THE TREATMENT OF BACILLARY (FLEXNER) DYSENTERY WITH SULPHANILYLGUANIDINE

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Search for an efficient intestinal antiseptic has in the past yielded poor results. *In vitro* tests of the bactericidal activity of antiseptics are no guide to their usefulness in suppressing pathogenic bacteria in the gut, for the substance must first pass the acid barrier of the stomach, withstand the action of bile and other intestinal juices, and exert a bactericidal action on the intestinal pathogens in a *milieu* of broken-down organic matter and hordes of saprophytic bacteria. Because the natural intestinal infections of man are so difficult to reproduce experimentally, the bactericidal or bacteriostatic activity of an antiseptic substance on the normal intestinal flora, particularly the coliform group, of experimental animals is probably the most useful index for its therapeutic trial in intestinal infections. Graham (1932) tested in this way a large series of substances which were powerfully bactericidal to the colon bacillus *in vitro*, but he failed to find any which produced an appreciable change in the intestinal flora when given by stomach-tube to mice. With the advent of the sulphonamides a fresh attack was made on the problem, and claims have been advanced for the therapeutic activity of different sulphonamide preparations against intestinal infections mainly on the basis of their bactericidal action on the typhoid-dysentery group of organisms *in vitro* or in the peritoneal cavity of injected animals. Since very large doses of typhoid or dysentery bacilli are usually required to kill a mouse infected intraperitoneally, the animal experiments were little better than tests in the tube. On the clinical side, different drugs, particularly those of the phenol series, have from time to time been popular as intestinal antiseptics. Lately, reports of an apparently beneficial result from the use of sulphapyridine or sulphathiazole on single or small groups of cases of typhoid and dysentery have appeared. As examples, Reitler and Marberg (1941) treated 20 cases of acute bacillary dysentery at Haifa with 1 gramme of sulphapyridine three or four times daily for two to four days, and found that most of the symptoms disappeared in two days' time; while Bell (1941) reported good results in the treatment of 14 cases of Flexner and Sonne dysentery with a total average dosage of 5 grammes of sulphapyridine: in neither group was there an adequate control series. The objections to the use of these drugs as intestinal antiseptics are that they are gastric irritants and readily absorbed from the gut, so that if given in full doses they may not only upset an already irritable stomach

but because of the risk of dehydration and oliguria may produce severe toxic effects on the kidneys.

Properties of Sulphanilylguanidine

Marshall and his colleagues at Baltimore conceived the idea of using as intestinal antiseptic a sulphonamide compound which, although soluble in water and efficient chemotherapeutically, was poorly absorbed from the intestine. Of a number of such compounds which they had prepared, sulphanilylguanidine was chosen as being the most active against various bacteria. In 1940 Marshall *et al.* described the preparation of this drug, its pharmacology and toxicity, and its bactericidal or bacteriostatic activity on the intestinal flora of mice; and in a second paper (1941) its therapeutic value was demonstrated in a small series of dysentery infections. In their experience the toxicity of the drug for animals was of the same order as that of sulphapyridine and sulphathiazole, but as only a fraction of sulphanilylguanidine given by mouth was absorbed the risk of toxic effects with it seemed small. What absorption occurred was mainly from the small intestine, so that for a maximum therapeutic effect on the lower bowel infrequent large doses seemed likely to serve best. The absorbed fraction, part of it acetylated, was quickly excreted by the kidneys. Its most remarkable property was its power, when fed to mice, to reduce to scanty numbers the thousands or millions of coliform bacilli in the faeces. For clinical use these authors recommended an initial dose of 0.1 gramme per kg. of body weight followed by a maintenance dose of 0.05 gramme per kg. every four hours until the number of stools daily was four or less; then 0.1 gramme per kg. (0.05 gramme per kg. for adults) every eight hours for another three days. With such dosage they reported good results in 7 cases of acute bacillary dysentery treated within three days of onset; the therapeutic effect was variable in 10 cases treated later in the course of the disease. They quoted Lyon as having obtained excellent results in 15 out of 20 treated cases of severe dysentery, compared with 20 untreated control cases.

