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AN EXPERIMENTAL INVESTIGATION ON DIARRHOEA AND VOMITING OF CHILDREN[1]

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Synopsis.

I. *The Object and Basis of the Research.*

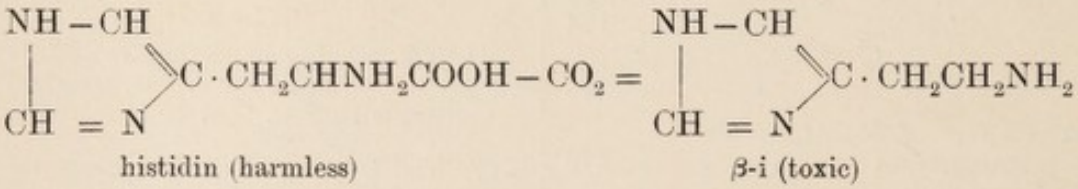
The following is a preliminary account of experiments undertaken in order to elucidate the condition of diarrhoea and vomiting of children. From a bacteriological point of view, this pathological condition has been extensively

[1] Giving results of an investigation subsidized by the Local Government Board.

investigated ; one has but to recall the association of diarrhoea and vomiting by various workers with the Bacillus enteritidis sporogenes, streptococci, Morgan's bacillus, Shiga's bacillus, and many others, to realize this. Much work has also been done on the bacteriology of milk, in order to demonstrate a relationship between diarrhoea and vomiting of children and infected milk. In consequence of the marked difference in the structure and properties of the bacilli isolated in different epidemics, it has been thought probable that, after all, there may be no specific bacillus responsible for the disease, and that, with increasing knowledge of the chemistry of the alimentary tract, there was room for an attack on this important subject along lines different from those which have already gained so much attention. My own work here described is, therefore, an account of an investigation undertaken along chemical lines, and this paper treats only of experiments dealing with the behaviour of animals when under the influence of a toxic substance normally present in the alimentary tract. (In a later paper, I shall describe the effects of the treatment of children suffering from diarrhoea and vomiting, the treatment following along lines indicated by this experimental work.) I do not propose in this paper to discuss the many important researches carried out by other workers on this disease along bacteriological and other lines not germane to the present investigation.

In the first place it may be well to state certain fundamental facts which bear upon this work.

Some years ago, an extremely active and toxic substance was isolated by Barger and Dale (1) from ergot. This substance is known as β-imidazolyl-ethylamine (which throughout this paper will be called β-i). The formula of this substance makes it clear that it is derived from histidin by the simple removal of carbon dioxide thus :



histidin (harmless) β-i (toxic)

Other substances, less active physiologically, now known as tyramine and isoamylamine were also obtained from ergot by the same workers, and the chemical constitution of the substances indicates that they are derived from tyrosin and leucin respectively, in a similar way to that by which β-i is derived from histidin.

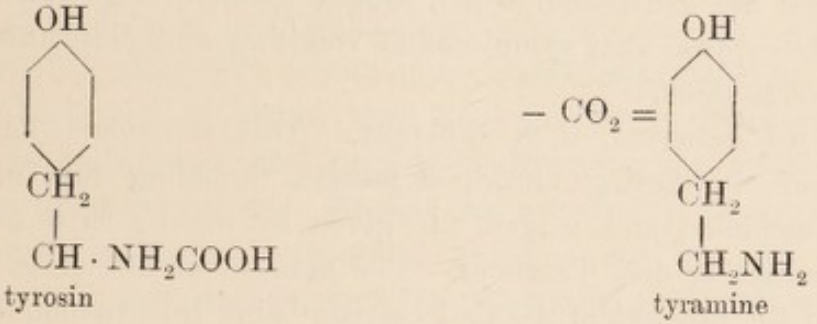
The next interesting discovery was also made by Barger and Dale (2) when they demonstrated the presence of β-i in the alimentary canal of normal herbivora. They were led to investigate this point by the well-known fact that a watery extract of the mucous membrane of the intestine has a marked depressant action on the blood-pressure, and in other ways has physiological effects similar to those produced by β-i.

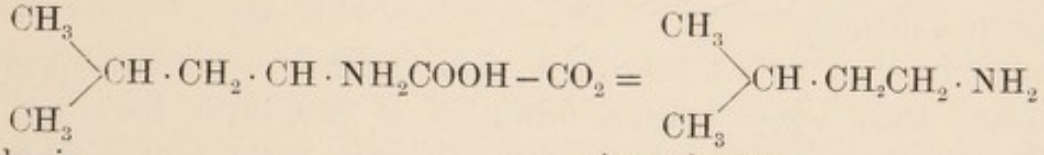
It is interesting to note that β-i may be placed in the same category as

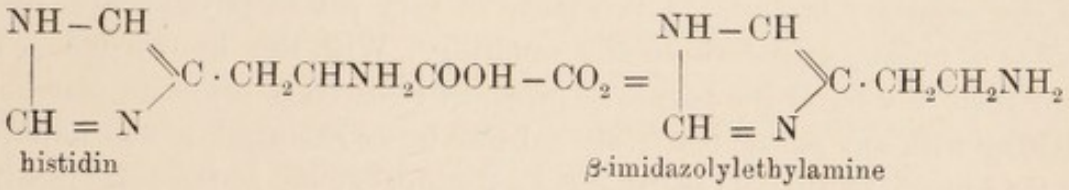
other ptomaines whose names are so familiar, viz. cadaverin and putrescin. The formulae of the two substances are:

$NH_2 \cdot CH_2CH_2CH_2CH_2CH_2NH_2$ cadaverin,
$NH_2 \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot CH_2 \cdot NH_2$ putrescin.

Their formulae show them to bear a close relation to lysin and the diamino-valerianic acid portion of arginin respectively. The similarity of formation of all these ptomaines can thus be seen:







$COOHNH_2CHCH_2CH_2CH_2CHNH_2COOH - 2\,CO_2$
lysin

$= NH_2CH_2CH_2CH_2CH_2CH_2NH_2$
cadaverin

$COOHNH_2CHCH_2CH_2CHNH_2COOH - 2\,CO_2 = NH_2CH_2CH_2CH_2CH_2NH_2$
diamino-valerianic acid (from arginin) — putrescin

The change from lysin and diamino-valerianic acid to cadaverin and putrescin was shown by Ellinger (3) to be the result of anaerobic bacterial decomposition. Similarly, Akermann (4) was able to produce β-i from histidin. Twort and I (5) have published a method for the isolation of bacilli capable of converting histidin into β-i. This bacillus was isolated from the contents of the intestine of human beings and other animals. The difference between the results of Ellinger and ourselves is that the removal of CO_2 from histidin does not at all depend on the presence of oxygen and goes on aerobically and anaerobically, while Ellinger's results depended on anaerobic conditions. The difference is probably to be explained by the fact that Ellinger carried out his experiments with a very mixed growth, which aerobically destroyed the amines as they were formed, while we worked with a single strain of bacillus, and so had

no such complicating factors. We did find, however, that one important factor influenced the change, namely, the presence of acid. In an acid medium, or what comes to the same thing, in the presence of carbohydrate—for bacteria always produce acid if sugar is present—the end product of the bacterial action on histidin was quite inactive physiologically, and certainly was not β-i.

While putrescin and cadaverin have been mentioned in order to show the analogous position they hold as compared with β-i, and that this last substance can also be classified in the ptomaine group, it is necessary to add that cadaverin and putrescin are, compared to β-i, almost inactive substances. It is, in fact, impossible to suppose that symptoms of vomiting and diarrhoea can, in any case, be ascribed to them.

In the case of β-i, however, it is different. This substance has an extraordinary power of stimulating unstriped muscle, including that of the alimentary canal, bronchioles, and uterus. If given by mouth to a cat, it immediately causes vomiting and diarrhoea. The amount of diarrhoea is of course proportional to the amount of β-i that escapes being thrown out of the stomach in vomiting.

Here, then, we have a substance present in the mucous membrane of the alimentary canal, whose constant formation, at least in the lower end of the intestine, is almost certain because of the presence of the amino-acid histidine and the necessary bacteria; a substance of very potent physiological activities, capable of producing diarrhoea and vomiting. With this knowledge, it seemed to me that, in view of the failure of previous research to associate diarrhoea and vomiting with any causal factor, either bacterial or chemical, it was important to test the hypothesis that β-i is either the substance, or one of the substances, which plays an important part in the production of such symptoms. Various hypotheses involving β-i as the cause of such symptoms can be formulated:

1. That in diarrhoea and vomiting there is excessive production of β-i in the intestine due either to:

   (*a*) A changed or increased bacterial flora;

   (*b*) A changed chemical or physical condition which prevents the absorption of histidine of the food and allows a subsequent increased production of β-i; or to

   (*c*) A changed condition which allows bacteria acting on histidin to produce a relatively large amount of β-i rather than innocuous products (such an innocuous product might be β-imidazolylacetic acid).

2. That in diarrhoea and vomiting there may or may not be an increase of β-i, but the β-i, present in the alimentary canal and mucous membrane, becomes active in some way, and is absorbed into the blood-stream at a time when the animal is incapable of resisting its toxic action and rendering it innocuous.

The experimental portion of this work has therefore been carried out on the assumption that β-i is one of the most important factors in epidemic diarrhoea and vomiting (I do not wish to imply it is the only one). Work has been done to discover the conditions under which it is absorbed from the intestine,

the conditions which prevent its absorption, the conditions which allow of its rapid decomposition after absorption into the blood-stream, and, on the other hand, the conditions which allow it to have its full toxic action on the animal.

Even if further research should show that β-i is not the important chemical agency at work in this condition, it is felt that many of the facts to be described will apply equally to other toxic substances and that the immunity against and liability to suffer the full toxic action of physiological substances, as met with in the animal body, follow general rules.

The problem is then resolved into two parts : (1) How to diminish or prevent the absorption of toxic substances from the intestine while at the same time interfering with other physiological activities as little as possible. (2) How to raise the immunity against such toxic substances as are absorbed.

In the experimental work to be described, the former of these problems has taken up the most time. At first this appeared to be the more important, and it is certainly the easier problem to be worked at. As the work progressed, however, it must be confessed that the second part impressed itself more and more upon me, and, although definite results were obtained along these lines, there remain many points to be cleared up. I feel confident that a solution of the problem as to the different powers of resistance to a toxic substance, possessed by animals under varying conditions, is most vital, not only from the point of view of epidemic diarrhoea and vomiting, but also in the case of all conditions of toxaemia. Why a small quantity of toxic substance should in one animal cause fall of blood-pressure, paralysis of the respiratory centre, and death, and in a second animal of the same type leave the animal practically untouched, seems to me to be the crux of a great number of pathological conditions.

In my opinion, the reasons for which I will give later, a child suffering from diarrhoea and vomiting is in a condition for experiencing the full effect of a toxic substance, so that the absorption into the blood-stream of a small quantity of β-i is capable of producing the full toxic action, while in a normal child a similar quantity would leave the child untouched.

The reasons, therefore, why β-i was chosen as a suitable substance in the following experimental work, and why, also, it is considered an important factor in diarrhoea and vomiting of children, are as follows :

1. β-i is present in the intestinal mucous membrane of normal animals (Barger and Dale).

