have been a previous undetected infection with *Bact. coli* B.G.T. in these cases. On the other hand, an appreciable number of children are admitted with primary gastro-enteritis without any identifiable specific strain of *Bact. coli* in their faeces.

Bacteriology

Routine Examination.—Faeces from cases of gastro-enteritis were examined to exclude the presence of protozoa and cultured on Leifson's desoxycholate citrate medium as well as in selenite enrichment medium to exclude the presence of salmonellae and shigellae. A MacConkey plate was also inoculated and at least four representative colonies were tested by slide agglutination, using Bray and Beavan's (1948) technique, which gave very clear-cut and reliable results. All positive colonies were inoculated into sugars, and a culture was sent to Dr. Joan Taylor, who very kindly confirmed their identity.

Sensitivity of the Organism to Chloramphenicol.—*Bact. coli* B.G.T. was inhibited in concentrations of chloramphenicol varying between 2 to 4 $\mu\text{g./ml.}$ Tests were made using representative colonies from each specimen from which *Bact. coli* B.G.T. was isolated whilst the child was on chloramphenicol treatment or if the organism reappeared after treatment had been stopped. No change in sensitivity of the organism was shown in these experiments. This is in agreement with the work of Smadel *et al.* (1949) and Alexander *et al.* (1949). This result is very different from those obtained with streptomycin, when insensitive variants began to appear in nearly all cases within 24 hours of the initiation of treatment. In most of the antibiotic-treated cases specimens of faeces were obtained once or twice daily during treatment and daily for two weeks after its cessation or until the patient was discharged.

Blood Levels.—Estimations of chloramphenicol blood levels were made; the technical difficulties were not fully overcome, but by the use of a modification of the technique described by Fleming and Smith (1947) levels up to 120 $\mu\text{g./ml.}$ were obtained. The test organism used was a strain of *Bact. coli* B.G.T. from Case 5. There seemed to be definite individual variations in the absorption of the drug by the patient, so a relatively high dose of chloramphenicol was employed.

Summary

In Birmingham, as in many other parts of Great Britain where it is sought, *Bact. coli* B.G.T. has been found to be intimately associated with epidemic infantile gastro-enteritis. The enteritis did not respond to the administration of sulphonamides, penicillin, or streptomycin, but the response, both clinically and bacteriologically, to chloramphenicol has been most encouraging.

We would like to express our thanks to the staff of the Children's Hospital, Birmingham, especially to Professor J. M. Smellie for his encouragement and most valuable criticism, to Sister M. Coker for her co-operation, and to Dr. Joan Taylor for much advice and help; also to Dr. C. Giles, who supplied the first antisera used in this investigation.

REFERENCES

- Alexander, H. E., Leidy, G., and Redman, W. (1949). *J. clin. Invest.*, **28**, 867.
 Bray, J. (1945). *J. Path. Bact.*, **57**, 239.
 — and Beavan, T. E. D. (1948). *Ibid.*, **60**, 395.
 Ehrlich, J., Bartz, Q. R., Smith, R. M., Joslyn, D. A., and Burkholder, P. R. (1947). *Science*, **106**, 417.
 Fleming, A., and Smith, C. (1947). *Lancet*, **1**, 401.
 Giles, C., and Sangster, G. (1948). *J. Hyg., Camb.*, **46**, 1.
 Holzel, A., Martyn, G., and Apter, L. (1949). *British Medical Journal*, **2**, 454.
 James, U., Kramer, I. R. H., and Armitage, P. (1948). *Lancet*, **2**, 555.
 Smadel, J. E., Woodward, T. E., and Bailey, C. A. (1949). *J. Amer. med. Ass.*, **141**, 129.
 Taylor, J., Powell, B. W., and Wright, J. (1949). *British Medical Journal*, **2**, 117.

PARATYPHOID B FEVER TREATED WITH CHLORAMPHENICOL

BY

MICHAEL CURTIN, M.D., M.R.C.P.I., D.P.H.

Registrar, Sheriff Hill Infectious Diseases Hospital, Gateshead-on-Tyne

The clinical course of paratyphoid fever is subject to much variation. Compared with typhoid fever it is in general a milder disease, although attacks of paratyphoid fever occur which both in severity and in duration rank with typhoid even of the most severe form. Owing to this variation in degree of clinical illness it is difficult to assess the response to any particular form of treatment. Reports on paratyphoid B fever treated with chloramphenicol (chloromycetin) have been few. Christie (1949) has reported briefly on seven cases, and Kiledjian (1949) and Montuschi (1949) have each reported on a single case. As much investigation remains to be done before the full effect of the drug on paratyphoid fever can be finally assessed, and as it will be some time before an optimum scheme of dosage can be arrived at, the following report may be of interest.