The value of a drug which, particularly in wartime, could be used for the control of dysentery was at once apparent to one of us (R. C.), and through the kindness of Sir Henry Dale a supply of sulphanilylguanidine was prepared by Mr. J. Walker, who appends a note on its preparation. Later we were fortunate in obtaining further supplies from

the American Red Cross Harvard Field Hospital through the kindness of Prof. J. E. Gordon and Dr. P. B. Benson, while a generous sample was also given us by Dr. Prescott of the Wellcome Foundation, Ltd.

Material and Methods

Although dysentery was the obvious choice of infection on which to test this new sulphonamide, the prevalent form of dysentery in the community was the usually mild Sonne infection, which, being self-limited and uncomplicated like bacterial food poisoning, did not lend itself to the therapeutic assessment of a new drug. However, with the good will of Dr. W. Allen Daley, Medical Officer of Health to the London County Council, and the sympathetic help of Prof. S. Nevin and Dr. S. A. Mann of the L.C.C. Mental Hospitals Pathological Service, arrangements were made to give the drug a therapeutic trial at Cane Hill Mental Hospital, where, opportunely, an epidemic of Flexner dysentery began in December, 1940, and continued with intermissions until June, 1941. Dysentery in mental hospitals is usually an endemic infection, often flaring up in the winter months and varying in severity from a mild diarrhoea to the most violent dysentery with frequent bloody stools and marked prostration. In this particular outbreak the infection was more severe and protracted than usual. Attempts to control its spread in the early stages by the use of a stock polyvalent Flexner vaccine were unavailing, and the prevalence and clinical severity of the outbreak may have been due to the introduction of a new type of Flexner bacillus along with a large influx of fresh patients. Once introduced into a ward the infection spread quickly to a large proportion of the inmates, who could not easily be taught the principles of personal hygiene. The prevalent type of organism was at first *B. dysenteriae* (Flexner Z) and later *B. dysenteriae* (Flexner X)*. The period of our investigation was from January to June, 1941.

All the patients were adults, of whom 130 were under observation. In this report, however, only patients who had both blood and mucus in their stools or who were bacteriologically confirmed to be suffering from Flexner dysentery were accepted for analysis. This reduced the number to 96, of whom 41 were treated with the drug and 55 were untreated controls. Their ages varied from 25 to 76 years; 55 were over 50 and many of them were in poor physical condition; 40 were males and 56 females.

Dosage of Drug.—The drug was given in powder form. It can be introduced as a suspension in water or milk; it has very little taste, and does not produce any nausea or vomiting. At first the prescribed dosage was 6 grammes a day in three doses of 2 grammes for four days, but after a preliminary trial this dosage was changed to 9 grammes a day in three doses of 3 grammes for two days, followed by 4 grammes a day in two doses of 2 grammes. If the diarrhoea was not controlled in four days the maintenance dose of 4 grammes a day was continued for one week. These are smaller doses than those recommended by Marshall *et al.*, but we were anxious to avoid the risk of any toxic effects from a new and clinically untried drug, and this dosage seemed to be therapeutically effective in most cases. However, later experience has convinced us that larger doses can safely be used, and may be advisable in the more severe cases of dysentery. A sample of blood was taken into oxalate twenty-four hours and again four days after starting treatment, in order to estimate the blood level of the drug. A morning specimen of urine was collected on the fourth day of treatment and examined for drug content, protein, crystals, casts, and cellular content. White cell counts were done on a number of treated and untreated cases. Bacteriological methods are discussed later. In all treated cases a liberal supply of fluids was insisted upon during the period of drug therapy, since sulphanilylguanidine, and particularly its acetylated derivative, might have irritative and toxic effects on the kidneys, similar to those produced by sulphapyridine and sulphathiazole, if adequate excretion was not maintained.

