since in that disease its conduct has been frequently investigated, and I have made no fresh observations. There still remain the cases of pneumonia and bronchitis to account for. These will be referred to later. Suffice it for the present that each was complicated with an underlying phthisis.

It is not sufficient, however, to have merely recorded the presence of the positive reaction in certain diseases; the clinical progress of each case has therefore been followed with reference to the behaviour of the reaction from time to time, with a view to ascertaining if its presence or absence has any bearing upon prognosis, as has been claimed by several writers. Very early in the research it was discovered that the positive reaction appeared very variably in the several cases of tuberculosis under observation. Many cases never yielded it; others did so at intervals of days or weeks, while it was at once obvious that its presence coincided with exacerbations of the disease. This was remarked by Ehrlich. I realized, therefore, that if the results were to have any value, the testings must be done frequently with each patient, and a record kept of their clinical condition. Accordingly, I performed the test in many cases every day the whole time they were under investigation, and kept a constant record of their clinical progress and physical signs. No case was tested less frequently than twice a week. By this means I have avoided falling into several errors which must be inevitable under any other mode of investigation.

A. THE VALUE OF THE REACTION IN THE DIAGNOSIS OF TUBERCULOUS CONDITIONS.

In the summarized statistics at the opening of this record I have stated that the number of tuberculous cases investigated amounted to 672. These are made up as follows:

Disease.	Total No. of Cases.	No. of Cases Positive.	No. of Cases Negative.
Acute miliary tuberculosis...	2	2	0
Chronic pulmonary tuberculosis...	628	83	545
Tuberculous meningitis ...	6	6	0
" peritonitis ...	3	3	0
" adenitis... ..	12	1	11
" bones and joints ...	21	0	21

The question at once arises, Did the presence or absence of the positive reaction prove of any assistance in the diagnosis of these cases? I shall endeavour to answer this by discussing the several groups singly.

1. Chronic Pulmonary Tuberculosis.

From the tabulated records of the 83 positive cases I find that not one was in the early stages of phthisis, when a diagnosis might be difficult, but that in every instance there were abundant physical signs of advanced mischief. In this I agree with other investigators. Bolli found the reaction present in the second and third stages of pulmonary tuberculosis, and was supported in this by Blad, Videbeck, and Michaelis. The later investigations of de Boissière and Cummins are further confirmatory of the fact. My own cases may be thus summarized:

Unilateral apical consolidation ...	9 cases
Bilateral " " ...	12 "
Unilateral vomica... ..	23 "
Bilateral vomicae ...	39 "

When these physical signs were taken in conjunction with the usual clinical picture of phthisis, no urine test was needed to convince one as to the diagnosis. So far, then, its value is *nil*. On the other hand I believe it to be of assistance in those cases in which the tuberculous mischief is hidden under some other acute process. I have referred, for example, to the fact that the positive reaction was elicited in 4 cases of acute lobar pneumonia, in 1 case of acute bronchopneumonia, and in 1 of acute bronchitis, and that in each of these cases there was afterwards found to be an underlying tuberculous lesion. The reaction faded and then disappeared in each instance, upon the recovery of the patient from the intercurrent affection, and in 2 cases it was observed to appear with the pneumonia. Petri records the case of a phthisical girl who gave a positive reaction during an attack of acute follicular tonsillitis, so that it would appear that not only does the reaction depend upon the exacerbations of the tuberculous process (as observed), but that its presence may be also determined by the onset of any acute illness upon an existing tuberculous lesion. This may explain its presence in such diseases as erysipelas and so forth, and may have misled the experimentalist in thinking it indicative of that and other diseases. In cases such as these the presence of the positive reaction may be of diagnostic value. Chronic or acute bronchial catarrh, for instance, often covers tuberculous signs. If, then, the reaction is obtained in these conditions, it should at once lead one to suspect tuberculous mischief. Considering the large number of cases tested, and that every one yielding a positive reaction was tuberculous (excepting those diseases cited, measles, typhoid, typhus and puerperal septicaemia), I feel justified in concluding that there must be some hidden tuberculous process at work in any case yielding the positive reaction not known to be in the category of those diseases which give it.