Between June and October, 1949, six clinical cases of moderately severe paratyphoid B fever were admitted to Sheriff Hill Infectious Diseases Hospital. The diagnosis was confirmed in all cases by culture of *Salmonella paratyphi* B. All patients had been ill for some days before admission; treatment with chloramphenicol was started in three cases at the end of the first week, in two cases during the second week, and in the sixth case early in the third week of disease. The dosage and the period of administration of the drug varied, patients who were admitted later in the series receiving larger doses over a longer period. The frequency of administration remained constant, the drug being given at four-hourly intervals.

In addition to the above cases, three apparently healthy faecal carriers of *Salm. paratyphi* B were admitted and treated with chloramphenicol.

Case 1

A girl aged 11 was admitted on June 21, 1949, complaining of malaise, headache, anorexia, diarrhoea, and abdominal pain of five days' duration.

On examination she was obviously toxic, cerebation was dull, her mouth was dry, and her tongue was heavily furred. The abdomen was tumid and slightly tender. The spleen was just palpable. Her temperature was 99.4° F. (37.4° C.) and pulse 120.

Progress and Treatment.—On admission penicillin (200,000 units four-hourly) and sulphasuxidine (8 g. daily) were given and continued for five days. There was no improvement; diarrhoea, abdominal pain and tenderness, pyrexia, and drowsiness persisted. *Salm. paratyphi* B phage type 3A was cultured from faeces. On the tenth day of disease a course of chloramphenicol was

Chart 1.—Case 1.

started (1 g. in first hour, 0.5 g. four-hourly for 24 hours, and then 0.25 g. four-hourly for six days). On the following day the patient's general condition was improved and she was much brighter mentally. Within 48 hours diarrhoea and abdominal tenderness disappeared. Her temperature was normal in four days (Chart 1). There was no clinical relapse, but *Salm. paratyphi B* was excreted in faeces for six weeks after clinical recovery. *In vitro* tests showed that the infecting organisms were sensitive to chloramphenicol even when the temporary carrier state had developed.

Case 2

A woman aged 29 was admitted on July 1, 1949, with a history of the onset of headache, pyrexia, abdominal discomfort, and constipation 14 days previously. At the time of admission her only complaint was that of general weakness and fatigue. On examination her temperature was 100° F. (37.8° C.) and pulse 102. The tongue was furred, and there was evidence of recent wasting. A mitral systolic murmur was heard, but nothing else abnormal was found.

Progress and Treatment.—After admission the patient progressed favourably for three days. *Salm. paratyphi* phage type 3A was isolated from faeces. On the fourth day there was a sudden rise in temperature accompanied by headache, abdominal discomfort, and mild abdominal tenderness. As it was felt that a relapse had taken place, next day she was given chloramphenicol (1 g. in first hour, 0.5 g. four-hourly for 24 hours, and then 0.25 g. four-hourly for five days). Treatment coincided with general improvement. Her temperature did not show a dramatic response, returning to normal on the fourth day of treatment. A convalescent carrier state developed, and this persisted for eight weeks.

Case 3

A boy aged 5 was admitted on July 6, 1949, with a history of lassitude, anorexia, cough, abdominal pain, and diarrhoea.

On examination he was obviously toxic, drowsy, and irrational, and had bouts of noisy delirium. His tongue was heavily furred. The abdomen was distended, and palpation was resented. The spleen was moderately enlarged. A few rose-coloured spots were present on the abdomen. His temperature was 101.8° F. (38.8° C.) and pulse 98.

Progress and Treatment.—On admission on the seventh day of illness a course of chloramphenicol was started (0.25 g. four-hourly for six days). Next day there was a remarkable change in the patient's condition. He was alert and composed, and the abdomen was less tender. Within 48 hours diarrhoea had ceased. His temperature returned to normal on the third day of treatment (Chart 2), coincident with clinical recovery. *Salm.*

Chart 2.—Case 3.

paratyphi phage type 3A was cultured from specimens of blood and faeces taken on the day of admission. A convalescent faecal carrier state developed and lasted for nine weeks.

Case 4

A boy aged 6 was admitted on July 18, 1949, with a history of headache, vomiting, drowsiness alternating with restlessness, and constipation.

On examination his general condition was fair, his tongue was furred, the abdomen was tumid, and a few rose-coloured spots were seen on his abdomen and legs. His temperature was 100.6° F. (38.1° C.) and pulse 112.