2. Bacilli have been isolated from the intestine capable of producing β-i from histidin ; also from meat extract, in this latter case from carnosin (histidyl-alanin) (Twort and Mellanby).

3. β-i is a toxic substance (6) capable of causing—

(*a*) Diarrhoea and vomiting,

(*b*) Fall of systemic blood pressure,

(*c*) Depression of the respiratory centre,

(*d*) Coma,

that is, symptoms met with in diarrhoea and vomiting of children.

II. *Experimental Procedure.*

For the animal experiments in this work the cat was used. Various anaesthetics were employed, and, as will be seen later, with markedly differing results. In many of the earlier experiments, urethane was injected about an hour before the experiments were started, 1 to 1½ grm. per kilo of body-weight being used. This, together with a little ether or chloroform or A. C. E. at the beginning of the operative procedure, sufficed to keep the animal anaesthetized for as long as required. In the later experiments, for reasons which will be explained, urethane was discarded in most cases, and chloroform only or A. C. E. was used. After anaesthetizing the animal, tracheotomy was performed, and in most experiments the blood-pressure in the carotid artery recorded. The abdomen was next opened, the large omentum moved back, and loops of gut measured. Lengths (25 cm.) were tied off after the method of Moreau. The region chosen for the loops varied with the experiment. In preparing these, it was necessary to keep the blood-supply and lymphatics as normal as possible, and further not to expose the intestines for longer than could be helped. A measured solution, usually 5 c.c., of β-i of a known strength was then injected into the loops. If the effect of drugs was sought in the experiment, the drug to be tried was injected into the intestine prior to the β-i. The intestines were then carefully replaced in the abdominal cavity so as to avoid twisting of the mesentery, and the abdominal cavity closed up. The animal was then left for a specified time. At the end of this time, the loops of the intestine were removed, opened up and washed out carefully, so as to bring the volume of the washings up to 300 c.c. This was then boiled, and the β-i *not* absorbed during the experiment estimated. To estimate the β-i, it was not possible, with the small quantities used, to find a chemical method. A biological method was therefore employed. This method was one used by Kehrer (7), and consisted of suspending one horn of the uterus of a guinea-pig in oxygenated Ringer solution, and finding the smallest quantity of solution capable of causing a full contraction of the uterus. This quantity was compared with the amount of the original β-i solution capable of causing a full contraction, from which data it could be easily calculated how much β-i had disappeared from the loop during the experiment. If, for instance, it took 0·3 c.c. of the original solution, and 0·6 c.c. of solution after being in the intestine, to bring about the same change in the guinea-pig's uterus, it can be readily seen that 50 per cent. of the β-i originally used must have been absorbed.

The advantages of the method are :

1. It is fairly rapid with a suitable uterus.

2. It is very delicate and is therefore suitable for small quantities of β-i.

The disadvantages of the method are :

1. Every uterus varies in susceptibility, so that the contents of all loops of intestine together with the control in each experiment must be tested on the same uterus.

2. The uterus varies in irritability during the experiment, and this can only be eliminated by repeated trials.

3. The method varies in accuracy of results at different times, and can never be as good as a reliable chemical method.

In spite of the obvious disadvantages, good relative results can be obtained by taking care that such conditions as temperature, constitution of Ringer solution, and times of injecting the drug are kept constant.

In the majority of cases two cats were experimented on, one being a control animal. For a long time, until something was learned as to the susceptibility of the animals to β-i and the conditions which regulated such susceptibility, very great difficulty was experienced in making the experiments comparable and successful. On account of the differences between animals, it can be readily understood that each experiment has to be repeated several times before any result can be accepted, and the animals have to be obtained in similar conditions for similar experiments. For instance, it is useless to test the absorptive capacities in cats, one of which had been fed on milk and the other on meat. In all experiments, except where other points were being tested, the cats were kept without food for twenty-four hours prior to the experiment, so that the small intestine was clear of food when required. Except in one or two cases where it is otherwise stated, it can be assumed that several experiments were made to prove a given point, although details of all such experiments are not given in the protocols.

III. *Preliminary Experiments.*

(*a*) *The disappearance of* β-i *from the small intestine during the time of the experiment depends only on absorption.*

In experiments to test this point the blood-vessels in the mesentery supplying loops of 25 cm. of intestine were tied before the β-i was injected. The animals were then left for varying periods of time and the amount of β-i remaining in the intestine was compared with the original β-i solution injected. No β-i could be absorbed in such a case, and if there was any deficiency in the β-i after its sojourn in a loop of intestine, this must have been brought about by bacterial decomposition. In Experiment 71 the β-i solution remained in the intestine for two hours and five minutes, after which the cat was killed. It will be seen that there was no loss of β-i in this time.

Inference: There is no disappearance of β-i from the small intestine when the mesentery is ligatured.

(*b*) *The independence of the absorption of* β-i *in each loop of the intestine.*

The number of instances in which the activity of one part of the alimentary canal depends upon that of another part might lead one to expect that absorption from one loop of the intestine would be influenced by the condition of the

proximal loops. Without going so far as to deny some interdependence between neighbouring parts of the intestine, it is clear that cutting off the blood supply above the experimental loop does not do away with the absorbing power of that loop.

In Experiments 71 and 73 the mesenteric vessels supplying loops of intestine 25 cm. in length were tied at various parts of the alimentary canal. It will be seen that this interference with the blood supply did not prevent 70, 47, 55, 44, and 50 per cent. of the β-i injected disappearing respectively from other loops of small intestine with the blood supply intact.

Inference: The absorption of β-i is not affected by tying the blood-vessels of a part of the intestine other than that in which the β-i is placed.

(c) *The increasing rate of absorption in passing from the duodeno-jejunal flexure to the caecum.*

The absorption of β-i from the small intestine increases from the duodeno-jejunal flexure to the caecum. The difference in the rate of absorption between the extreme ends of this part of the small intestine is marked, but becomes less so as the loops approach one another.

In Experiment 52 it will be seen that 59 and 60 per cent. of the injected β-i were absorbed from loops of intestine at the duodeno-jejunal flexure, whereas 84 and 89 per cent. disappeared from similar loops at the caecal end of the small intestine in the same time.

The maximum rate of absorption is always attained at the lower end of the small intestine.

In Experiment 134 the loops of intestine in each cat are placed nearer to each other. Loops A_2 and B_2 were at the caecal end of the small intestine, and loops A_1 and B_1 25 cm. distance higher up than A_2 and B_2. In the time allowed for absorption, viz. 1 hour 18 minutes, 50 and 55 per cent. of the injected β-i disappeared from A_1 and B_1 and 55 and 66 per cent from the lower-placed loops A_2 and B_2.

Inference: The nearer the caecum, the greater is the absorptive capacity of the small intestine.

IV. *The Effect of Food-stuffs and Bile on the Rate of Absorption of* β-i *from the Small Intestine.*

In this place I wish only to refer to the effect of feeding on the rate of absorption of β-i in the small intestine. Later, it will be seen that feeding of some food-stuffs has also an important effect on the resistance offered by the animal to the β-i after absorption.

(a) *The effect of milk.*

If milk is given to a cat some hours prior to the injection of β-i, then the rate of absorption of the β-i from the intestine is diminished.

For instance, in Experiment 123, 100 c.c. of milk were given to a cat 4 hours 30 minutes before the β-i was injected. In this cat, 50 per cent. of the β-i was absorbed in two hours, while in the control cat 66 per cent. was absorbed.

Experiment 37 shows a similar result.

Milk, therefore, depresses the absorption of β-i from the intestine.

(*b*) *The effect of meat.*

A cat digesting and absorbing meat also absorbs β-i at a slower rate than a non-digesting cat.

This fact is seen in Experiments 125 and 129.

In Experiment 125, 30 grm. of meat (without fat) were eaten 1 hour 50 minutes before the β-i was injected. During the time of intestinal absorption, 1 hour 30 minutes, the meat-fed cat absorbed 36 per cent. of the injected β-i, against 46 per cent. in the control non-digesting cat.

In Experiment 129 the meat was eaten 4½ hours before the injection of β-i into the intestine. The time of absorption allowed was 2½ hours. The meat-digesting cat absorbed 70 per cent. of the β-i, and the control cat 75 per cent. This difference is small, but the time allowed for absorption was long, 2½ hours, and this length of time always tends to nullify effects, because of the natural slowing in the rate of absorption that takes place in such experiments as these. Consequently, in course of time, the amounts of β-i remaining unabsorbed in all loops tend to approximate closely.

During the digestion of meat the rate of absorption of β-i from the intestine is diminished.

(*c*) *The effect of fat.*

Fat is known to have such a marked inhibitory action on intestinal functions that it might be expected that it would have an inhibitory action on the absorption of toxic and other substances. For instance, fat depresses the movements of the stomach and alimentary tract, and also inhibits the secretion of gastric, and therefore of pancreatic, juices.

The experimental work here described demonstrates that fat also depresses the absorption of β-i from the intestine, but the effect is not so marked as one might expect.

In Experiment 145, 20 grm. of fat were eaten by cat *A* six hours before the β-i was injected into the intestine. In one hour, 36 per cent. and 30 per cent. of the β-i were absorbed in cat *A*, while in the control non-digesting cat *B* 42 per cent. and 42 per cent. disappeared from corresponding loops of intestine. Similar results were obtained in other fat-feeding experiments quoted in protocols.

The question now arose as to whether the inhibitory action of fat on absorption was due to a local effect on the absorbing portion of intestine, or whether it was due to some previous action in its passage along the intestine,

in which latter case the action might be considered indirect. In order to test whether the fat action was direct or indirect, other experiments were performed in which olive-oil emulsion was placed directly into the loops of intestine before injecting β-i solution into the same loops.

Experiments 144 *a* and 144 *b* demonstrate that fat placed directly into the intestine has as large an inhibitory action as when the fat is eaten. In these experiments it is seen that 60 per cent. and 60 per cent. of the β-i are absorbed from the loops also containing olive oil, while the control loops without fat absorb 81·6 per cent. and 70 per cent. of the β-i respectively. It is probable therefore that the inhibitory action of fat on absorption is a local and direct one.

Inference :

1. The absorption of β-i from the small intestine is slightly delayed if the animal is previously fed on fat.

2. This action of fat is a direct one on the epithelium of the absorbing portion of intestine.

(*d*) *The effect of carbohydrate.*

The result of this portion of the work is not clear, for the direct feeding of pure carbohydrate to cats before the injection of β-i into the intestine was not tried. Instead of this, a solution of dextrose was injected into the intestine, and in all the experiments performed there was a marked diminution in the rate of absorption of the β-i. In further control experiments, in which water was injected into the intestine instead of a dextrose solution, the inhibited rate of absorption was also marked, so that clearly it is not possible to state that carbohydrate in the intestine had in itself an inhibitory effect.

The effect of a dextrose solution is seen in Experiment 130. In cat *B* 25 c.c. of a 10 per cent. solution of dextrose were injected into the small intestine 15 minutes before the β-i solution was injected. Cat *A* was the control, and only received the β-i solution. 70 per cent. of the β-i was absorbed from the intestine of *A* in 1½ hours, while 49 per cent. was absorbed from the dextrose cat. Here, then, is a marked inhibitory effect apparent in the dextrose cat.