2. Tuberculous Pleurisy and Peritonitis.

I have not had under observation any case of this condition distinct from phthisis—that is to say, none which could be affirmed to be tuberculous. From a total of 46 cases of pleurisy, 16 were obviously tuberculous, being accompanied by other signs and symptoms, and of these only 3 gave a positive reaction, and that because they were accompanied by advanced apical mischief. For this reason they are not grouped separately. No reaction was obtainable in any of those cases where one was in doubt as to the nature of the process. Bolli found the reaction present in 6 cases out of 12, and noted that the disappearance of the characteristic colour coincided with the absorption of the pleuritic effusion. He obtained similar results in 8 cases of tuberculous peritonitis. I have not watched a sufficient number of cases of either to justify any new conclusion. While doubtful if the reaction can have any value in the diagnosis of either condition, I believe it only to occur in pleurisy and peritonitis of a tuberculous nature.

3. Tuberculous Meningitis.

Of 6 cases of this disease every one gave a strongly-marked positive reaction. In 5 of the cases the diagnosis was made, or proved, by *post-mortem* examination; in the sixth, the patient was a child presenting typical symptoms. This is mentioned because no evidence could have any value without an autopsy to corroborate the record. It is interesting to know that two of the cases were regarded as typhoid fever, the presence of the diazo reaction being taken as confirmatory of this view, nor was the possibility of meningitis fully realized until the *post-mortem* examination proved negative, and the brain was exposed accordingly. It is a matter of regret that the reaction should be most constant in an affection which often closely resembles enteric fever, and that it is on this account of no value in the differential diagnosis of the two complaints. Indeed, I find it present frequently in other conditions resembling these—namely, typhus fever, acute miliary tuberculosis, and acute lobar pneumonia in a phthisical subject, precisely the very group of diseases which, in certain cases, may be mistaken for each other.

4. Tuberculous Adenitis.

Of 12 cases of tuberculous disease of the lymphatic glands one only yielded a positive diazo reaction—a boy suffering from glandular enlargements in both axilla, round the neck, both groins, and in the anterior mediastinum. It is to be regretted that the patient was of a morose and sullen temper, refusing treatment and full examination, so that no further record has been kept. Faint inspiratory crepitations at one pulmonary apex, with the presence of the diazo reaction, led

to the conclusion that the patient was tuberculous. As in the case of pleurisy and peritonitis the paucity of cases prevents any conclusions, but the question is left open as to whether the reaction might not prove of occasional service in differentiating the various glandular affections.

B. THE VALUE OF THE REACTION IN THE PROGNOSIS OF CHRONIC PULMONARY TUBERCULOSIS.

This is now considered as the most important feature in the behaviour of the diazo reaction, and it has been the goal towards which the energies of most of its investigators have been directed. A few of their conclusions may be briefly indicated. Bolli looked upon the presence of the reaction in the early stages of phthisis as not necessarily of bad import, and its absence in the latter stages as indicative of a less rapid course. Michaelis made observations upon 167 phthisical patients, of whom 111 gave a positive reaction, and 56 a negative one. Of the 111, none were cured, 15 were improved, 13 remained unchanged, while 30 died; whereas of the 56 negative cases, 5 were cured, 44 improved, 4 remained unchanged, and only 3 died. Blad and Videbeck also worked at the same subject, their results agreeing with those of Bolli, and with the records of Lorentz at the Halla Sanatorium. Their tabulated results show that, in general, the reaction is negative, or if present, weak and inconstant, in those cases which clinically appear favourable. On the other hand, it was positive and well marked in bad cases. They agreed that patients yielding the reaction were not suitable for sanatorium treatment. Friedenwald considered the positive reaction justified a gloomy prognosis. Nissen believes that the more constant its presence the more acute and extensive is the tuberculous process. Raoul de Boissière examined 130 cases of chronic phthisis in the Edinburgh Hospital for Consumption, of which 18 were found to give a positive reaction. Thirteen of these died, 1 lay in *extremis*, and 2 recovered. He found that the presence of the positive reaction coincided with the appearance of the graver symptoms, and concluded that its appearance was of bad import, and its disappearance a good omen. Cummins examined 436 urines, of which tuberculosis cases numbered 221; 67 of these responded to the test. They were tested periodically, and led him to the following conclusions, namely, "That the presence of the positive reaction has great prognostic significance in pulmonary tuberculosis. It does not appear till late, and is usually noted in a rapid case. When it once appears, it persists until death. The average duration of life after the appearance of the diazo reaction is about six months. An unfavourable prognosis should be given in every patient presenting this reaction." Ehrlich himself looked upon its appearance as a bad portent, while, on the other hand, Petri, Georgiewsky, Becker, Giesler, and others declare the reaction to have no significance in the prognosis of phthisis.