Progress and Treatment.—On admission on the seventh day of illness he was put on a course of chloramphenicol (0.5 g. four-hourly for 24 hours and 0.25 g. four-hourly for four days). On the morning following admission several fresh rose-coloured spots were noted. However, his general condition was much improved. The temperature settled to normal in 24 hours. *Salm. paratyphi* phage type 3A was isolated from specimens of blood and faeces taken on the day of admission. The patient continued to excrete the infecting organisms in faeces for seven weeks after termination of treatment.

Case 5

A woman aged 40 was admitted on August 27, 1949, with a history of headache, generalized abdominal pain, constipation, and malaise for six days.

On examination cerebation was dull and speech was sluggish. There was well-marked abdominal tenderness. The tongue was furred. The temperature was 101° F. (38.3° C.) and pulse 92. There were no other abnormal findings.

Progress and Treatment.—On admission the patient was put on a course of penicillin (200,000 units four-hourly) and sulphadimidine (sulphamezathine) (six tablets in the first hour, three four-hourly for 24 hours, and then two four-hourly to a total of 40 g.). Abdominal pain and tenderness, drowsiness, and pyrexia persisted. Faeces were reported as positive for *Salm. paratyphi B* phage type Taunton. Serum gave a positive agglutination with this organism to a titre of 1 in 250 (H). On the 14th day of disease a course of chloramphenicol was begun (2 g. in first hour, 0.5 g. four-hourly for five days, and then 0.25 g. four-hourly for four days—a total of 23 g. in nine days). Within 24 hours there was a change in the patient's condition. She was brighter and said she felt much better.

Chart 3.—Case 5.

This was in marked contrast to her previous outlook. The abdominal tenderness improved, disappearing after three days. Her temperature was normal on the fourth day (Chart 3). Faeces were still positive for *Salm. paratyphi B* on the fifth day of treatment, but negative thereafter. *In vitro* tests showed that the infecting organism (*Salm. paratyphi B* phage type Taunton) was sensitive to chloramphenicol.

Case 6

A boy aged 12 was admitted on September 19, 1949, with a history of headache, abdominal pain, pyrexia, and constipation for four days.

On examination the abdomen was moderately distended. The tip of the spleen was palpable. Rose-coloured spots were present on the abdomen and legs; there was slight generalized adenitis; and the tongue was furred. His temperature was 99.8° F. (37.1° C.) and pulse 94.

Progress and Treatment.—Cultures of blood and faeces were positive for *Salm. paratyphi* phage type 3A. Serum gave a positive agglutination with this organism to a titre of 1 in 250 (H). Chloramphenicol was begun on the seventh day of illness (2 g. in first hour, 0.5 g. four-hourly for five days, and then 0.25 g. four-hourly for four days—a total of 23 g. in nine days). There was a dramatic change in the patient's condition. Within 24 hours he no longer complained of headache or abdominal pain. His temperature became normal in 18 hours and, apart from a slight rise (99° F., or 37.2° C.) on the fifth day, remained normal. Faeces were negative for *Salm. paratyphi* on the fifth day. Four subsequent specimens were also negative.

Carriers

During July three apparently healthy faecal carriers of *Salm. paratyphi B* phage type 3A were admitted and treated with chloramphenicol. The dosage given was 16.5 g. in seven days (0.5 g. four-hourly for four days, and then 0.25 g. four-hourly for three days). This dosage had no effect on the carrier state.

Comment

The satisfactory evaluation of the effect of chloramphenicol is not possible in so small a series of cases, especially in the absence of a control group. The rapid improvement and clinical recovery which coincided with treatment, however, were most impressive. This was particularly so in those treated early in the disease.

The dosage of chloramphenicol adequate to produce a well-marked clinical response did not in all cases render the faeces free from the infecting organisms. A similar observation was made in the case of typhoid fever by Woodward *et al.* (1949) and by Rumball and Moore (1949).

In vitro tests showed that both *Salm. paratyphi B* phage type 3A, and *Salm. paratyphi B* phage type Taunton were sensitive to chloramphenicol. It was encouraging to note that in a case in which sensitivity tests were carried out after treatment the organism had not become resistant to chloramphenicol.

It remains to be seen whether larger doses over longer periods will prevent the development of the convalescent carrier state, and whether the drug is effective in treatment of the chronic carrier condition.