(*e*) *The effect of water.*

That the foregoing effect may be due to the water of the solution and not the dextrose is apparent from Experiments 57 and 58.

In Experiment 57, 60 c.c. of Ringer solution were injected into the intestine at the duodeno-jejunal flexure of cat *A*, 40 minutes before the β-i was injected. No fluid was injected into *B* except the β-i solution. *A* only absorbed 33 per cent. of the injected β-i in 1 hour 40 minutes, compared to the 70 per cent. absorbed in *B*.

In Experiment 58, where 25 c.c. of Ringer solution were injected into *A*, a similar, but not so pronounced a result, was obtained.

Inference: The inhibitory action on the absorption of β-i from the intestine, evident when a carbohydrate solution has been previously placed in the intestine, cannot be ascribed to the carbohydrate itself, because Ringer solution has a similar marked inhibitory action.

(*f*) *The effect of bile on the absorption of* β-i.

While discussing the effect of food-stuffs in the intestine on the absorption of β-i, it is right that the effect of bile should be considered at the same time. For it is evident that one of the effects of food passing from the stomach into the duodenum is a liberation of bile. It is necessary therefore to see whether the increased flow of bile may account for some of the effects previously demonstrated.

From another point of view, the inquiry into the effect of bile is interesting. In diarrhoea and vomiting of children, one of the results must be the ultimate deficiency of bile salts and other bile products in the child's economy, following from the persistent intestinal condition. It is further evident that the body values its bile salts, for otherwise the cycle of events which it employs, namely, the excretion by the liver and subsequent reabsorption into the portal circulation, would not occur with such persistence and regularity. The elimination of bile may therefore be of some importance in the aetiology of diarrhoea and vomiting.

In addition to these facts, the important part played by bile in allowing purgatives like aloes and rhubarb to have their full physiological effect seems to indicate that it may have some action on the absorption of substances from the intestine.

In the experiments performed in this connexion it will be seen that bile has some inhibitory action on the absorption of β-i from the intestine, and there is also evidence that this action depends on the bile salts present.

In Experiment 105, 1 c.c. of the cat's own bile withdrawn from the gall bladder was placed in each of two loops of the intestine, while in an intermediate loop no bile was injected. In 2½ hours 66 per cent. of the injected β-i was absorbed from the control loop, while 50 per cent. and 34 per cent. were absorbed from the bile-containing loops.

In Experiment 106, where ½ c.c. of bile was injected into one loop, 60 per cent. of β-i was absorbed from this loop containing bile in the time that 90 per cent. disappeared from the control loop.

We see, therefore, that bile depresses the rate of absorption of a toxic substance like β-i. It may be added that the results obtained with bile at the duodeno-jejunal end of the small intestine were not consistent, and the effect in any case was not so clear as at the caecal end.

Experiments 108 *a* and 108 *b* further indicate that bile salts alone are capable of inhibiting the action of β-i. In these experiments it will be seen that less β-i has been absorbed from those loops which contain bile salts than from those without bile salts. One must be careful in the use of bile salts, for they

have such an irritant action on the intestinal mucous membrane that it is easy to cause an inflammatory condition in the intestine by means of them, and this inflammation itself will naturally depress absorption. An effort was therefore made to keep the amount of bile salt in the intestine at an amount likely to be met with normally, and below the amount capable of producing an inflammatory condition of the epithelium. In the bile-salt experiments a solution containing 6 per cent. sodium glycocholate and 2 per cent. sodium taurocholate was used, and 0·25 c.c. was injected into the loop of intestine with the 5 c.c. of 1 per cent. β-i.

The results here described seem to me to be important from a practical point of view, and it may well be that the deficiency of bile salts will prove to be one of the factors worthy of further consideration in the case of diarrhoea and vomiting of children.

Whether bile salts play any such part as this in normal life seems to depend, in view of the above results, on whether they are absorbed back into the blood-stream in the upper or lower part of the small intestine. If they are normally absorbed for the most part from the jejunum, then it would not appear that they will have much opportunity of exerting any inhibitory influence on the absorption of toxins formed in the ileum. Two further points are suggested by these results:

1. It has been seen that all the feeding-experiment results indicate that food in the intestine inhibits the normal absorption of β-i. The result of feeding is always to increase the amount of bile in the intestine, and the depressed absorption of the β-i may therefore be due to the bile present.

2. The presence of bile in the intestine, even in localized patches, as is evident by the pigmentation, is a condition which may be met with in any intestinal experiment. Consequently, one has always to be careful in observing this factor in absorption experiments.

Summary. In this section it has been demonstrated that the presence of food-stuffs and water in the intestine delays the absorption of β-i contained in a watery solution. An inhibitory effect is also produced when bile or bile salts are present. In other words, an empty non-digesting intestine is in the best condition for absorbing into the general circulation a toxic substance like β-i.

V. *The Effect of* $MgSO_4$ *on the Absorption of* β-i.

The effect of magnesium sulphate on the absorption of β-i from the intestine can be tested either by giving the salt by mouth and then injecting the β-i into loops of the intestine, or by first injecting magnesium sulphate and then β-i into the same loop. The former of these methods is the better, because it conforms more exactly to the normal; but it is very difficult to perform, for, when $MgSO_4$ is given to a cat with an empty stomach, it is immediately vomited. The results to be described were therefore obtained by the second method.

In Experiment 137 it will be seen that 30 per cent. of the β-i injected was

absorbed from the loop containing $MgSO_4$, and 56 per cent. from the control loop containing no $MgSO_4$.

In Experiment 138, 33 per cent. of the β-i in the $MgSO_4$ loop was absorbed, while from the control loops containing no $MgSO_4$ 76, 66, and 69 per cent. of the β-i disappeared. In these two experiments the amount of $MgSO_4$ present was 6 per cent., and undoubtedly with a higher concentration of the salt the depression of the absorption would be greater. On weakening the $MgSO_4$, however, a limit is reached below which this salt has no effect. This can be seen in Experiment 136, where the amount of $MgSO_4$ in the loop was 0·83 per cent., and there is no inhibition in the rate of absorption.

The effect of $MgSO_4$ is no doubt complicated by other factors which I have not up to the present been able to test. Just as its cathartic properties depend upon the amount of fluid in the body, so also it cannot be doubted that the effect of $MgSO_4$ will be considerably greater in a normal individual than in one already deprived of fluid, as is met with in the case of diarrhoea and vomiting. What strength of $MgSO_4$ in an animal with deficient body-fluids is necessary to inhibit the absorption of toxic substances from the intestine can only be ascertained by further work.

Summary. Experiments have been performed to show that a strong solution of $MgSO_4$ (6 per cent.) has a depressant action on the absorption of β-i from the intestine, whereas a weak solution (0·8 per cent.) has no such effect. Experiments with strengths varying between these limits show that a solution as low as 2 per cent. in the normal animal exhibits this depressant effect.

VI. *The Effect of Morphine on the Absorption of* β-i.

Morphine is so often used in cases of intestinal disturbance, and with such beneficial results, that its effect on the absorption of β-i from the intestine was investigated. In these experiments two difficulties arose, each of which made the results more complicated:

1. The direct depressant action of the morphine on the respiratory centre, even in small doses.

2. With this depressed respiratory centre, the animals are on the point of death for some time before actual death occurs, and it is doubtful whether any absorption takes place during this period.

In spite of these difficulties, however, experimental results show that although morphine may have some depressing influence on the absorption of toxins from the intestine, yet the effect is small and more than counterbalanced by more serious results.

In Experiment 134 there is seen the effect of 5 minims of the liquor morphinae hydrochloridi placed in two loops of the intestine. In these morphine-containing loops, 50 and 55 per cent. of the β-i injected were absorbed in the time that 55 and 66 per cent. of β-i were absorbed from the corre-

sponding control loop containing no morphine. The inhibition of the morphine on the absorption is small.

In order to see whether one could completely suppress the absorption of β-i by morphine, Experiment 133 *b* was performed, in which a larger dose, i.e. 1 c.c. of the liquor morphinae hydrochloridi, was injected into the intestine along with the β-i. In this experiment, after a period of 70 minutes, 37·5 per cent. and 40 per cent. of the β-i had been absorbed. It is clear, therefore, that morphine, even in large doses, allows considerable absorption of a toxic substance like β-i from the intestine.

A more serious point of consideration is, however, that in all the experiments performed, the morphinized cats died of respiratory failure. The importance of this will be discussed at the end of the paper.

Summary. Morphine seems to depress slightly the absorption of β-i from the intestine, but this inhibitory action is negligible in view of the dangers of respiratory failure caused by the morphia in these experiments, even when used in comparatively small doses. These experiments suggest that there is considerable danger in the use of morphia in cases of diarrhoea and vomiting in infants.

VII. *The Effect of Alkali and Acid on the Absorption of* β-i.

Differences of hydrogen ion concentration met with in the digestive tract play such an important part in ferment action and other functions of the alimentary canal that it was thought probable that they would also play an important part in absorption and, from the point of view of this research, in the absorption of β-i. Experiments were therefore made to test this point.

Alkali.

Two points of interest are noticeable about these experimental results:

1. Alkali has a greater adjurant action in the absorption at the upper than at the lower end of the small intestine.

2. That the amount of increased absorption due to the presence of alkali varies very considerably at different parts of the intestine and even in the same part of the intestine in different cats.

As for the first point, a possible reason why alkali is more potent at the jejunal end than at the ileal end is that the normal reaction of the mucous membrane at the latter end is more alkaline, and this may be the optimum condition of alkalinity for absorption in this position. It may in itself also explain why absorption from the lower end of the small intestine is normally greater than from the upper end. As regards the second point, the marked difference in the effect of the added alkali is not surprising when one considers the variations in reaction met with in the intestine even in corresponding parts.

In Experiment 120 is seen the effect of alkali at the jejunal end of the small intestine. To one loop was added 1 c.c. of 5 per cent. Na_2CO_3, and from this

loop 82 per cent. of the β-i was absorbed in 2 hours 34 minutes, while 50 per cent. only disappeared in the same time from the control loop containing no alkali.

In the ileum (Experiment 133 *a*) the same amount of alkali, viz. 1 c.c. of 5 per cent. Na_2CO_3, was injected into one loop, and from this loop 69 per cent. of the β-i was absorbed, compared with 62·5 per cent. from the control loop.

It will be seen there is a marked increase in the β-i absorbed under the influence of alkali at the jejunal end, whereas at the ileum the increase is small.

Acid.

One definite remark which can be made about the presence of acid in the small intestine is that, if it is allowed to remain free in the intestine, then there is a marked inflammatory condition set up, the blood-vessels are congested, and the mucous membrane reacts as if to a great irritant, a large amount of mucus being secreted and the mucous membrane denuded. The underlying muscle also becomes congested. It is true that small quantities of acid can be placed in the intestine without irritating the mucous membrane, but this, I think, is due to the presence of neutralizing substances, such as alkali and protein, already present or immediately secreted when the acid is injected. For instance, it is possible to inject stronger acid solutions into the lower end of the small intestine than into the jejunal end without producing the irritant action. This is to be correlated with the larger amount of neutralizing alkali at the lower end.