My own opinion is, as has been stated at the opening of this paper, that this variableness of results has depended upon too infrequent testings, so that I would again emphasize the fact that I have followed the behaviour of the reaction in every case at least twice a week, and often daily throughout their course.

It has been said that of the total number of phthisical patients examined, 83 yielded a positive test, and that every one of these was in the second or third stage of the disease. To complete their clinical picture we would further add that 67 ran an evening temperature of over 100°; 32 of these being intermittent, 15 remittent, and 7 continuous. Of the remaining cases 5 had a swinging temperature of between 99° and 100°, and 10 were apyrexia.

The 545 negative cases present a contrast to this. Of them 386 were in the earliest stages of phthisis, 111 presented unilateral consolidation, and 33 bilateral; 12 presented unilateral cavities, and 9 had bilateral vomicae. With regard to their temperature, 436 were apyrexia, 12 ran up between 99° and 100°, and 63 had a swinging temperature of over 100°.

Of the 83 positive cases, 60 died, 6 were unchanged, while 17 may be said to have shown slight improvement. Of the negative cases, 17 died (6 of intercurrent affections), 30 were unchanged, and 454 improved.

Several other facts were also noticed. Forty-eight of the positive cases showed a gradual fading of the reaction as they drew near to their end, and ultimately ceased to yield it at all, sometimes as long as months before death; generally it ceased on the day of death. In several cases the urine was drawn off *post mortem* by means of an aspirating needle introduced above the pubis, but a positive reaction was never obtained. Petri performed the same experiment, but never

found a positive reaction after death. Ehrlich first noticed this *ante-mortem* fading of the reaction, but it has since been contradicted by some writers. That it really does fade is a fact. Those declaring the contrary have simply not met with the cases.

Petri considered a concentrated state of the urine to be necessary to the production of the characteristic positive tint. With a view to testing the accuracy of this observation, I have recorded the specific gravity of every case, and found that of the sum total of positive cases (including measles, typhoid, etc.) the average specific gravity was 1025. Only in 6 cases was it less than 1016; but in reference to this point we have to consider that the concentration of the urine is largely dependent upon the temperature of the patient previous to and at the time of voiding, and, in the case of phthisis, upon night sweats. Those with the highest temperatures and the most profuse sweats are generally the worst cases, and therefore the class in which we may expect the reaction. In the same way the reaction cannot be wholly dependent upon pyrexia, as some have affirmed, since it is often present in apyrexial cases.

I have further observed that those cases giving the reaction to the end have run a more rapid course, and have died in a less exhausted condition than those who had once given the reaction; and then had ceased to do so. This I believe to be the cause of the constant affirmation that a diazo patient had the worst outlook, and I believe that writers who have made this statement have not met with cases which ceased to yield the reaction a month or six weeks prior to death. There seems, in short, to be required a certain amount of vitality for the production of the characteristic aniline colour of Ehrlich's reaction. This is borne out in a remarkable way by the following case:

G. C., aged 20. Admitted to Walton Workhouse, December 16th, 1901. Temperature 97.6°, pulse 168, respirations 56. In extreme exhaustion. A large vomica at right apex. Diazo negative. Patient was so ill that no expectation of his surviving the night was felt. He rallied, however, and showed marked improvement the next and four following days. On December 21st, that is five days after admission, he gave a well-marked reaction. A day or two later he began to sink; his temperature, which had gone up to 100°, fell to subnormal again, the diazo reaction faded, and he died on the 29th of the same month.

We further have to record a very singular case in which the patient, although in the last stages of phthisis, failed to give the reaction. He was under observation for a month, during which time his urine was tested daily. On the last day of the month he died from hæmoptysis, and in idle curiosity I set the blood aside in a measure glass, decanted the serum and subjected it to the diazo test. To my astonishment it yielded a rich reaction. Encouraged by this event I repeated the experiment in 6 other cases of hæmoptysis, but did not again succeed in eliciting the reaction. I aspirated the heart immediately death occurred in a large number of cases, but never obtained a positive coloration. My attempts to get it in pleuritic effusions also failed.

