

THE TREATMENT OF CHOLERA WITH BACTERIOPHAGE.*

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ASIATIC cholera is an acute disease running its course with extraordinary rapidity. It is quite common in cholera to see many a healthy man doing his normal work in the evening who passes the first stool in the morning and is dead within the next 24 hours. This rapid course of the disease is the most important factor to be reckoned with in the treatment of Asiatic cholera. We are of the opinion that the so-called early or the mild cases of cholera are entirely unsuitable for determining the efficacy of any treatment. Statistics based on the "cures" of such cases are worthless. So we have excluded such cases from our consideration. We shall deal with the treatment of cases of fully developed true Asiatic cholera. These are the cases who are admitted already in a collapsed condition or who fall into collapse within 6 hours of admission.

Again, it must be emphasised that we do not contribute to the view ordinarily held that the symptoms are largely or entirely due to the loss of fluid from the body. We do not propose to discuss experimental laboratory evidence in favour favour of our view because that would carry us out of the scope of this paper. We are, however, convinced by our clinical experience alone, that in cholera we are dealing with cases of a disease the victims of which suffer from obvious symptoms of severe intoxication. It is also beyond the scope of this paper to enter into the controversy regarding the nature of the toric the toxins responsible for the syndrome of cholera. But there is no doubt that the patient suffering from cholera is evidently suffering from the effects of a very potent poison.

We also believe that Asiatic cholera is caused by an infection with the Vibrio choleræ, the classical vibrio of Koch. Whether the toxins which cause the syndrome of cholera are derived from the bodies of the vibrios or from the tissues of the host we do not propose to discuss, but their action coincides with the infection with the V. choleræ and their active multiplication in the intestine of the patient. We are, therefore, confronted with two important problems in the treatment of cholera. The first is to destroy the vibrios, to stop or inhibit their multiplication in order to prevent further damage. The second is to undo the mischief that has already been done, to eliminate the toxins already present, to enable the system to pass over the collapsed stage in order to recover from the initial shock it has received from the poison already absorbed. Bearing in mind the rapid course of this disease our remedy must be such as to bring about these results in the shortest time possible.

With regard to bacteriophage we know that a small quantity of it destroys a very large number of cholera vibrios *in vitro* in less than 2 hours and, as it destroys them, it regenerates itself and grows at their expense. This quality of the bacteriophage makes one expect it to be the ideal remedy for the treatment of cholera inasmuch as it is entirely harmless to man and can be taken in incredibly 'large doses.

Our preliminary work on the study of bacteriophage enabled us to prepare a sufficiently potent cholera bacteriophage, which we used in the treatment of patients admitted into the cholera ward of the Patna Medical College Hospital during the cholera season of 1929.

The results showed that in bacteriophage we have a promise of a very potent remedy for the treatment of cholera. The results should be as good or even better in the hands of everybody as a bacteriophage can now be obtained which is even better than that we used in 1929.

We ourselves could hardly believe our own results. It certainly is a remarkable statement to say that the mortality from cholera by our method of treatment was reduced almost to nil. But this is what our results showed. We have never come across such striking results in the treatment of cholera obtained by any other method previously. And when we say cholera, we mean by it a severe attack of virulent true epidemic Asiatic cholera; the patient being pulseless and severely collapsed, passing in his stools the true cholera vibrio in almost pure culture. To reduce the mortality in such cases to about nil is certainly remarkable.

Our results can be summarised as follows:— For comparison we give also the results obtained in the same hospital during the same epidemic but without our methods being applied. At that time we were only observing the cases without treating them.

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		cent.
The total number of cases treated in the cholera ward before we took it over	24	
The total number of deaths amongst them	5	21
The number of bacteriologically proved cases of cholera amongst them	16	
The number of deaths amongst those 16 cases	4	25
The total number of cases treated in the cholera ward <i>after</i> we took it over	266	
The total number of deaths, disregarding the actual cause, amongst them pneu-		
monia, toxæmia, post-partum, typhoid	18	7

fever, leprosy, etc. . 18 The number of cholera cases proved bac-140 teriologically

The number of deaths amongst those 140 cases (excluding those who died within 2 hours of admission, i.e., where no therapeutic intervention could have any effect) . . . : . .