My results point to the probability that acid stops the absorption of β-i absolutely when it destroys the mucous membrane, and inhibits it only below this point. Lactic acid, 0·6 per cent., delays the absorption of β-i, and an increase of acid above this figure prevents absorption and destroys the mucous membrane. I have obtained no evidence that the addition of acid in any quantity can stimulate the absorption of β-i. It is interesting to see how differently the intestine reacts to acid and alkali, how susceptible it is to the former, an inflammatory condition being easily set up, and how indifferent it is to the latter, even to much larger quantities. The effect of these substances on the absorption of β-i is in keeping with the different results on the mucous membrane which are noticed when acid and alkali are brought into contact with them.

Summary. Alkali stimulates the absorption of β-i from the small intestine, and more especially at the jejunal end, the effect at the ileal end not being so obvious.

Acid has an inhibitory action on the absorption of β-i, and a small concentration completely stops absorption and destroys the epithelium of the mucous membrane.

VIII. *The Effect of altering the Volume of Body Fluids on the Absorption of β-i.*

It is well known that an animal has an excellent mechanism for keeping its blood-volume constant, in spite of conditions tending to produce alterations. Thus, bleeding an animal causes fluid to be withdrawn from the lymph and other fluid dépôts, whereas injecting saline into the circulation causes the excess of fluid to accumulate in the splanchnic and other veins, and subsequently to be passed on to the lymphatic spaces before being finally excreted by the kidneys. It is, therefore, difficult to raise the arterial blood-pressure by the injection of fluid, although, if the arterial blood-pressure is low to begin with, in consequence of a deficiency of body fluid, then the injection of water does have a more pronounced heightening effect on it. It will be seen therefore that the injection of Ringer solution into the blood-stream does not make as important a difference to the arterial and capillary circulation as might at first be expected. Previous experiments described have been concerned only with the effects of the intestinal contents on the absorption of β-i, but it is evident that the blood circulating through the intestine is also of great importance. As to what is the relative importance of the various factors in the blood, such for instance as the blood-pressure, blood-volume, viscosity, and osmotic pressure in regulating absorption from the intestine, very little is known. No great effort has been made in this research to differentiate between these factors, but the experiments described below demonstrate clearly that absorption from the intestine can be readily modified by varying the blood circulation.

(*a*) *The effect of diminishing the body fluids by bleeding.*

When this type of experiment was first attempted, it was with the idea that the condition might be comparable to that of an animal deficient in body fluids, such as is met with in diarrhoea and vomiting of children or when an animal has had fluid withheld from its diet. It is obvious, however, that the conditions set up by withdrawing blood cannot be regarded as equivalent to that resulting from an abnormal loss of fluid through the kidney or alimentary canal, or produced by withholding water from the diet. The bled animal has a deficient cellular and protein element in its blood, and its blood-volume may be quickly restored to the normal by the mechanism mentioned above. Though the experiments are not comparable to the condition aimed at being produced, the results seem to be sufficiently interesting to merit description.

In Experiment 140, 40 c.c. of blood were withdrawn from the carotid artery in one cat. In this cat 50 and 66 per cent. of the β-i injected into the intestine were absorbed, whereas in the control normal cat 75 and 80 per cent. were absorbed from corresponding loops of the intestine during the same time.

In Experiment 141, 15 c.c. of blood were withdrawn from the carotid artery three minutes before the β-i was injected into the intestine, and in thirty minutes 20 and 33 per cent. of the β-i were absorbed, comparing with 33 and 39 per cent. absorbed from the intestine of the control cat.

It is evident from these two experiments that the effect of bleeding is to diminish markedly the absorption of the β-i from the intestine.

To set against this inhibited absorption of a toxic substance, there must however be placed the fact that the bled cat has lost a great amount of its power to resist the toxic substance which is absorbed. For instance, in Experiment 140, the fall of blood-pressure caused by the absorbed β-i in the fed cat was 40 mm. Hg, whereas in the bled cat, although less β-i was absorbed, the fall in blood-pressure was 68 mm. Hg. In Experiment 141 the bled cat died thirty minutes after the injection of the β-i into the intestine.

Experiment 56 shows very well this greatly diminished resistance resulting from bleeding. 30 c.c. and 20 c.c. of blood were withdrawn respectively from cats *B* and *C* before injecting β-i into the intestine. Cat *B* died fifteen minutes, and cat *C* ten minutes, after the injection of β-i. Cat *A*, the control cat with no blood withdrawn, lived $3\frac{1}{4}$ hours after the injection, during all of which time the β-i was being absorbed into the blood-stream.

It is evident from the above-described experiments that bleeding has two effects:

1. It diminishes the rate of absorption of β-i from the intestine.

2. It reduces very greatly the resistance of the animal to the toxic substance absorbed.

I have no doubt in my own mind, from other experiments described in this paper, that an animal deficient in body fluids caused by loss through the alimentary canal or kidney would absorb β-i at a greater rate than the normal animal, and in this respect be different from the bled animal.

Summary. A bled animal absorbs a toxic substance from the alimentary canal at a less rate than a normal animal, but it has but little power of rendering such an absorbed substance innocuous, and rapidly succumbs.

(*b*) *The effect of increasing the body fluids by injecting Ringer solution.*

It can be definitely stated that any large increase of the body fluids caused by injecting Ringer solution into the blood-stream has a definite inhibitory action on the absorption of β-i from the intestine. That is, in the case of cats, the injection of quantities of fluid greater than 100 c.c. has this inhibitory effect. Quantities of fluid below this amount do not appear, from the experimental results obtained, to have such an action. However, it must be remembered that the problem is somewhat complicated, and this may explain the indefinite results obtained when smaller quantities of solution than 100 c.c. were injected into the blood-stream. This complication is as follows: If an animal has but small power of resistance to β-i, as is the usual condition of a control non-digesting animal, the first β-i absorbed from the intestine brings the blood-pressure down very rapidly from 110–120 mm. Hg to 50–60 mm. Hg, the respiratory centre is affected, and the animal may be on the point of death for some time. During this time the further absorption of β-i seems to stop, or is depressed, while the Ringer injected cat, being in a better position for

metabolizing the absorbed β-i, continues to absorb it, and may ultimately, over a definite period of time, absorb as much as the control cat.

When large quantities of Ringer solution are injected, two definite results are obtained : (1) the animal's power of absorbing β-i from the intestine is markedly depressed, (2) the animal has a greatly increased power of resistance to the β-i absorbed.

Before describing the results of experiments performed, it may be well to mention one other important fact. It is useless in such experiments, where the animal has to be left on the operating table for periods over one hour, and where a salt solution is injected into the blood-stream, to use ether, chloroform, or A.C.E. as anaesthetics in cases. When fluid is injected into animals anaesthetized in this way, oedema of the lungs is frequently produced, and this may kill the animal and certainly deprives the results obtained of any real value. In such experiments, therefore, I found it necessary to use urethane to such an amount that little or no other anaesthetic was necessary. Urethane, however, has its disadvantages : (1) urethane itself depresses the respiratory centre ; (2) although the initial blood-pressure under urethane is high, yet it seems to make the vasomotor system more susceptible to depressant substances such as β-i.

(*b*) 1. *The injection of large quantities of Ringer solution into the blood-stream.*

Experiments 50 and 52 show the effect of injecting several hundred c.c. of Ringer solution. In Experiment 50 quantities of 200, 100, and 100 c.c. were injected into the external jugular of one cat. During the time of absorption, viz. 1 hour 50 minutes, 25 and 35 per cent. of the β-i were absorbed from the intestine, the control cat, without fluid injection, absorbing 40 and 70 per cent. in the same period. When the time allowed for absorption is increased, as in Experiment 52, the time in this case being 2 hours 40 minutes, the difference is less marked, especially if, as usually happens, the control cat dies. In this experiment, quantities of 200, 100, and 100 c.c. were injected, and the amounts of β-i absorbed were 50 and 82 per cent. in the Ringer cat and 66 and 89 per cent. in the control cat.

(*b*) 2. *The frequent injection of small quantities of Ringer solution into the blood-stream.*

It seemed probable that, in view of the mechanism animals possess for ridding the circulatory system of excessive fluid, the injection of small quantities of fluid at frequent intervals would be more efficacious in suppressing the absorption of β-i than the injection of large volumes of fluid less frequently. For, in the former case, the circulatory system would probably, over the period of the experiment, have an average greater quantity of fluid circulating through it. Experiment 110 was carried out to test this point. Into the external jugular vein of one cat, quantities of 10 c.c. of Ringer solution were injected at intervals of fifteen minutes, 110 c.c. being injected altogether. 50 and 73 per cent. of

the β-i in the intestine were absorbed by the Ringer cat, and from corresponding intestinal loops of the control cat without Ringer solution 80 and 84 per cent. of the β-i disappeared.

In all the other seven experiments in which large quantities of Ringer solution were injected, there was marked depression of the β-i absorbed from the intestine.

Of greater importance still, is the fact that in all the experiments of this nature performed, viz. ten, eight of the control cats, i.e. those without the injection of fluid, died. In one case only did the Ringer-injected cat die, and this took place after thirty minutes' further β-i absorption than the control cat. In several cases the Ringer cats were allowed to live for some hours after the control cats had died, and had ultimately to be killed.

The result of these experiments is, therefore, quite definite.

1. The absorption of a toxic substance like β-i is depressed by the injection of large quantities of Ringer solution.

2. The resisting power of a Ringer-injected cat is greatly increased against the toxic action of β-i.

(*b*) 3. *The injection of small quantities of Ringer solution into the blood-stream.*

Other experiments similar to those already described were performed, but in these cases smaller quantities of Ringer solution were injected (75 c.c. or less over the whole period of the experiment). None of these will be quoted, as the difference between the β-i absorbed in the experimental and control cats was too small to be of any importance.

IX. *Some Miscellaneous Experiments.*

In the following experiments only one of each type of experiment was performed, and it is impossible therefore to attach importance to them as they stand. The only one that points to any positive result is the first, where the osmotic pressure of the blood was increased by injecting dextrose into the blood-stream.

The effect of increasing the osmotic pressure of the blood by injecting dextrose solution. Dextrose was used in this experiment for two reasons:

1. It is a suitable means of raising the osmotic pressure of the blood by a non-toxic physiological substance.

2. It is commonly believed that carbohydrate plays an important part in neutralizing toxic substances, so that the injection of dextrose was hoped to increase materially the resistance of the animal to β-i.

In Experiment 143 it will be seen that 25 c.c. of 100 per cent. solution of dextrose were injected into the external jugular vein immediately before the β-i solution was injected into the intestine. After one hour of absorption, 55 and 64 per cent. of the β-i had left the intestine of the dextrose cat, and 37 and

55 per cent. in the case of the control cat. The dextrose cat died one hour from the time of injection.

This experiment seems to indicate :

1. That increasing the osmotic pressure of the blood increased the rate of absorption from the intestine.

2. That the animal's resistance was not increased by the dextrose, since this cat died.

The effect of injecting magnesium sulphate into the circulation on the absorption of β-i from the intestine. $MgSO_4$ injected into the circulation is said to have some of the distinctive actions that are associated with this substance in the intestine. For instance, when injected subcutaneously, $MgSO_4$ increases intestinal peristalsis and may produce purgation, and it is therefore believed that some of the $MgSO_4$ injected subcutaneously is passed back from the blood into the gut. From this fact it might be expected that the injection of this substance into the blood would affect the rate of absorption from the intestine. Against such an expectation, however, may be placed the fact previously demonstrated that it takes a concentration of 2 per cent. $MgSO_4$ in the intestine to have any marked depressant action on the absorption of β-i.