Clinical Results

While the principle of the alternate case was adopted in selecting cases for treatment, a fair number of patients did not develop the typical features of dysentery in the first day or two

of illness, but were later diagnosed on either clinical or bacteriological findings as Flexner infections. As a result treatment with the drug was reserved for an unduly large proportion of the more severe and classical infections. For purposes of analysis, therefore, the cases, both treated and untreated, were divided into two groups, labelled "severe" and "mild." The criteria of a severe case were a high initial temperature of 101° to 105° F., frequent stools (four to sixteen a day) containing both blood and mucus, and a prostrated toxic patient. The typical course of such a severe untreated case was an abrupt onset with high fever, headache, general malaise, severe abdominal pain, and in a few cases vomiting. Very soon there were frequent stools containing blood and mucus and accompanied by much tenesmus; the patient became quickly dehydrated and was very weak and toxic. Many of these cases had a typical, almost diagnostic, odour. The diarrhoea, usually with blood and mucus, continued for a week or ten days, and sometimes longer, and when the blood and mucus disappeared the motions often remained relaxed for a further few days. The temperature fell by lysis to normal in four to seven days; appetite improved after a week but convalescence was slow and protracted. In the milder case there was an initial rise of temperature to 99° to 102° F., abdominal pain and malaise, and relaxed stools, usually with blood and mucus, for two to five days. The temperature settled fairly quickly; abdominal pain disappeared, but malaise and anorexia persisted for a week or so, and the patient made a slow recovery.

Of 41 treated cases, 28 were classified as severe and 13 as mild; for the 55 untreated cases the distribution was 16 severe and 39 mild. Thus there was a much higher proportion of severe cases in the treated than in the untreated group. Two patients died among the 28 severe treated cases: one, aged 42, on the eighth day of infection despite a dramatic response to drug therapy; the other, aged 62, on the twenty-second day after making a good recovery from his dysentery. Three patients among the 16 severe untreated cases died: one, aged 29, on the third day of infection; one, aged 47, on the seventh day; and one, aged 60, also on the seventh day.

The criteria of clinical response to sulphanilylguanidine were: (1) a change in the character and frequency of the stools; (2) a rapid fall in temperature; and (3) the disappearance of toxic symptoms. A careful daily record was kept of the number and naked-eye appearance of the stools of every patient under observation, particular attention being given to the presence of blood and/or mucus. As the investigation proceeded it was felt that the number of motions a day was a less reliable guide to the response to treatment than the time in days of the disappearance of blood and mucus from the stools. In this connexion the day of disease on which chemotherapy was begun was an important factor in the reckoning. Among the severe cases the average time was 3.0 days from onset, ranging from two to five days, and in this group blood and mucus were present for an average period of 4.8 days, or 1.8 days after treatment was begun. In two patients blood and mucus persisted for more than a week after treatment was started. Among the severe untreated cases the average duration of blood and mucus in the stools was 6.8 days, or exactly two days longer than in the treated series. In the group of mild treated cases chemotherapy was begun at an average of 3.2 days from onset, and blood and mucus were present for 2.8 days from onset, so that in many cases blood and mucus had disappeared before specific treatment was given. The average duration for bloody mucoid stools in the untreated mild group was 2.5 days. This criterion of clinical response was therefore valueless in the mild type of dysentery.

Temperature charts of all cases were kept for analysis, but a cursory examination of these showed that the difference in duration of pyrexia between treated and untreated cases was not likely to be statistically significant. The fact that in many cases treatment with the drug was not begun until the third day of illness precluded any striking effect on pyrexia, which in untreated patients was beginning to settle by the third or fourth day of disease. There were, however, outstanding cases with a critical fall of temperature apparently in response to drug therapy, just as there were untreated cases with pyrexia continuing at a fairly high level for five to seven days. It is impossible to say, with the limited numbers under review, whether these were chance occurrences or not.

* We are indebted to the late Dr. W. M. Scott and to Dr. Marion Watson for the final identification of these strains.