I am indebted to Dr. J. Grant, medical superintendent, for his invaluable advice in the preparation of this paper, and to Dr. R. Norton, Director of the M.R.C. Public Health Laboratory, Blakelaw, Newcastle-upon-Tyne, who carried out all bacteriological investigations, including the chloramphenicol sensitivity tests.

REFERENCES

- Christie, A. B. (1949). *Post-Grad. med. J.*, **25**, 409.
Kiledjian, Y. (1949). *Lancet*, **2**, 629.
Montuschi, E. (1949). *Ibid.*, **2**, 675.
Rumball, C. A., and Moore, L. G. (1949). *Brit. med. J.*, **1**, 943.
Woodward, T. E., *et al.* (1948). *Ann. intern. Med.*, **29**, 131.

DIFFRACTION METHODS OF MEASURING RED CELL DIAMETERS

THEIR USE IN CLINICAL HAEMATOLOGY*

BY

ALAN J. HIRD, M.B., Ch.B., B.Sc., M.R.C.P.

(From the Gardiner Institute of Medicine, University of Glasgow)

An important feature of many anaemias is the alteration which occurs in the size of the average red cell. This alteration is important in diagnosis and in treatment, since, for example, many anaemias associated with increased red cell size have a common aetiological basis and respond to liver therapy.

The recognition of changes in red cell size can be made in various ways. Probably the oldest is direct inspection of blood films, stained or unstained, under the microscope—a method fraught with difficulty even for those accustomed to making blood examinations and with positive danger for those not so accustomed. Actual measurement of the diameters of a large number of cells (Price-Jones, 1929) converts the method into one of considerable accuracy

but of such tediousness that it is ruled out except for special research. An entirely different method, depending upon measurements of the packed cell volume and of the red cell count, gives a value for the mean corpuscular volume (Wintrobe, 1946). While the packed red cell volume can be determined with an accuracy of $\pm 1\%$, the red cell count (and consequently the mean corpuscular volume) is subject to errors of at least $\pm 5\%$.

An ingenious alternative method for measuring the mean diameter of red cells depends upon the diffraction of light around the edges of the cells in an unstained film in a manner similar to that which occurs in a diffraction grating. By suitable optical methods the diffracted light can be projected upon a ground-glass screen as a series of coloured haloes (when ordinary white light is used). The diameter of the haloes varies inversely with the average diameter of the red cells. This method was originally described by Pijper (1919), and a later article (Pijper, 1947) gives a full historical review, including references to modifications by other workers, together with a detailed account of the technique and its application. It is claimed not only that the mean cell diameter can be determined but also that the degree of anisocytosis can be estimated by the degree of spread between the inner violet and outer red rings. Brief descriptions of the method will also be found in the standard textbooks on haematology (Whitby and Britton, 1939; Todd and Sanford, 1943; Wintrobe, 1946). Some workers have cast doubt upon the accuracy of claims made for diffraction methods (e.g., Fullerton *et al.*, 1933), and the general impression derived from a study of the literature is that the method is a useful confirmatory one but not reliable enough to be of use as a single critical method of diagnosis. A modification of the method, using two lamps (Pryce, 1929), has simplicity to commend it, but has been criticized by Pijper on the ground that it does not measure the diameter of the average cell but that of the smallest cell present in any considerable number.

Objects and Methods of Present Investigation

The object of this investigation was to determine whether the simple diffraction method of measuring the mean red cell diameter was of value in addition to or in place of more elaborate studies. Several uses were considered: (1) as a "screening" device in the diagnosis of untreated cases of anaemia; (2) as a control in the treatment of cases of pernicious anaemia; and (3) in the investigation of certain other conditions, particularly liver disease.

The study covers a period of almost two years, during which time all new cases showing features of haematological interest in one medical unit were personally investigated, and many followed up at intervals. In addition to conditions usually grouped under the heading of primary blood diseases a considerable number of disorders primarily involving other systems were also dealt with. Excluding a large number of investigations in which the diffraction method was used but for various reasons a complete blood examination was not made, there remain 353 blood examinations on 205 individuals in which haemoglobin, red and white cell counts, haematocrit, absolute values, sedimentation rate, and icterus index were determined in addition to the measurement of the mean cell diameter by the diffraction method and examination of stained films. Table I shows the numbers of each type of disease investigated.

Blood in all cases was withdrawn from a vein into a small bottle containing the appropriate amount of oxalate mixture (Wintrobe, 1946). Haemoglobin was determined by the oxyhaemoglobin method (Bell *et al.*, 1945), using a

*The proofs of this article were passed by Dr. Hird shortly before his tragic death on December 11, 1949.