The principles which we applied in the treatment of cholera were very few, but were strictly observed. They were as follows:-

1. The treatment of the patient must begin as soon as possible, without any delay.

2. To get the best results we cannot rely on bacteriophage alone. We mentioned above that our bacteriophage destroyed vibrios in less than 2 hours, in vitro, where from the very beginning bacteriophage is brought in close contact with the vibrio. It is different in the body. There we can introduce bacteriophage in two ways:-

(a) By the mouth.-Although in the case of cholera the peristalsis is usually very energetic yet it takes a considerable time for bacteriophage to reach every vibrio in the intestines and to destroy them.

(b) By the intravenous route.—It has been proved by animal experiments that bacteriophage has a definite predilection for the intestinal tract, it was also proved that bacteriophage possesses a definite bacterio-taxis, that is to say, it moves quickly in the direction of bacteria sensitive to its action. Therefore, in case of intravenous injection bacteriophage quickly directs itself towards the intestines and comes in contact with the vibrio quite soon. But we must not forget that bacteriophage does only one thing: it destroys living cholera vibrios, but neither neutralises the toxin already present in the body, nor cures the lesions produced by vibrios and their toxin.

Therefore, we must definitely understand the rôle of bacteriophage: sooner or later it stops the further activity of vibrios in the body. To neutralise the damage already done we must use other methods which will help to eliminate the toxin from the body, will support the heart action and will provide the necessary amount of liquid to the dehydrated tissues of the body. Such a method par excellence is the saline alkaline intravenous injection which, therefore, must be used along with adminis-tration of bacteriophage.

3. No antiseptics like calomel, potassium permanganate, essential oils, etc., or absorbents like kaolin must be used. Bacteriophage is a living being and cannot act properly in the presence of antiseptics. Kaolin, etc., absorbs bacteriophage also and prevents its reaching the vibrios.

We shall now describe the details of our method of treatment under two headings, viz., (1) the use of intravenous saline, and (2) the administration of bacteriophage.

(1) The use of intravenous saline.

At the outset it must be mentioned that our method of intravenous saline is a modification of that originated by Sir Leonard Rogers.

We consider it very important that the saline to be used intravenously shall be properly prepared and sterilized. We have made it a rule to prepare it under our own supervision. Its preparation should not be entrusted to nurses or the compounders. We prepare the saline in two parts, one is hypertonic and the other isotonic, the latter is used for the preparation of the alkaline saline. For the hypertonic saline Erlenmeyer's flasks of 2 litre capacity are used. Owing not only to the varying degree of hardness in the water but also to other reasons, we consider it important to use distilled water always. For the preparation of the saline we use only sodium chloride and no other ingredient, the strength of the hypertonic saline being 120 grains of sodium chloride in a pint of distilled water, and of the isotonic saline 60 grains of the salt in a pint of distilled water. We have found that the addition of calcium and potassium chlorides, originally recommended by Sir Leonard Rogers, has no advantage. Tablets of sodium chloride containing calcium are on the market, but the saline made with them becomes turbid when alkali is added to it. We have, therefore, abandoned the use of these tablets in favour of the sodium chloride without any other ingredient. After the salt is dissolved in the distilled water the saline is filtered through a filter paper: three pints of the hypertonic and one pint of the isotonic saline are then placed in a two-litre and one, litre flasks respectively. The flasks are plugged with plugs of gauze instead of cotton-wool to prevent fibres of cotton-wool dropping inside them. They are then sterilized in the autoclave for 30 minutes at 120°C.

Saline in this way can be prepared beforehand and kept ready for the expected number of cases. Sterilization at this temperature prevents the growth of moulds and the saline, if kept protected from dust, may be used after several weeks of storage.

The saline must be sterilized in the autoclave. We have found that sterilization under pressure is essential. It is not desirable that the saline for intravenous use be sterilized by

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boiling. The dissolved air driven off by boiling renders the saline less efficacious and perhaps even makes it dangerous if used intravenously. We want to draw special attention to this detail as this has not been noted in the literature before. Some of the failures of the intravenous saline are the result of want of attention to this detail.