The general response and behaviour of an ill patient under treatment with a new remedy is not readily susceptible to analysis, and the evidence must be assessed as objectively as possible by a physician with experience of the disease and with an adequate control series. At first, when the infection was rather mild and the initial dosage small, it was difficult to be positive about the beneficial effect of sulphanilylguanidine. Later, particularly with early treatment of the more severe cases, the clinical response to the drug was very definite and sometimes dramatic. Nursing staff became enthusiastic about its curative value. In a typical severe case improvement of the patient's condition was noted in twenty-four hours after giving the drug, and by the third day the patient was feeling much better with disappearance of toxic symptoms and return of appetite. Bowel movements became much less frequent within two days, though blood and mucus often persisted for three to four days. Most striking of all, convalescence was much shorter than in the untreated cases. In the milder infections, too, the treated patient was quickly sitting up, with good appetite, while the relaxed stools were followed by a period of constipation.

Relapses.—In the untreated series it was not uncommon for relaxed mucoid stools to continue for some time after the patient was clinically well, whereas the treated patient usually became constipated. But in the treated group four of the severe cases and one mild case had slight relapses, with blood and mucus in the stools for one or two days. These fresh attacks all occurred within a week after cessation of drug therapy, and were probably true relapses rather than reinfections. In only one of them was the Flexner bacillus isolated during the relapse.

Bacteriology

In the early stage of this investigation material for bacteriological examination was taken every day of the acute illness direct from the rectum by means of a rectal speculum and stout loop. The material obtained—usually mucus mixed with some faecal matter—was either planted immediately on to plates of MacConkey's medium or emulsified in warm saline and inoculated in the laboratory within one to two hours of taking the specimen. Suspicious colonies were picked next day, inoculated into the appropriate sugars for fermentation reactions, and tested by the slide-agglutination technique with the Flexner I and II polyvalent antisera of the Oxford Standards Laboratory. The results obtained by these methods were not very satisfactory, and towards the end of April a new medium became available which gave a much higher yield of positive results. This medium is a modification of the Difco S.S. formula, which is based on Leifson's (1935) work on the selective inhibitory action of sodium desoxycholate, citrate, and other chemicals on the normal intestinal bacteria. Full details of this desoxycholate-citrate agar and its usefulness in the isolation of the various intestinal pathogens will be published separately by Dr. Martin Hynes. Table 1, prepared by Dr. Hynes, shows the results obtained with material from patients with Flexner infections, and demonstrates in particular the value of the new medium in isolating the pathogen during convalescence.

TABLE 1.—*A Comparison of the Isolation of B. dysenteriae (Flexner)*

Day of Disease	No. of Specimens			% Positive	
	* McC. + † D.C. +	McC. - D.C. +	McC. - D.C. -	McC.	D.C.
1	16	4	0	80	100
2-4	19	15	1	54	97
5-7	5	10	3	28	83
>7	6	27	82	5	29

* McC. = On MacConkey's medium.
† D.C. = On desoxycholate-citrate agar.

These figures refer to the total number of specimens examined on desoxycholate-citrate agar. So far as the results from individual patients are concerned the Flexner organism was isolated from 59 (61.4%) of the 96 patients in this series. During the period when Hynes's modification of the S.S. medium, or in a small number of cases the S.S. medium itself, was being used, 37 (92.5%) out of 40 patients yielded the Flexner organism; in 6 of these cases neither blood nor mucus was noted in the stools at any time, and others were suffering from

mild infections. It is only fair to state that during this period, when additional technical assistance was available in the hospital laboratory, the Flexner bacillus was isolated on MacConkey's medium in specimens from 27 (67.5%) out of 40 patients, which was a considerable improvement on the early results. Latterly specimens of faeces were being used as often as material obtained by the rectal loop and speculum.