In Experiment 115 two lots of 25 c.c. of Ringer solution containing 0·5 per cent. $MgSO_4$ were injected into the external jugular vein. After 65 minutes of absorption, 53 and 73 per cent. of the β-i had disappeared from the intestine of the $MgSO_4$ cat, and 53 and 50 per cent. from the corresponding loops of intestine of the control cat.

This experiment indicates that:

$MgSO_4$ in the circulation has not diminished the rate of absorption of β-i from the intestine.

The effect of injecting secretin into the blood-stream on the absorption of β-i from the intestine. On the supposition that absorption from the intestine is a kind of inverted secretion, it is possible that, just as the pancreas is stimulated to activity by secretin, so also this substance might have some action on absorption from the intestine. To test this point, Experiment 47 was performed.

Secretin was made in the ordinary way from the mucous membrane of the duodenum of a cat, and injected at different periods into the external jugular vein of another cat absorbing β-i from the intestine. The total amount of secretin injected in 3 hours was 100 c.c. The control cat died after 2¼ hours. The amounts of β-i absorbed from the secretin cat were 80 and 80 per cent., and from corresponding loops in the control cat 83 and 75 per cent.

The following points are indicated by this result :

1. Secretin has had no stimulating effect on the absorption of β-i (this cat absorbed for ¾ hour longer than the control).

2. The increased vitality of the secretin cat can probably be associated with the injection of fluid into its blood-stream.

X. *Evidence that β-i is not absorbed from the Large Intestine.*

In any consideration of the subject of alimentary toxaemias, it is clearly necessary that absorption of toxic substances from the large intestine must be investigated, for this situation is generally regarded as the abode of undesirable and toxic substances, and, if absorbed, they would probably produce pathological symptoms. The subject is, however, a large and complicated one, and the facts so far ascertained in this research can but form the basis of further work, although they were thought sufficiently interesting to mention in this paper.

In considering the part played by β-i in the large intestine, the following questions present themselves for solution :

1. Is β-i normally formed in the large intestine ?
2. If formed, is it absorbed into the blood-stream ?
3. If formed and not absorbed, does it have a local action on the colon or is it further changed to an innocuous substance ?

As regards Question 1, there is no doubt but that β-i can be formed in the intestine, for, in this position, Twort and I constantly found the bacillus which produced β-i from histidin. On the other hand, so far as I am aware, nobody has isolated β-i from the contents of the colon or even from its mucous membrane. I should think it unlikely that β-i is formed to any real extent in the large intestine of a normal animal, for its presence would stimulate the plain muscle, and bring about evacuation when only a small quantity was present.

As regards Question 2, this point has been investigated, and it will be seen that no evidence of the absorption of β-i from the large intestine has been obtained. It does not seem to me likely that such a substance would be absorbed in this part of the alimentary canal, for the following reasons : (*a*) There is but little evidence that anything except water is absorbed from the large intestine ; (*b*) It is obviously undesirable that toxic substances which accumulate in the large intestine should gain access to the blood and general circulation.

As regards Question 3, it will be seen that if β-i is formed in, or enters the large intestine from the small intestine, then bacteria can, and probably do, convert it into an innocuous substance.

In Experiment 71 we see that all the β-i injected into the large intestine was recovered after it had been allowed to remain there 1 hour 55 minutes. From a loop of small intestine placed immediately above the ileo-caecal valve, 40 per cent. of β-i was absorbed in this time. This experiment seemed to indicate that the large intestine was incapable of absorbing β-i. When the experiment was subsequently repeated, however, a different result was often obtained, namely, some of the β-i injected into the large intestine could not be recovered.

Experiment 132 is an example of this result. It will be seen that 50 and 50 per cent. of the β-i disappeared from loops of the small intestine, and 30 per cent. from the large intestine. In view of the contradiction of these experimental results, Experiment 144 *c* was performed, in which, before

injecting β-i into the large intestine, all the blood-vessels to this part were tied. In 2 hours 20 minutes, 33 per cent. of this β-i disappeared, and since it could not be absorbed it had obviously been destroyed by bacterial decomposition and the end products, at least so far as its action on plain muscle was concerned, were without physiological action.

The above facts indicate that the following statements concerning β-i and its relation to the large intestine may be true:

1. That any β-i formed in the large intestine or passed on here from the small intestine is not absorbed into the general circulation.

2. That, as a rule, any β-i formed in the large intestine is further destroyed by bacteria and made harmless, so that it does not even have its local stimulating action on this organ.

The complexity of the subject is emphasized by the different actions bacteria have on histidin as determined by the presence or absence of extraneous factors like oxygen, sugars, acids, and alkalis, and no doubt other unknown influences. It is an interesting problem to consider what part is played by β-i in the production of diarrhoea, and if it is responsible, why in such cases it has not been destroyed by other bacteria. For it is clear that β-i is not absorbed from the large intestine, and if formed, will only bring about evacuation of the colon and rectum.

As to what end product is formed when the bacterial flora of the large intestine makes β-i inactive, but little can be said. This is a change easily carried out, and only requires the removal of ammonia from the β-i molecule. That a considerable amount of ammonia is removed by bacteria in the colon is evident from the work of Folin, who goes so far as to claim that all the ammonia found in excess in the portal circulation really comes from the large intestine as the result of bacterial decomposition. This power possessed by bacteria in the large intestine of rendering toxic amines innocuous seems to me a factor of some importance, and worthy of further consideration, for it is almost certain that all active amines would undergo the same fate as that which obtains in the case of β-i.

Mutch (8) has recently found that the presence of the Bacillus aminophilus in the ileum is characteristic of patients suffering from chronic constipation, and associated with this, a low blood-pressure and toxaemic condition. It may be mentioned that the Bacillus aminophilus removes carbon dioxide from histidin and forms β-i, and Mutch assumes that the formation and absorption of β-i from the ileum is responsible for the low blood-pressure.

My results, here described, are in agreement with those of Mutch, in so far as it is the small intestine and not the large intestine which is the position for the absorption of β-i, if it should happen that this substance is responsible for the toxaemic symptoms of chronic constipation. It seems to me there are too many difficulties still unexplained by the meagre facts so far ascertained, to allow the claim that the production and absorption of β-i in the ileum cause a toxaemic condition in such people. To mention one obvious difficulty, an increased

formation of β-i would mean a hurried evacuation of the contents of the intestine rather than a condition of constipation, and I doubt whether any physical disability of the ileum and colon, short of complete obstruction, would satisfactorily meet this difficulty. In fact, it seems impossible with our present knowledge of bacterial production, bacterial destruction, and the absorption of β-i in the intestine, to be able to claim any causal relationship between chronic alimentary toxaemias and this substance.

Summary.

1. No evidence of the absorption of β-i from the large intestine was obtained.

2. Where β-i has disappeared after injection into the large intestine, it can be satisfactorily explained by bacterial decomposition.

3. This change of a toxic physiological substance into an inactive substance in the large intestine may extend to all amines. If this is so, it is an important second line of defence against the entry of such substances into the blood-stream from the large intestine.

XI. *The Resistance of Animals to β-i under Varying Conditions.*

In order to gain some insight into the processes underlying the resistance offered to β-i by animals under varying conditions the blood-pressure effects were specially studied.

The blood-pressure of the animals in these experiments depends on two factors :

1. The rate of absorption of β-i into the blood. The greater the amount absorbed, the greater the fall of blood-pressure.

2. The rate at which the animal can metabolize the absorbed β-i. If the β-i can be rendered innocuous immediately on absorption, naturally there is no effect on the blood-pressure.

The action of β-i on the blood-pressure is complex, and is not understood by physiologists. The outstanding points about its action are :

1. That it is an extremely strong stimulant of all plain muscle, and outside the body constricts all arteries.

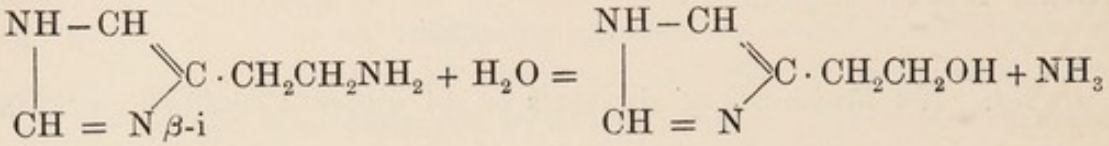
2. That in the intact animal it lowers the general blood-pressure and dilates the arterioles of the systemic system.

The difference between these two actions can only be reconciled by assuming that β-i either has a paralysing effect on the vasomotor centre or on some part of the sympathetic nervous system. That it does affect the central nervous system is evident from the anaesthetic action of the drug ; for it is nearly always possible to predict the relative amount of β-i absorbed during an experiment by the amount of anaesthetic required to keep the animal anaesthetized. An animal absorbing β-i rapidly does not require so much anaesthetic as one absorbing it slowly. It is further clear that β-i has a depressant effect on the respiratory centre, for whenever a cat has died under its influence, it has been observed that it is the respiratory centre that fails. It is true that the effect of injecting β-i

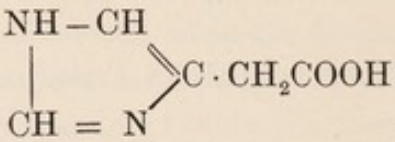
into the blood-stream is to cause a more rapid and a deeper respiration, but this effect is due to the constriction of the bronchiole muscles, which makes the breathing efforts more difficult because of the resistance met by the ingoing and outgoing respiratory air.

It seems probable, therefore, that the fall in general blood-pressure is due to the depressant action of β-i on the vasomotor centre or sympathetic system, which more than counterbalances its direct contracting action on the plain muscle of the arteries.

The next preliminary question is—what does the body do with β-i when it metabolizes it and renders it innocuous? Here again we are on unknown ground, but arguing by the analogy of other animes (9) one can say that its mode of disposal is probably as follows: The β-i is, probably, after absorption into the portal circulation, attacked by the liver cells, and the amino group taken away thus:

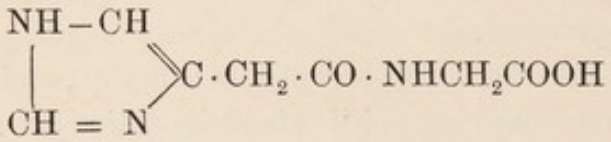


The alcohol group is probably then oxidized to—



β-imidazolylacetic acid.

Some of this acetic acid compound may then be combined with glycin and excreted as the aceturic compound thus:



These chemical changes are those undergone by parahydroxyphenylethylamine and indolethylamine, the bases of the amino-acids tyrosin and tryptophane respectively (10). So far as I know, the above chemical changes have not been shown to occur to β-i, at least in the perfused liver. One would expect there would be difficulty in proving the reaction with a substance of such marked physiological potency. I wish to point out that all that is necessary to render β-imidazolylethylamine innocuous is to remove the NH_2 group.