The alkaline saline is prepared from the isotonic saline. It is not desirable to heat sodium bicarbonate in solution with sodium chloride. Sodium bicarbonate is weighed in 160 grain portions, put in paper packets and sterilized in a glass jar also covered by paper in the autoclave at 120°C. in such a way that they will not be damaged by condensing water. Just before use one of the powders of the sterilized sodium bicarbonate is dissolved in a pint of the sterilized isotonic saline. This is the alkaline saline. A glass funnel of suitable size with a filter paper is wrapped in a paper and sterilized in the autoclave. The alkaline saline is filtered through the filter paper into a graduated flask from which it is run into the vein.

For giving the saline intravenously very few extra instruments are required. It is not necessary to purchase the cholera outfits that are in the market. A few graduated funnels with rubber tubing of suitable length and calibre and several silver cannulæ of small and medium sizes provided with stopcocks should be obtained, such as those recommended by Sir Leonard Rogers.* The funnels with rubber tubing and a cannula assembled are wrapped in a towel and paper and then sterilized in the autoclave at 120°C. for 30 minutes. A number of them may be thus kept ready for use when required.

The saline injections must be used rationally, and their application guided by the specific gravity of the blood of the patient. To measure it the usual water and glycerine mixture in bottles must be prepared fresh. Owing to age the mixture in the bottles supplied with some of the outfits is useless. It is essential to have suitable areometers registering specific gravity suitable areameters registering small gravity from 1050 to 1080. About 8 small elongated bottles are required. Glycerine and the distilled water are mixed in a cylinder and the specific gravity is read with the areometer. By addition of more glycerine or distilled water Carbolic acid should be added so as to make its strength in the mixture about 2-5 per cent. This prevents the growth of moulds. The mixture should be made fresh as often as necessary of the mixnecessary. The specific gravity of the mixture should be checked frequently, about once

^{*}Supplied by Messrs. Smith Stanistreet & Co., Ltd.,

a week. In the rainy season due to the absorption of moisture by glycerine it should be checked even twice a week.

For finding the specific gravity of the blood, the finger is pricked with a needle and the drop of blood is sucked up with the help of a rubber teat into a thin glass pipette. It is very important to take special care not to suck into the pipette any air with the blood. The drop of blood is then released in the middle of the glycerine mixture. Special care again is taken not to discharge air bubbles with the blood. Supposing we release the drop of the blood in the middle of the mixture of specific gravity 1068 and we find that it floats, then the specific gravity is less than 1068. But if the drop sinks the specific gravity of the blood is more than 1068. The drop should begin to sink as soon as it is released from the pipette, because even the drop that has risen up previously will begin to sink after the lapse of a very short time.

On admission the degree of the collapse from which the patient is suffering should be ascertained. The use of the sphygmomanometer is helpful but is not indispensable. Feel the radial pulse and see whether it is still perceptible (though it may be very thin and weak) or not perceptible at all. In both these instances immediate intravenous saline will probably be required. It is worth while to feel the radial pulse of both wrists as it may be imperceptible on one side on account of abnormality which, to our surprise, we met with quite often.

Take the specific gravity of the blood always ! This is important. If the specific gravity is below 1058, the patient usually does not yet require an intravenous saline. Patients are, however, not infrequently encountered, especially those who have already had intravenous saline, whose pulse is so weak that judging by the pulse alone one would order an immediate intravenous saline. But on seeing the specific gravity of the blood it is found to be very much below 1058. Now in some of these cases the administration of an intravenous saline is known to have disastrous results due to heart failure. We, therefore, never give an intravenous saline without first ascertaining the specific gravity of the blood and the condition of the heart. Finding the specific gravity of the blood does not take more than a minute. In such cases the intravenous saline is withheld for the time being, and every effort is made to improve the pulse by strengthening the weak heart. We have found that the intravenous or subcutaneous administration of 1/250 grain of strophanthin is very beneficial in bracing up the heart. This is our routine in treatment of the failing heart in cholera. The subcutaneous administration of 10 c.c. of 10-20 per cent. camphor in oil is also helpful.