Convalescent Carriers

With the new desoxycholate-citrate medium, which detects relatively small numbers of pathogens in the faeces, it became possible to assess the value of sulphanilylguanidine in eliminating Flexner bacilli from the bowel. While it is generally believed that the Flexner organism quickly disappears from the gut after an acute attack, there is little doubt, from epidemiological evidence, that convalescent carriers must exist, and these cases, together with cases of chronic dysentery, are mainly responsible for spreading the disease and for maintaining it as an endemic infection in a community.

If an intestinal antiseptic should prove to be not only beneficial against the acute infection but also able to prevent the convalescent carrier state or the development of chronic dysentery, it would be a great boon to the public health in general and to asylum communities in particular. With the help of the new medium an examination was therefore made for convalescent carriers of Flexner's bacillus in a series of treated and untreated patients, all of whom gave positive cultures in the first week of infection. Among 16 treated cases 3 gave positive cultures after the end of the first week; 2 of these were positive on the eighth or ninth day respectively, and neither of them gave positive specimens after a week's treatment with the drug. The third case had a mild relapse on the tenth day of disease, and gave two positive cultures following this fresh attack, which may have been a true reinfection. In contrast to these findings, among 17 untreated cases 8 were positive after the first week of infection, and 5 of them were still positive in the third week, 2 in the fourth week, and 1 in the sixth week. The difference between the two groups may be expressed in another way. Of 43 specimens of faeces examined from the 16 treated patients after the seventh day of disease, only 4 were positive, whereas 14 out of 54 similar specimens examined from the 17 untreated patients were positive. Thus sulphanilylguanidine given in the acute stage of a Flexner dysentery infection seemed to be effective in preventing the convalescent carrier state. It should be noted, however, that of the eight convalescent carriers in the untreated group three were initially mild infections, and it may be that such cases are more likely to become carriers than those with more acute infections. Our experience with sulphanilylguanidine in the treatment of Sonne dysentery, in which convalescent carriers are common (Cruickshank and Swyer, 1940), suggests that if the drug is given after the diarrhoea has ceased it is less likely to influence the carrier state. As the drug will now encounter much more organic matter and many more saprophytic bacteria in the bowel, larger doses than seem to be effective in the acute diarrhoeal phase may be required to eliminate the pathogens from the intestine of carriers. For the same reason, sulphanilylguanidine may not be an effective prophylactic in a ward outbreak of dysentery.

Absorption and Excretion

It was desirable to find out what proportion of sulphanilylguanidine was being absorbed from the intestine of patients with acute bacillary dysentery and whether it was readily excreted by the kidneys. The concentration of the drug in the blood twenty-four hours and four days after starting treatment and the urinary concentration on the fourth day were therefore estimated on 28 patients by the method of Hynes (1940). In addition an estimate of the total amount of drug excreted in the urine was made in three patients in order to determine more exactly what proportion of the total dosage given by mouth was absorbed and excreted. The average concentration of the drug in the blood, examined about two hours after the last dose of 2 or, more often, 3 grammes and a total amount of 6 to 12 grammes in the previous twenty-four hours, was 1.55 mg. of free sulphanilylguanidine and 2.57 mg. of total drug per 100 c.cm.; on the fourth day, after a dosage of 4 grammes in the previous twenty-four hours, the corresponding blood levels were 0.74 and 1.28 mg. per 100 c.cm. The drug concentra-

tion at twenty-four hours varied for free sulphanilylguanidine from 0.5 to 3 mg. and for total drug from 0.75 to 6 mg. per 100 c.cm.; at four days the corresponding extremes were 0.25 to 1.75 mg. free drug and 0.5 to 2.5 mg. total drug per 100 c.cm. These figures, when compared with the blood levels after similar doses of the other sulphonamides, show that sulphanilylguanidine is poorly absorbed from the bowel and that the amount absorbed bears some relation to the dosage given. The urinary concentration of sulphanilylguanidine on the fourth day after treatment averaged 94.3 mg. per 100 c.cm. for free drug and 150.2 mg. per 100 c.cm. for total drug. The highest concentration, 240 mg. of free drug and 300 mg. of total drug, occurred in the patient who gave the highest blood concentration—namely, 3 and 6 mg. per 100 c.cm. Thus the urinary findings indicated that the absorbed moiety was being readily excreted. The rate of excretion could best be appreciated from the drug concentrations in the measured twelve-hourly collections made on three patients during and immediately after the period of drug therapy. The findings in one of them are given in Table II as an example which was closely paralleled by the other two.