It was hoped, during the course of these experiments, to find there would be some simple relation between the rate of absorption of β-i and the blood-pressure. If, for instance, in animals placed under similar conditions, one animal could metabolize the β-i as it was absorbed, and so retain a high blood-pressure, while the second animal, with a poor metabolizing power, had a low blood-pressure, it might be expected that the rate of absorption would be a measure of the blood-pressure, so that an animal with a vigorous metabolism would absorb more β-i

than one with a poor metabolism. This condition cannot be said to hold from the experiments so far performed, although, I believe, when the blood-pressure is very low—say below 50 mm. Hg—the absorption from the intestine is small.

Normal Blood-pressure Changes during the Absorption of β-i.

In all experiments the blood-pressure is at its lowest about thirty minutes after the β-i has been injected. If the animal has but little resisting power to the absorbed β-i, then the blood-pressure remains stationary for some time, until a further fall of pressure occurs with death. If the animal has a more vigorous metabolism, a reaction follows and the blood-pressure gradually rises, sometimes even to the normal, but more frequently to a level lower than the normal.

The usual blood-pressure effects are to be seen in the following experiments:

Experiment 140 *a*.

β-i injected into intestine, 12.25.

Blood-pressure.

Before injection,		12 18	120 mm. Hg.
After	,,	12.29	103 ,,
,,	,,	12.35	86 ,,
,,	,,	12.45	80 ,,
,,	,,	12.50	62 ,,
,,	,,	12.55	52 ,,
,,	,,	1.3	56 ,,
,,	,,	1.17	60 ,,
,,	,,	1.38	60 ,,
,,	,,	1.55	78 ,,
,,	,,	2.14	76 ,,

Experiment 140 *b*.

β-i injected into intestine, 12.33.

Blood-pressure.

Before injection,		12.30	96 mm. Hg.
After	,,	12.36	79 ,,
,,	,,	12.43	65 ,,
,,	,,	12.51	55 ,,
,,	,,	1.2	58 ,,
,,	,,	1.16	60 ,,
,,	,,	1.30	60 ,,
,,	,,	1.55	70 ,,
,,	,,	2.10	74 ,,

This lowering of the blood-pressure and subsequent rise may be correlated with the rate of absorption of β-i, which is greater during the first hour, and afterwards becomes slower.

(*a*) *The blood-pressure effects in animals injected with Ringer solution.* It has previously been pointed out that injecting large quantities of Ringer solution into the blood-stream has two effects:

1. It delays the absorption of a toxic substance like β-i from the intestine.

2. It makes the cat less susceptible to the β-i absorbed, so that, whereas the control cat usually dies, the Ringer-injected cat lives.

The question is, how does the Ringer solution have this second action?

This question is difficult to answer, but it seems quite definite that the increased fluid does not prevent the β-i from lowering the blood-pressure, almost in as marked a manner as in the control animal. It does, however, prevent the final lowering of blood-pressure preceding death.

That the injection of fluid does not prevent the initial lowering of blood-pressure is seen in Experiment 110. In this experiment it will be seen that the blood-pressure is depressed from 110 mm. to 60 mm. in the Ringer-injected cat and from 120 mm. to 70 mm. in the control cat. It will be further observed that the blood-pressure of the Ringer cat gradually rises to 86 mm., while that of the control cat gradually sinks to 64 mm., at which time the fall is rapid and the cat

dies in five minutes. In other words, the injection of fluid does not prevent the initial large fall in blood-pressure, but gives to the animal the power of asserting itself once more, and the blood-pressure rises to some extent.

The question now arises, why does the Ringer solution prevent the fall of blood-pressure preceding death? I have already mentioned that I do not consider the fall of blood-pressure to be the primary cause of death. Death in these animals undoubtedly is the result of the failure of the respiratory centre. Of course the activity of the respiratory centre depends on the blood-pressure, but a Ringer-injected cat will have an efficient respiratory centre with a lower blood-pressure than a non-injected control cat will have with a higher blood-pressure. One must further suppose that the β-i depresses the vasomotor centre as well as the respiratory centre, for the asphyxial rise of blood-pressure, due to the increased hydrogen in concentration of the blood which accompanies respiratory failure, is absent.

The beneficial effect of injecting fluid into these cats depends, therefore, on some effects produced in the medulla oblongata which prevent the β-i from completely knocking out of action the respiratory and vasomotor centres. From this consideration, it appears possible that the original fall of blood-pressure, experienced to an equal extent both by the Ringer-injected and the control cats, may depend upon some interference of the vasomotor mechanism not situated in the medulla oblongata, and for this reason, that whereas injected fluid does not prevent the normal fall of blood-pressure produced by β-i, it does prevent the onset of that fall of pressure and failure of the respiratory mechanism accompanying death, the latter of which actions are obviously related to the medulla oblongata.

Two suggestions can be made as to the increased resistance conferred on animals by injecting Ringer solution:

1. A large part of the Ringer solution injected must be filtered off from the blood to the lymph spaces. This in itself may carry off a portion of the β-i to places where it is innocuous.

2. The increased body-fluids may increase the gaseous and other interchange between the blood, the lymph, and the cells of the body, including the cells of the nervous system. The cells would consequently get a greater opportunity of selecting nutriment, and therefore be in a position for resisting more completely the full toxic action of the β-i.

There does not appear to me to be any definite evidence to allow one to state that β-i can be rendered innocuous at a greater rate when the body fluids are increased by injecting saline solutions into the blood. On the other hand, the beneficial results of such treatment, as seen by the increased vitality of the animals, are unmistakable.

Summary. Injecting fluid into the blood-stream does not prevent the fall of blood-pressure caused by β-i as seen in the normal animal; but it does prevent the fall of blood-pressure preceding death. This latter important action probably depends on the increased vitality of the centres in the medulla.

(*b*) *Blood-pressure effects in animals deprived of blood.* Loss of blood makes an animal very susceptible to β-i absorbed from the intestine, and although less is absorbed than in the normal animal, as has been previously shown, the bled animal always dies as the result of the experiment, and often very rapidly. The animal seems to lose most of its power of rendering the β-i innocuous, the blood-pressure rapidly falls and the respiratory centre fails.

In Experiment 141, after withdrawing 15 c.c. of blood from the carotid artery, β-i absorbed from the intestine brought the blood-pressure down from 90 mm. to 40 mm., and the cat died within thirty minutes of injection. The blood-pressure of the control cat came down in the same time only from 160 mm. to 130 mm. It is further of interest to observe that the control cat absorbed 10 per cent. more β-i from the intestine than the bled cat, so that the latter cat was quickly killed by a smaller quantity of β-i than that which had but little effect on the control cat.

It can be said, therefore, that just as increasing the body fluids increases the resistance of the tissues, and more obviously of the medullary centres, to β-i, so also diminishing the body fluids by bleeding makes the same tissues extremely susceptible to β-i, so that the death of the animal quickly comes about.

I wish to point out again that the results here obtained cannot be applied completely to the more natural condition, from the point of view of this work, in which the body fluids are reduced by vomiting and diarrhoea, or when water is withheld from the diet.

An animal in this latter condition, with a large cellular and protein element of its blood, will undoubtedly be better able to render β-i innocuous than the bled animal, but I think it probable that, in both cases, the tissues of the body, and especially the respiratory centre, will have a reduced power of resisting that β-i which is not rendered innocuous.

I recognize it is difficult to dissociate the resistance of a tissue to a toxic substance and the power that tissue has of rendering a toxic substance innocuous ; for it seems evident that a substance only acts on a tissue in the inverse proportion to the power that the tissue has of rendering it innocuous by chemically transforming it. However, when a substance like β-i, which apparently kills by acting on a small part of the medulla oblongata, is being dealt with, I think it is permissible to discuss these two conditions as being, to some extent, independent. For the power the cells of the nervous system have of rendering a substance innocuous by causing a chemical transformation must be small compared with the liver and cellular elements of the blood. Consequently, I do not think there will be much difference in the resisting power of the medullary centres to β-i in the two types of animals deficient in blood, although the blood deficient in cellular elements will less efficiently make β-i absorbed from the intestine harmless than more normal blood.

Summary. In a bled animal, β-i absorbed from the intestine causes a more rapid fall in blood-pressure and the animal soon dies of respiratory failure.

(*c*) *Blood-pressure effects in fed animals.* It has been seen in previously described experiments that the absorption of water and food-stuffs depresses the rate of absorption of β-i from the intestine.

Considered from the point of view of blood-pressure, several interesting facts are revealed in the case of digesting animals. I do not here refer to the comparison of well-fed and starving animals, for experiments on this point have not been made. Incidentally, however, I may say that there is no doubt, from observation in well-fed and poorly conditioned animals, that the former can deal with large amounts of β-i and their blood-pressure be little depressed, while the latter die very readily and experience a rapid and marked fall of blood-pressure after absorbing small quantities of β-i. In comparative experiments such as comprise a large part of this work, this question regarding the state of nutrition has always been a difficulty, and can only be surmounted by keeping the animals for some days on a known diet before the experiment.

In the following experiments, diets of meat or milk or fat were given to cats at varying periods from one to six hours before being anaesthetized, and the effect on the blood-pressure produced by β-i absorbed from the intestine was observed.

1. *Meat.*

If the animal had eaten meat four hours or less before the injection of β-i into the intestine the results were too indefinite to be interpreted, for we have previously seen that the digestion and absorption of meat depresses slightly the absorption of β-i from the intestine. Consequently it is not surprising that the fall in blood-pressure in the meat-fed cat is not so pronounced in the majority of cases as in the control cat.

When the interval between eating the meat and injecting β-i is about six hours then the effect is more obvious, as can be seen in Experiment 139. Cat *B* was given 30 grm. of meat about seven hours before the β-i was injected into the intestine. Cat *A* was not given meat. It will be seen that the blood-pressure in *B* fell from 134 to 122 in the time that the blood-pressure in *A* (no meat) fell from 125 to 82. There was only 3 per cent. difference in the amount of β-i absorbed in these two cats, so that this could not explain the large difference in blood-pressure effect.

The increase in an animal's resistance to β-i produced by meat is clearly different from the increased resistance conferred on an animal by injecting a saline solution into the blood. In the former instance the blood-pressure is not allowed to be depressed to anything like the same degree as in the latter animal, although in both cases the blood-pressures recover and the animals survive. The entrance of the digestive products of the meat has a stimulating action on the cells of the body, giving them an increased metabolic power, so that the β-i is acted upon with greater readiness and more quickly made innocuous. In this change, the cells of the nervous system no doubt participate, and, as has been previously mentioned, if these cells can transform β-i more easily, then they will not be so susceptible to its toxic action.

Summary. When an animal has digested and absorbed a meal of meat the

depression of the blood-pressure caused by β-i absorbed from the intestine is considerably less marked than in a non-digesting animal. The animal has an increased capacity for chemically changing the β-i and making it harmless.

2. *Milk.*

The feeding of milk seems to have a similar effect to that of meat, but, in this case, the depression on the absorption of β-i from the intestine is more marked and the difference in the blood-pressure effect smaller, so that one can only say that the resistance of the tissues, although it is probably increased to some extent, is not so great as in the meat-fed animal.