Because in cholera several pints of saline are to be administered it is much better to

cut down on the vein. Finishing off the flow of several pints of saline into a vein by a prick of needle through the skin may be successful in some instances, but it is usually fraught with difficulties. We, therefore, as a routine cut down upon the vein. Any prominent vein of good size may be chosen. The veins in front of the elbow usually suffice in most cases. No anæsthesia is required for the trivial operation. The vein is made turgid by tying a piece of bandage cloth on its proximal side. The skin incision should be made parallel with the vein but not directly over it. With the thumb of the left hand pull the skin over the vein to one side and holding it there incise through it with the knife. The skin and the subcutaneous tissues are incised with a single sweep of the knife. When the skin is released the incision in it comes to lie directly upon the vein which shines a distinct blue colour and is directly in view. Very little further freeing of the vein will be necessary if all the subcutaneous tissue has been cut by the first incision. A director is placed under the vein and two catgut ligatures are passed underneath it. The distal ligature is tied over the vein and cut short. Next pour the hypertonic saline into the graduated funnel and let it run through the cannula to ensure that the flow is unimpeded and all air has been driven out. This is a very important detail. Every care should be taken to ensure that there is no air in the tubing. Fill the graduated funnel early to allow time for the fluid to fill the tubing completely. Then let a good deal of the saline run from the cannula. Air will be noticed squirting out, so continue to run until there is no more squirting of the air and the flow becomes a continuous regular stream. Now close the stopcock of the cannula. The graduated funnel must be held on a stand. Avoid changing its height because this may let air get into the vein. Hold the front wall of the vein with fine forceps and snip the vein with sharp scissors. Open the stopcock of the cannula and let some of the saline run out of it again, then introduce the cannula through the flap-like incision into the vein while the saline is still running out of it. By doing so the running saline washes the blood oozing from the rent of the vein, admitting a clearer view of it and at the same time prevents air from being driven into it. Do not loose the hold on the vein with the forceps until the cannula is introduced.

When the cannula is in the vein, tie it in the vein with an incomplete surgical knot of the proximal catgut ligature which was previously passed under the vein. If the graduated funnel is now filled with saline up to the top the downward flow can easily be seen in the narrow neck of the flask, if it is going freely into the vein.

The amount of saline to be given intravenously should be arrived at with discretion, paying attention to the condition of the heart, the lungs and the specific gravity of the blood. As a rule we give to all adult patients three pints of the hypertonic saline, and finish it off with an additional pint of alkaline on the first sitting. We seldom give more than four pints of hypertonic and one pint of alkaline saline at a time, and that also we give only in cases of very high specific gravity of the blood of about 1068 or over. If the specific gravity of the blood is below 1058, but the patient is pulseless, defer intravenous saline and try to brace up the heart with 1/250 grain of strophanthin hypodermically, or 10 c.c. of camphor oil also given hypodermically. If the patient still remains pulseless intravenous saline may be given but in small amounts not more than 2 to 3 pints in all. For children, of course, the amount of saline to be given intravenously will be varied as the case may be. If the patient collapses again after a previous saline and the specific gravity is not unusually low, showing the weak pulse to be due to heart failure, we do not hesitate to give another intravenous saline as many times as necessary, though the amount of saline may now be reduced (2 to 3 pints in all). The alkaline saline may however be omitted in order not to overdose the patient with alkali. If you are not sure that the failing pulse is due to a week heart, give the benefit of doubt in favour of saline and give saline even if the specific gravity is below 1058. When about 10 to 15 ounces are run in and the pulse improves, 2 to 3 pints in all can then be confidently given with very beneficial results. If, however, the pulse does not improve stop the saline.

The temperature of the saline to be given intravenously should receive special attention. We never give the saline at a temperature higher than the normal body temperature (98.4°F.). We have never had an occasion to regret having given it even at a lower temperature in all cases irrespective of the rectal temperature. On the contrary we have had disastrous results by using intravenously even slightly warmed saline. In countries where cholera is prevalent the room temperature will in almost all cases be found to be adequately warm for such purposes.

During the course of the administration of saline intravenously there is often severe shivering. It may occur earlier with cold saline but we have never considered such shivering to be of much consequence. It usually passes off quickly without untoward results.

As to rapidity of the flow we think it to be very important and must be carefully looked to. Three pints in 45–60 minutes is quite a good rate, because the vaso-constriction often sets in at about this time, rendering the flow slower and difficult. But the flow should not be accelerated for fear of the vaso-constriction. The rate of an ounce a minute should rarely be exceeded. After the saline is finished and the cannula is taken out of the vein, the incomplete surgical knot is tightened, completed and cut short. After the application of iodine the skin incision is closed by one or two sutures of silkworm gut. The wound is dressed and bandaged.