TABLE II.—*The Urinary Excretion of Sulphanilylguanidine after Doses of 9, 9, 4, and 4 Grammes daily from April 12, 1941*

Date	Time	Volume	Sulphanilylguanidine per 100 c.cm.	
			Free	Total
April 12-13 ..	8 a.m.—8 p.m.	32	30	35
	8 p.m.—8 a.m.	12	170	260
,, 13-14 ..	8 a.m.—8 p.m.	16	120	220
	8 p.m.—8 a.m.	14	150	250
,, 14-15 ..	8 a.m.—8 p.m.	24	60	150
	8 p.m.—8 a.m.	14	110	190
,, 15-16 ..	8 a.m.—8 p.m.	32	70	105
	8 p.m.—8 a.m.	22	110	180
,, 16-17 ..	8 a.m.—8 p.m.	16	60	170
	8 p.m.—8 a.m.	10	100	110
,, 19	8 a.m.—8 p.m.	10	10	15
	8 p.m.—8 a.m.			

It will be noted that the concentration rose to a maximum between twelve and thirty-six hours after beginning treatment, fell to a lower level on the third and fourth days when the dosage was smaller, and quickly dropped to a very low concentration after treatment was stopped. There is therefore no evidence that the drug is retained in the tissues for any time after absorption. The proportion of acetylated drug varied both in the blood and in the urine between an eighth and a half of the total, and, as with other sulphonamides, there was no clue as to why certain patients conjugate a large proportion and others very little.

In three cases a rough estimate was made of the total amount of drug excreted in the urine during and immediately after the period of treatment. The calculation, based on the total volume of urine and on concentrations of the drug in twelve-hour collections, gave amounts of 7.5, 8.5, and 13 grammes respectively out of a total of 26 grammes ingested by each patient. If these figures represent the actual amount of drug absorbed, then about one-half to two-thirds must remain in the intestine, to be excreted in the faeces. Estimations were made of the drug concentration in twelve specimens, from three different patients, of fluid or semi-solid stools passed during the period of treatment; the concentration varied from 10 mg. (in a very watery specimen) to 700 mg. per 100 c.cm. Actually only half of the specimens gave concentrations above 200 mg. per 100 c.cm., which is the saturation level of the drug. These figures are lower than those quoted by Marshall and his colleagues (1941), and might be regarded as an indication for bigger doses than were used in this investigation.

Toxicity

With the doses used and the small amount of drug absorbed systemic toxic effects such as occur with the other sulphonamides were not anticipated with sulphanilylguanidine. In fact with one exception no clinical evidence of toxicity was noted: a rash of morbilliform type occurred on the twelfth day of treatment in a patient who, because of persistent diarrhoea, had been given a total dosage of 58 grammes of the drug (9 grammes daily for two days and 4 grammes daily for ten days). Marshall