3. *Fat.*

The results observed after feeding on fat are interesting. Of the experimental results quoted in the protocols, only one, Experiment 146, will be mentioned here, but it will be seen that they are all of the same nature. In Experiment 146 cat *B* ate 20 grm. of beef-fat 4¾ hours before the β-i was injected into the intestine. Cat *A* had no food. In cat *B* (fat) the blood-pressure came down quickly, after the β-i was injected, from 124 mm. to 98 mm., after which it rose steadily to 150 mm. In the control cat *A* the blood-pressure came down after injection from 160 mm. to 88 mm., and then steadily rose to an average of 115 mm. A similar subsequent rise of blood-pressure following a fall is seen in Experiment 95 on a fat-fed cat.

In a cat fed on fat the following blood-pressure effects are observed:

(1) The blood-pressure is only depressed to a small extent following the first absorption of β-i from the intestine.

(2) The blood-pressure afterwards rises to a point considerably higher than before the injection of β-i.

(3) After a certain period of time—often after the remarkably coincident time of one hour and forty minutes after the injection of β-i—the blood-pressure falls more or less suddenly, sometimes quite suddenly, and the animal dies.

A perusal of the amounts of β-i absorbed in these experiments compared to the results in the control animal will show that the fat-fed cats absorb less β-i than the control animals. The difference, however, is not sufficiently marked to explain the slight fall of blood-pressure in the fat-fed cats. For an explanation of this, it might appear at first sight that fat has a similar influence to the meat in previously described experiments, where it was thought that the meat had stimulated the tissues to have a greater metabolizing action on the absorbed β-i. I do not consider that this explanation will hold in the case of fat, for the reason that this would not explain why the cats should die suddenly after a certain length of time. Any explanation must certainly be able to cover this phenomenon, which, so far as my experiments have gone, seems fairly constant. I suggest that a reasonable explanation is offered along the following lines, although I have not yet been able to carry out the obvious experiment to prove it.

A substance can be absorbed from the intestine in two ways:

1. Via the blood-stream into the portal circulation.
2. Via the lacteals into the lymph and then into the blood.

Ordinary physiological teaching would lead one to expect that β-i would be absorbed only into the blood-stream and not into the lacteals. Now, in a fat-fed animal, the lacteals throughout the intestine, except probably for a few inches above the ileo-caecal valve, are full of fat, and if one looks at the mesentery when injecting β-i into a fat-fed animal one notices that the milkiness of the lacteals disappears in a minute or two. This probably means that the β-i solution is entering into the lymphatic circulation via the lacteals. What is the difference to the body between the β-i entering the blood via the lacteals and that which is directly absorbed into the blood? The difference is this: the β-i entering the portal blood has to pass through the liver, where a large part will probably be made innocuous, while the lymphatic absorbed β-i obtains access to all the tissues of the body without the possibility of being destroyed by the liver. In other words, portal-blood β-i may be innocuous, lymphatic β-i is probably toxic. Thus, if it should happen that the intestinal lymphatic system is full of fat, so that the lymph stream from the intestine is extremely slow, until the fat is removed from this position, the β-i entering the body from the intestine will have to pass into the portal circulation, and the liver can then deal with it and render it harmless. It would be expected that when the fat has been completely emptied out of the lymphatic channels into the blood-stream there would be a sudden flow of the lymph charged with β-i into the general circulation, and that this β-i would be extremely toxic. This suggestion would adequately explain why, in Experiment 146, the blood-pressure suddenly fell from 132 mm. to 50 mm., also in Experiment 96 from 130 mm. to 40 mm., in both cases the fall of pressure taking place 1 hour 40 minutes after the injection of β-i into the intestine. This phenomenon was also observed in all other cases, although the fall of pressure was not always instantaneous. It was at first thought to be due to the anaesthetic, for these fat-fed cats take an anaesthetic very badly, the smallest inhalation of ether or chloroform sending down the blood-pressure suddenly and killing them, unless great care is taken in the administration. Careful observation made it clear that the phenomenon was not related to the anaesthetic.

It is therefore concluded that the high blood-pressure met with in fat-fed cats, during the absorption of β-i from the intestine, is due to the fact that, owing to the temporary blocking up of the intestinal lymphatic system by fat, more of the absorbed β-i enters the body via the portal circulation, so that a large part of it is rendered innocuous by the liver before getting access to the central nervous system, the heart, and general circulation.

One fact the above suggestion does not adequately satisfy, viz. why the blood-pressure should go much higher when β-i is in the intestine than prior to its injection. At present I know no facts which explain this interesting phenomenon, and it seems useless to discuss it.

Summary. After a meal of fat, β-i absorbed from the intestine causes a slight fall in blood-pressure, followed by a rise to a point higher than that attained before any β-i is absorbed. This high pressure is retained for some

time—usually about $1\frac{3}{4}$ hours—and then falls more or less suddenly, and the animal dies of respiratory failure.[2]

XII. *Summary of the Experimental Results with Animals.*

The most important results obtained in this research from a practical point of view, that is, results which seem to me to have a practical bearing on the subject of diarrhoea and vomiting of children, are as follows:

First, as regards the absorption of toxins like β-i from the intestine:

1. There is a marked delay in the absorption of toxic substances, normally in the intestine, when animals are injected with large quantities of fluid.

2. That water in the intestine delays the absorption of toxic substances.

3. That during the digestion of food-stuffs generally, meat, milk, sugar, and fat, there is a delay in the absorption of toxic substances.

4. That the presence of bile in the intestine delays the absorption of toxic substances.

5. That magnesium sulphate, of a concentration of 2 per cent. or over, delays the absorption of toxic substances from the intestine. A less concentration of $MgSO_4$ has no effect.

6. That morphine, below the point of being a serious menace to the respiratory centre, has no effect on the absorption of toxic substances.

Secondly, as regards the resisting power of the animal towards absorbed toxic substances:

1. The resistance of an animal against toxic substances is very greatly increased by the injection of fluid into the circulation.

2. An animal with a diminished amount of fluid, and particularly after the loss of a small amount of blood, has little power of resistance against toxic substances.

3. An animal's resistance is greatly increased after it has absorbed from the intestine food, and more particularly meat.

4. During the digestion of fat, toxic substances absorbed from the intestine have a diminished action, at least for some hours.

XIII. *The Bearing of the above Results on the Aetiology of Diarrhoea and Vomiting.*

The research work I have described only allows me to discuss this subject from one point of view. I can pronounce no opinion on the important questions of differences of the bacteria, or superadded bacteria (microscopic or ultra-microscopic), or even the increase of chemical toxic substances in the intestine of diarrhoeic and vomiting children. At the same time my experimental

[2] Since this paper was written some months ago, I have satisfied myself that the rise of blood-pressure following the fall is not so constant as earlier experiments led me to believe. Further, experiments made on lymphatic absorption of β-i from the intestine indicate that too little is absorbed in this way to explain the foregoing phenomena in the above manner.

results seem to indicate that given only the toxic products present in the normal alimentary canal, together with a child reduced to such a condition as that produced by an ordinary attack of diarrhoea and vomiting from any dietetic indiscretion, these conditions alone can explain the serious results that may follow.

What is the condition set up in a child suffering from an acute attack of diarrhoea and vomiting? The primary cause of this condition I cannot discuss, beyond saying that a child's alimentary canal is capable of being deranged by many causes not necessarily bacterial in their nature. Such a child has then—

1. No food in its alimentary canal and no water.
2. A deficiency of body fluids to a greater or less degree.
3. A loss of bile, including more particularly bile salts.
4. No reserves of absorbed food-stuffs in its whole economy.

These are precisely the conditions demonstrated in animal experiments for—

1. Allowing toxic substances to be absorbed from the alimentary canal at a maximum rate.
2. Allowing the absorbed toxic substances to have their maximum effect.

It is thus clear that once the condition has been set up, the effects are cumulative, for the diarrhoea and vomiting first produced allow substances such as β-i to be absorbed, which by exerting their own action produce further diarrhoea and vomiting and fall of blood-pressure and loss of fluid. The child becomes more and more susceptible, and death ultimately ensues.

Of these results the most important seems to me to be the loss of body fluids. The one condition associated with diarrhoea and vomiting which has been regarded by clinicians as particularly prominent is the onset of this disease in an epidemic form when the temperature of the air arrives and remains at a certain high degree. This temperature has constantly been regarded as a causative factor in the production of bacterial products of a toxic nature and it is supposed that these products are responsible for the epidemics. All this seems likely. What I consider of greater importance, however, is that the high temperature causes a great diminution of body fluids by evaporation, so that when ordinary digestive derangement causing diarrhoea and vomiting is superadded, the further loss of fluid, and other conditions described above, place the child in the most susceptible state and deprive it of all power of resistance to poisonous influences.

The body temperature of an animal is the result of a balance between heat production and the loss of heat through radiation and conduction and loss by evaporation. The following figures obtained by Rubner show how the loss of heat through evaporation becomes more important when the temperature is raised:

External Temperature.	Total Heat produced. *Cal. per kg.*	Loss by Conduction and Radiation. *Cal.*	Loss by Evaporation. *Cal.*
7·6° C.	83·5	71·7	11·8
15° C.	63·0	49·0	14·0
20° C.	53·5	37·3	16·2

These figures represent loss of heat in a dog, and it is seen how greatly the loss of heat by evaporation increases, especially when considered relatively to the loss by conduction and radiation. In a child the difference would be more striking, for the loss by evaporation forms normally a greater percentage of the heat loss:

	Loss of Heat. Radiation and Conduction.	Loss of Heat. Evaporation.
Man	77·1 %	22·9 %
Dog	79·5 %	20·5 %

It is evident, then, that a child is capable of losing a large percentage of its body fluids by normal evaporation during hot weather, so that when further loss of water by vomiting and diarrhoea is added, it will be seen how easily a child may enter the danger zone.

Apart from the experiments described in this research, it is well known from other facts, although to my mind it has never been fully appreciated, how susceptible the body is to loss of fluid. Arguing from the condition of the blood in cholera, Hill makes the remark that a fasting mammal can use up the whole of its body fat and 50 per cent. of its protein before dying, while a thirsting one becomes moribund when it has lost little more than 10 per cent. of its body fluids.

As for the draining away from the body of bile, and more especially the loss of bile salts, the fact that these substances inhibit the absorption of toxic substances from the intestine is important. I am inclined to think their importance is still greater for reasons I have not yet fully grasped. For instance, I think that bile salts, besides stimulating the liver to excrete bile, are also an effective stimulant to the liver cells, and enable them more completely to metabolize and render toxic substances innocuous.

To sum up:

1. A child suffering from diarrhoea and vomiting owing to loss of fluid, loss of bile salts, with an empty intestine and in a starving condition, is in an ideal position for allowing toxic substances normally present in the alimentary canal and mucous membrane to be rapidly absorbed and have their full toxic action.

2. That the association of this disease with a high atmospheric temperature in an epidemic form is to be explained largely by the additional loss of fluid due to evaporation of water, whereby the child keeps down its body temperature.