Every cholera patient must be watched constantly from the time of admission until he is out of danger. The temperature should be recorded every hour for six hours after the intravenous saline. One ought to be on special guard against hyperpyrexia. If the body temperature tends to rise above 103°F. it should be kept down by cold sponging and an ice cap.

If the nursing is satisfactory and the patient is seen frequently, an attendant being always present, hot water bottles may be used with great benefit during the initial collapse and the shivering stage of the intravenous administration of saline. During the reaction stage the hot water bottles should be removed. During the first 24 hours intravenous saline may be needed repeatedly even 2 hours after the previous one. It is very important indeed to watch the patient constantly night and day until discharged. A dangerous collapse may occur even on the 5th or 6th day of the disease. Besides there is always the danger of uramia.

With the intravenous use of alkaline saline the danger of uræmia will largely be averted. After the acute stage is over, in a certain proportion of cholera cases uræmia will still have to be reckoned with. During convalescence, measuring the amount of urine passed in 24 hours is of considerable value. If 2 pints of urine are passed in 24 hours the patient is most probably out of danger. But if, however, the quantity of urine passed in the 24 hours is inadequate, the patient should be carefully watched for any signs of uræmia which usually manifest themselves early in the shape of mental confusion, dullness and restlessness. On the approach of these early signs an intravenous administration of a pint of the alkaline saline usually wards off an attack of fatal uræmia. With the use of the alkaline saline in this manner, uræmia has ceased to be of much concern to us.

We have found that the use of sodii bicarbonas by the mouth and dry cupping of the kidneys is of real help in establishing free flow of urine.

(2) Bacteriophage.

The administration of bacteriophage is very simple indeed. Our bacteriophage is stocked in 50 c.c. bottles and in 5 c.c. glass ampoules. On admission one of the bottles containing 50 c.c. of the bacteriophage is placed near the patient. He is given bacteriophage by the mouth in about drachm doses every 30 minutes. The bacteriophage is given undiluted and sipped directly from the bottle. It is an almost tasteless and odourless clear fluid. When taken in those doses by the mouth it is usually retained and does not induce vomiting. If a dose is vomited due to the usual vomiting of cholera, it may still be given undiluted as usual. If, however, it is believed that the vomiting was induced by the dose of bacteriophage then it should be given in smaller sips. Two bottles of bacteriophage each containing 50 c.c. of the bacteriophage will thus be finished in the following 16 hours. After the first 24 hours of admission a bottle in 24 hours for the next 48 hours will usually suffice.

It will be seen that in this manner an enormous amount of bacteriophage has been given to the patient. We have found the bacteriophage to be entirely harmless. It can be taken ad libitum.

If the bacteriophage is given intravenously there is evidence to show that it appears much more quickly in the stools, than when given by mouth. There is also evidence to show that there is a kind of chemotaxis between the bacteriophage and the microorganism susceptible to its action. Due to this property the bacteriophage present in the blood stream is attracted to the micro-organisms susceptible to its action and in the case of cholera the bacteriophage, given intravenously, is concentrated in less than 2 hours in the intestines—the site of the proliferation of V. choleræ.

We, therefore, give bacteriophage by the intravenous route also. In this case, however, we do not give more than 5 c.c. of the bacteriophage and that very considerably diluted, because there is the possibility of anaphylactic shock on account of the foreign proteins present in the broth used for preparation of bacteriophage, although we have never had any such misadventure. Apart from this, as our experiments on guinea-pigs have shown, the bacteriophage by itself has no toxic action at all. The only safe way to administer bacteriophage intravenously is to give it along with the saline. Used in this way it never produced any reaction. A five cubic centimetre ampoule of the bacteriophage is broken and the contents poured into the hypertonic saline and thus given intravenously with it.

Intravenous or subcutaneous injections of undiluted bacteriophage should never be given as they are liable to produce definite anaphylactic shock, of passing but rather unpleasant character.

We have no experience of the effects of administering bacteriophage per rectum. We have not tried this method so far and we are, therefore, not in a position to express any opinion about it.