et al. (1941) commented on the absence of toxic effects in their series of 25 treated children: in a group of 25 adults treated with the drug there were two cases of drug fever and one of mild haemolytic anaemia. The complication that seemed most likely to occur, as a result of the dehydration and diminished urinary excretion, was deposition of the drug in the urinary tract, with consequent irritative or obstructive effects. In no patient in our series, however, was there oliguria or urinary suppression, and no one complained of lumbar pain. Laboratory examination of the fourth-day specimen of urine of 28 patients showed a trace or slight trace of protein in 5: it seemed likely that this finding was related to the infection rather than to the drug. Epithelial cells and leucocytes were seen in about half the specimens, which were all non-catheter; neither red blood cells nor casts (apart from occasional hyaline casts) were noted in any specimen of urine. Crystals, usually in the shape of narrow rectangles, were present in the urinary deposits of 21 out of 28 patients: sometimes scanty, sometimes abundant, the presence and the amount of crystalline deposits bore a close relation to the urinary concentration of the drug. In this series of specimens crystals were usually present when the urinary concentration of free drug was over 100 mg. per 100 c.cm., which seemed odd in view of Marshall's statement that the drug is soluble to 200 mg. per 100 c.cm. Test-tube experiments were therefore made in order to find out the saturation level of the drug in urine of pH 5.6. At body heat (37° C.) the solubility was 175 mg. per 100 c.cm., at room temperature (18° C.) 150 mg., and at 4° C. 125 mg. Thus our finding of crystalline deposits with low urinary concentrations of the drug was probably related to the fact that many specimens of urine in this investigation were sent by post to the group laboratory during the winter months; solubility may also be affected by a high level of other inorganic constituents such as would be present in concentrated urine from febrile patients. Where the urinary concentration of the drug was high—as, for example, in the second-day specimens of the patients whose urine was examined daily—sheaves of narrow pointed crystals rather like those of tyrosine were seen in the urinary deposit and were regarded as the crystals of the acetylated drug. Similar crystals were observed in the urine of treated patients not in this series. One of these was a woman aged 86 with a severe *Salmonella reading* infection who was not benefited by large doses of sulphanilylguanidine and who died on the eleventh day of her infection. Histological examination of the kidneys showed marked degeneration of the tubular epithelium but no evidence of any irritative reaction. Nevertheless where large doses of the drug are being given it seems a wise precaution to insist on a liberal fluid intake.

White cell counts were done on 12 treated cases at the end of the period of drug therapy and in 9 untreated control cases. In the treated group the counts varied from 7,900 to 13,000 cells per c.mm., with an average count of 9,666; in the control group the extremes were 7,300 and 14,800 cells per c.mm., with an average of 9,733. Two treated patients with counts of 12,000 at the onset of infection were retested two weeks later and had counts of 9,800 and 10,200 cells per c.mm. respectively, which merely suggests that there is a slight leucocytosis at the onset of an acute dysentery. When it is remembered that more than half of the patients in the series were over 50 years of age, with a natural tendency to impairment of function, it is obvious that sulphanilylguanidine in the doses given has practically no toxicity for the tissues.

Conclusions

Sulphanilylguanidine, a new intestinal antiseptic, has been used in the treatment of 41 adult cases of acute bacillary (Flexner) dysentery with beneficial results, particularly in the more severe infections. A series of 55 cases in the same epidemic served as controls. While a dosage of 3 grammes three times a day for two days followed by 2 grammes twice a day for two to five days was therapeutically effective, larger doses are safely tolerated and may be advisable in severe or refractory infections.

Treatment with the drug in the acute stage of the infection apparently helped to prevent a convalescent carrier state, which was present in about half of a bacteriologically controlled untreated group.

Sulphanilylguanidine probably owes much of its effectiveness as an intestinal antiseptic to its poor absorption from the intestine, since one-half to two-thirds of the drug remains unabsorbed.

Blood and urinary concentrations of the drug were low relative to the dosage given, and toxic effects were conspicuous by their absence. Since crystalline deposits occur in the urine with concentrations over 175 mg. per 100 c.m., an adequate fluid intake and output should be maintained to avoid local toxicity or irritation in the urinary tract, although there was no evidence of any such effect in this series of treated cases.

The use of a new culture medium, desoxycholate-citrate agar, resulted in the isolation of *B. dysenteriae* (Flexner) from a high proportion of both typical and atypical infections. Its superiority over MacConkey's medium was most noticeable in the isolation of the pathogen during convalescence.

The combination of an efficient intestinal antiseptic and a culture medium which will readily detect mild infections and convalescent carriers may be instrumental in eliminating dysentery from mental hospitals.