CONCLUSIONS.

The conclusions I draw from this investigation are that the reaction is not of any value in the prognosis of chronic tuberculosis. It is certainly true that those patients who gave the reaction have nearly all died; it is certain that in most cases the course was rapid; it is also true that its appearance and disappearance corresponded to the exacerbations and to improvement; it is also a fact that those not giving the reaction are patients who have shown improvement in their clinical course. This, to a large extent, agrees with what has been before written. Were this all, such facts would indeed be significant, but they reveal only half the truth. The positive cases have died, but they have been precisely those cases for whom no hope could ever be entertained; they have run, many of them, a rapid course, but an equal number have run a slow course and even improved; its appearance coincides with the exacerbations of the tuberculous process, but it follows the onset of the graver symptoms, and though it disappears with improvement, such disappearance again follows the suspension of the alarming signs; while, again, it may disappear because the patient is growing weaker. A reaction blowing hot and cold in this manner cannot be of very great reliability. Lastly, patients who have not given the reaction have mostly been in the early stages when hope is justifiable, while if it is absent in the later stages it means nothing, since its appearance is intermittent: is present one week, absent the next.

It is probable that every fatal case of tuberculosis will yield the reaction at some period of its course. In view of the

frequency of the intermissions, we cannot say that any case is negative unless it has been tested constantly for long periods. Of the 17 cases which were negative and yet died, 6 succumbed to intercurrent diseases. Of the remainder, 2 were admitted *in extremis*, when the reaction is known to fade, so that it is not an unjustifiable conclusion to suppose that the other 9 were tested during an intermission.

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TETANY AS A COMPLICATION OF GASTRIC DILATATION,

WITH NOTES OF A CASE.*

By ANDREW TRIMBLE, M.B.,

Belfast.

THERE are few subjects in the whole range of medical literature round which so much controversy has raged as that of tetany. Its first description, in association with gastric dilatation, is attributed by some to Newman¹ in 1861, by others to Steinheim.² Its etiology was the despair of theorists and experimental workers alike, Maestro³ regarding it as an "essential neurosis," Oddo⁴ as "a toxæmia produced in the presence of a hypersecretion of hydrochloric acid and gastric juice," while Berliozheimer⁵ cites a case in which no ptomaines could be found.

Its pathology was for years the will-o'-the-wisp of the men of the microscope and of the morbid specimen, but to their credit be it said they promise fair to be the guides out of the present morass. Its treatment has been the shuttlecock of the battledores of the physician and surgeon, the former of whom have tried everything, from calomel to the latest antiseptic, while the latter have performed operations as varied in intensity as gastric lavage, puncture of the spinal cord, and gastro-enterostomy.

I shall leave, however, the domain of controversy and speculation, and pass on to the clinical notes of a case, and subsequently make some observations on them and on tetany in general.

Personal History.

R. H. S., aged 45, married, by occupation a commercial traveller for a distillery, but, nevertheless, very moderate in his use of alcoholic liquors. Of an active, nervous temperament, slightly built, and rather short of stature. For years he had complained of attacks of indigestion, which were the more remarkable to him because of the enormous quantities of food and fluid he was capable of taking at one meal. Pain ensued during these attacks about an hour after eating, and was only relieved by vomiting, which he sometimes produced artificially, in quantities as large as a chamberful, often wondering "where it had all come from," and being astonished to see in it at times food he must have eaten several days previously—for example, barley groats.

History of the Present Illness.

First Day.—I was called to see the patient first about 8 p.m. on Thursday, September 8th, 1904, and on arrival was informed that he had "had a fit" a short time previously, in which the upper arms were pressed to the sides, the forearms bent at the elbows, the fingers bent at the knuckles, with the thumbs inside the palms. The feet were also contracted, and there were twitchings about the mouth. During the entire attack he was quite conscious—indeed, rather alarmed at the appearance of his hands, exclaiming, quite excitedly, as if watching something entirely outside himself, "Look! Look at my hands!" Bathing in hot water had relieved the contracture and the pain. He was very flushed and sweating when I entered, sitting propped up in bed, carefully holding his hands under the bedclothes lest another

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