After the administration of bacteriophage the stools rapidly become free from V. choleræ. Most of the vibrios are destroyed and the few that escape are usually of atypical kind. It appears that they lose their pathogenic power. At the same time there is marked improvement as regards the diarrhœa. The stools become less frequent, less abundant, contain fewer flakes and rapidly become fæcal in nature. With the improvement as regards diarrhœa further necessity for an intravenous saline does not arise. As the diarrhœa lessens there is rapid improvement in the general condition of the patient. It is remarkable how soon very good appetite returns. With the bacteriophage the recovery is very much quicker than without it. The period of convalescence is considerably shortened. In about 3 or 4 days' time the patient is much better and asks for permission to go home.

The bacteriophage is the only remedy we give internally. The patient is given cold water to drink *ad libitum*, only care is taken not to give it in unusually large quantities at a time in order to prevent unnecessary vomiting. Ice may be given freely to be sucked.

All other drugs are avoided. No potassium permanganate is given in water or in pill form. Essential oils, camphor, calomel, etc., are not used at all.

No food is given for the first 48 hours. If after this there is satisfactory improvement barley water and whey or fruit juices may be given. If these do no harm and the diarrhœa improves milk may be allowed. Then semisolid food and finally ordinary diet. Unnecessary prolonged fasting enhances the acidosis and should be avoided in cases with a tendency to uræmia.

A word of warning about the cholera bacteriophage should be given here. It is necessary that the bacteriophage should be of the highest virulence possible. Besides, it is also very important that it should have a very wide range of action. The bacteriophage we have been able to produce so far is of high virulence and its range of action is fairly wide, that is to say it destroys all the strains of V. choleræ we have met with up to now in the epidemics in various parts of India. Its range of action, however, is not extensive enough to complete our satisfaction. We cannot deny the possibility of coming across strains of cholera which are not acted upon by our bacteriophage. Several such examples we encountered previously during the car festival of Jagannath in Puri. In such cases of course the beneficial results of the bacteriophage are wanting. We hope, however, to surmount these difficulties, as our technique for the preparation of bacteriophage improves. We hope to prepare a highly virulent bacteriophage that will attack all the strains of V. choleræ met with throughout India.

A PLEA FOR THE LOWER UTERINE SEGMENT CÆSAREAN SECTION.*

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IN 1925, during a surgical visit to America, I saw Professor de Lee doing the operation in Chicago of which he is the sponsor. I was immediately impressed by the importance of this operation to India for, as you are aware, antenatal care is only in its infancy in this Peninsula and as a result dire obstetric difficulties are not realized until the patient has been long in labour with the result that the woman is not taken to hospital until her life and that of the baby are in danger, and certainly not before she has had multiple examinations.

On returning to India I at once began to teach and perform this operation. Since 1925, 75 lower uterine Cæsarean sections have been performed at the Eden Hospital and in private practice, and of the hospital patients 48 arrived late in labour after many examinations had been made outside, and in 28 the membranes had ruptured.

Out of the 75 cases there were 11 maternal deaths and 10 fœtal deaths-3 of these were stillborn and 7 died within a few days. This mortality may seem excessive, but when you consider that 48 of the cases were essentially septic at the time of operation, and that 62 per cent. had been in labour for many hours, I think you will agree that these statistics are worthy of your serious consideration; for only a few years back a large proportion of these septic cases would have been treated by craniotomy or by the older method of classical Cæsarean section with complete eventration of the uterus. I have been associated with the Eden Hospital now for 20 years and can well remember the disastrous results to mother and baby of this eventration operation and, I can assure you, that before the War 50 per cent. of the mothers who suffered this operation, after they had been hours in labour and were septic due to multiple examinations or trauma, died of general peritonitis due to the spill of septic contents of the uterus into the peritoneal cavity.

Nobody is more aware than I am that a mortality of 11 out of 75, that is, 14.66 per cent., is grievous, but until such time as we can induce patients to come to hospital early, or can rely on efficient antenatal supervision, maternal mortality due to obstructed labour must remain great in India.

^{*} Being a paper read at the Medical and Veterinary Research Section of the Indian Science Congress held at Nagpur, January 1931.