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THE PREPARATION OF SULPHANILYLGUANIDINE

It appears that sulphanilylguanidine (I) was first obtained by Buttle *et al.* (1938), who, from its mode of formation, unwittingly ascribed an erroneous constitution to their product (Dewing and Smith, 1941). The compound was first described as such by Roblin and his co-workers (1940), who encountered it accidentally in the course of their study of the synthesis of heterocyclic derivatives of sulphanilamide. These authors, for identification purposes, then obtained the substance by reduction of the nitro group in *p*-nitrobenzenesulphonylguanidine (II), which they synthesized (cf. Winnek, 1940, 1941). A few weeks later Marshall and his colleagues (1940) published a description of a large-scale preparation of sulphanilylguanidine, and their method was followed in the present work. Condensation of acetyl-sulphanilyl chloride with guanidine nitrate in the presence of a large excess of sodium hydroxide yields acetyl-sulphanilylguanidine (III), which is subsequently hydrolysed with hydrochloric acid to free sulphanilylguanidine:

Two points of interest emerge and deserve comment. As obtained initially, the acetylsulphanilylguanidine (III) forms a somewhat pasty white solid which tenaciously retains water. Drying is a very slow process under the ordinary conditions (hot oven at 90° C., or vacuum desiccator, etc.), and furnishes apparent yields of the same order as those quoted by Marshall *et al.*—namely, 65 to 70% of theory. Hydrolysis of this product yielded free sulphanilylguanidine also in the amount reported by Marshall *et al.*—namely, 45 to 60% of theory. The process of drying can, however, be greatly accelerated by washing the cake of crude acetyl derivative on the filter with small volumes of spirit and then ether, in which solvents the

compound is insoluble. Drying in the hot oven at 90° C. then gives an absolutely dry product in an hour or two, and, on account of the more efficient drying, the yields obtained at this stage appear to be somewhat smaller—namely, 59 to 62% of theory. On the other hand, hydrolysis of the acetyl derivative dried in this way gave free sulphanilylguanidine in the improved amounts of 61 to 72% of theory.

Tests for acute and chronic toxicity of this preparation on mice gave results very similar to those reported by Marshall *et al.* (1940).

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A TELLURITE-IRON-ROSOLIC ACID MEDIUM SELECTIVE FOR *B. DYSENTERIAE* (FLEXNER)

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Dysentery bacilli, unlike those of the typhoid-paratyphoid groups, have not shown any peculiar characters enabling the investigator to devise a medium which, while inhibiting the growth of other organisms, allows them to develop and be readily identified.

On litmus lactose agar and on the media of MacConkey, Conradi-Drigalski, and Endo the unrestrained growth of *B. coli* leaves little place on the plates for the development of dysenteric bacilli, so that their successful isolation is usually possible only in the early acute stages of the disease and when the stools are planted out within a few hours of their collection. To the antiseptic action of such dyes as malachite green, methyl green, victoria green, brilliant green, fuchsin, crystal violet, and gentian violet dysenteric bacilli are more sensitive than most strains of *B. coli* (Churchman, 1912, 1913; Krumwiede and Pratt, 1914; Kligler, 1918). The new Difco Bacto S S agar specially devised for the isolation of *Salmonella* and *Shigella* strains is designed to meet a long-felt want, a want which may also be satisfied by the medium described in this paper.

The Action of Sodium Tellurite

In the course of an investigation of the suitability of a modified bismuth sulphite medium for the selective growth of Flexner's bacillus, addition of sodium tellurite was found to reduce the growth-inhibitory action both of the bismuth and of the tellurite.

We have shown in the development of our bismuth sulphite medium that the addition of iron salts enabled the *B. typhosus* to develop while the growth of many other organisms continued to be suppressed (Wilson and Blair, 1927). It was this experience which led us to try a combination of tellurite and a ferric iron salt. Having found the bismuth sulphite media unsuitable for use with tellurite, we tried the addition of tellurite to MacConkey's medium, and discovered that the addition of 0.5 c.cm. of a 1% watery solution of sodium tellurite to 100 c.cm. of the medium completely inhibited the growth of nine strains