XIV. *The bearing of the Experimental Results on the Treatment of the Disease.*

At this point I wish to discuss the lines along which, according to my experimental results, such treatment ought to be directed. It seems proper, also, to point out here that the experimental conditions which form the basis of this work are artificial, and that it is impossible to say whether the deductions made from the results can be applied altogether to the treatment of

diarrhoea and vomiting of children. The fact that they are on the whole in close agreement with a recognized treatment of this disease, a treatment which has been evolved by empirical methods, makes one hope that they can be extended to clinical uses, and that, if they fail to cure, it is not because they are inherently wrong, but because in the disease there are other factors to be accounted for besides those studied, namely, the toxic action of substances absorbed from the alimentary canal. It is possible that there are other factors in the disease which have not been met in the experimental work, such, for instance, as a bacterial invasion of the blood or a deficiency in the body of chemical substances necessary for the sustenance of life. If this is the case, then such additional factors can only be discovered by combined experimental and clinical observation. For if the methods evolved from laboratory work, such as is here described, are radically inefficient when applied to the specific clinical cases, one can only assume that the experimental conditions do not sufficiently resemble the pathological state met with in the disease.

(*a*) *The necessity of increasing the body fluids to normal.* The first and fundamental line of treatment, according to the experimental results, is to raise the body fluids up to and beyond the normal amounts. The injection of fluid is, of course, a recognized treatment already used clinically, and the animal experiments above described only confirm this view and possibly place it on a more scientific basis. I wish to point out, however, more explicitly the importance of this measure. There are two things to remember: (1) That an animal with deficient body fluids has lost most of its power of resisting any toxic action; (2) that by increasing the body fluids well above the normal, the resistance to toxic action is greatly increased. From these facts it will be seen that, if a child continues to vomit everything, including water, then it is better to inject a sterilized saline solution into the blood-stream directly rather than subcutaneously. By injecting fluid into the blood-stream one can raise the body fluids well above the normal, but in subcutaneous injection the tissues seem to refuse to imbibe the fluid when a certain maximum is attained. Intravenous injection into a young infant with empty vessels is generally difficult and often impossible, and in these cases one is reduced to subcutaneous injection of salt solution.

As soon as vomiting subsides, even if only partially, the child should be given large quantities of fluid by mouth. To be given a few sips of water occasionally is of little value, and I do not consider it is possible to give too much water. As much as two ounces of water every hour should be given while the child is in a moribund condition, and until vomiting completely ceases. Water given by mouth and absorbed into the circulation from the alimentary canal is undoubtedly better than water injected in any other way. For it has been previously pointed out, that when water is in the intestine, the absorption of β-i, and no doubt other toxic substances present in the intestine, is more markedly diminished than when the body fluids are increased by the injection of salt solution into the blood. There is also evidence that when water is absorbed

through the alimentary mucous membrane it liberates substances capable of stimulating the organs of the body, such for example as the kidney, while water injected intraveneously or subcutaneously has no such action (11). Thus, for instance, it is said that water taken by mouth is a better diuretic than an equal quantity of water injected into the blood-stream, because in the former case a specific diuretic substance is liberated by the water from the mucous membrane of the alimentary canal which stimulates the kidney to activity. If this is true of renal activity, it seems probable that stimulants from other organs may also be liberated at the same time. All the evidence therefore points to the advisability of giving water by mouth to these children, if this is possible.

In order to administer a saline solution subcutaneously to a child the best plan is to place the sterile solution in a thermos flask, and to give it continuously in this way. Quantities of a pint, 568 c.c., can be injected in this way in a few hours, and this should be repeated until the child can take water by mouth.

It may be said, however, that if large quantities of fluid are injected intravenously into a child there may be a danger of producing oedema of the lungs. On this point my experience with children is not sufficiently great to speak with authority, but from a large experience with animals I should say there is but little danger so long as the child is not suffering from bronchitis and broncho-pneumonia. It is certainly true that, if anything has caused irritation to the lungs of cats, such for instance as the inhalation of ether or chloroform, and especially the former, oedema of the lungs is produced even when only small quantities of Ringer solution are injected into the blood. This is an important practical point and cannot be emphasized too strongly. I should say that treating shock on the operating table, after or during the inhalation of ordinary anaesthetics, by injecting salt solutions intravenously into a patient, may be a most dangerous proceeding in consequence of the ease with which oedematous lungs are produced after anaesthetics, especially after ether.

Broncho-pneumonia often develops as a terminal event in diarrhoea and vomiting of children, and in such cases intravenous injection of salt solution is contra-indicated, and more especially so, for by this time vomiting has usually disappeared, and it is possible for the child to retain water in its stomach.

In recent years a good deal of publicity has become attached to the 'sea-water' treatment of diarrhoea and vomiting. In so far as, in this treatment, fluid is injected it must be of value, and I think it should be understood that it is the water that is the important element of this treatment. It is, of course, useful to have the fluid of a similar osmotic pressure and of somewhat similar constitution to the salt constituents of the tissues, but this is not an essential. It might be supposed that the magnesium element of the sea-water would be of importance, but my experimental results give no support to this view, and there is no doubt that normal salt solution or other physiological saline is quite as effective in the treatment of diarrhoea and vomiting of children as diluted sea-water.

(*b*) *Feeding.* The feeding of the child should start as soon as possible; but, at the same time, it is useless to feed the child with any food requiring digestion so long as there is a deficiency of fluid in the body. This is obvious, for until the body fluids are normal, there will be no secretion of the digestive juices. The parched mouth of such children indicates the suppression of saliva, and the gastric juice has been observed to be similarly depressed. On the other hand, a large secretion of gastric juice can often be obtained by injecting saline into the blood-stream of an animal. If the secretion of gastric juice is absent, then all the other digestive juices of the pancreas and alimentary canal are suppressed, and in addition the absence of hydrochloric acid allows bacterial decomposition to develop to its fullest extent, so that food taken by mouth decomposes and a worse condition of sapraemia is produced. The custom of giving children albumin water at an early and severe stage of the disease seems to me to be contra-indicated in consequence of the decomposition-changes the albumin is liable to undergo in the absence of a secretion of proteolytic juices and hydrochloric acid. Speaking theoretically, I should say that a solution of 10 per cent. dextrose made up with 0·5 per cent. HCl would seem the most reasonable food-stuff to give the child. If this is retained, one might then pass on to a solution of whey containing a large proportion of lactose and so on to milk, the latter being diluted. The importance of feeding the child at as early a stage as possible is necessary, in order to afford some resistance to the tissues of the child in view of the sequelae, such as broncho-pneumonia, which intervene so frequently. After a child has had no nourishment for some days, it can offer no resistance to such a disease as broncho-pneumonia, and the possibility of this development must always cause one to remember the importance of supplying a source of energy as soon as possible.

I am aware that the administration of carbohydrates is regarded as bad treatment, but given with dilute hydrochloric acid, I cannot see any possibility of it doing harm, and if absorbed in the intestine, as I think it would be, it can only do good.

(*c*) *The use of purgatives.* My experimental work has afforded but little insight into the value of purgatives in diarrhoea and vomiting of children. It was seen that magnesium sulphate very efficiently retarded the absorption of β-i from the intestine in concentration above 2 per cent., but below this figure it had no effect. If this substance is used in treatment, therefore, it must be in fairly large doses to be at all effective. Here, again, it is useless to give such a drug until the body fluids are up to or above normal, and there is no doubt but that, the more fluid there is in the body, the better will $MgSO_4$ exert its cathartic action and suppress the absorption of toxic substances into the blood-stream.

Another substance which ought to be effective is castor oil, for we have seen that fats have a protecting action on the organism. If the fats of the oil are absorbed, their large caloric value must be very useful to children in this condition. Still recommends castor oil in 5 minim doses, and states that if thus given it has a constipating effect. One must assume that the oil in

these cases is not hydrolysed by the lipase of the pancreatic juice, and if this is so, the question arises as to whether it is absorbed from the alimentary canal. Whether calomel is a good purgative to give, I am unable to say. I should think there is always some danger in giving a substance like this, because of the possibility of absorption, and especially so when the body fluids are below normal, as in diarrhoea and vomiting. Calomel, when absorbed, like any other ionized mercurial compound, may have bad results.

(*d*) *The danger of morphine.* Standard clinical text-books on diarrhoea and vomiting of children teach that opium and morphine are very useful drugs in the treatment of this condition, especially when the alimentary canal is in a state of hyper-irritability, and there can be but little doubt that for suppressing the diarrhoea and vomiting, and generally reducing the movements of the alimentary canal, small doses of tinctura camphorae co. or Dover's powder, or tinct. opii, or even a hypodermic injection of morphine (although this latter has a preliminary effect of increasing vomiting) are excellent drugs. It is further stated in the same text-books how dangerous morphine can be if not used carefully and correctly. I have been most impressed in the animal experimental results above described by this fact. A cat is not very susceptible to morphine, and in order to produce Cheyne-Stokes breathing I have sometimes injected as much as 10 c.c. of liq. morphinae hydrochlor. into the circulation without causing death. In the above-described experiments, however, 5 minims injected into the alimentary canal were sufficient to bring about collapse of the respiratory centre, and death, when the animal was also absorbing β-i. It seems to me that, in a moribund and collapsed child, a most important point in treatment must be to avoid depressing in any way the respiratory centre, so that, although morphine is useful for suppressing the symptoms of diarrhoea and vomiting, I feel it should not be administered in this condition from any other point of view. In the amounts likely to be given to the child, it was seen above that it can have no action in suppressing the absorption of toxic substances from the alimentary canal.

Finally, of all treatment in this condition, the one thing essential is to get the volume of body fluids of the child back to normal or above normal, for otherwise—

1. The absorption of toxins from the intestine goes on at a maximum rate.

2. The absorbed toxic substances have their full toxic action and cannot be made harmless by the tissues to any marked extent.

3. All the digestive juices are suppressed and food only decomposes with the further production of toxins.

Protocols.

Note on Experiments.

The experimental procedure is fully explained in the text. In order to understand the following results, it may be stated briefly that the active substance β-i was placed in loops of intestine of 25 cm. length and left there for varying periods of time. Then the loops were removed and the contents washed out carefully with hot water, and made up to a volume of 300 c.c. In order to estimate the amount of β-i which had disappeared during the time of absorption, the power of the washings to contract a guinea-pig's uterus was compared with that of the original β-i solution. If, for instance, it took 0·5 c.c. of the experimental solution to contract the uterus to the same height as that produced by 0·25 c.c. of the original β-i solution, then it can be seen that 50 per cent. of the β-i placed in the intestine must have disappeared during its sojourn there.

A reading such as 0·2 β-i = 0·4 A_1 = 0·6 A_2 means that 0·2 c.c. of the original β-i solution, 0·4 c.c. of the washings from the loop of intestine A_1, and 0·6 c.c. of the washings of loop A_2 all had a similar power of contracting the same uterus. Since these solutions were equal in strength at the beginning of the experiment, some of the β-i (50 per cent.) must have been absorbed from loop A_1, and more (66·6 per cent.) from A_2.

Experiment 71 (p. 171).

Loop (2) at caecal end.
Loop (1) taken 2 inches above loop (2).
Mesenteric blood-vessels of loop (1) tied.
5 c.c. of a 1 per cent. solution of β-i placed in each loop.

Time of absorption:

2 hours 5 minutes.

Result:

0·3 c.c. of β-i = 0·3 c.c. loop (1) = 1 c.c. loop (2).

Amount of β-i absorbed:

Loop (1) 0 per cent.
Loop (2) 70 per cent.

Experiment 73 (p. 172).

Two cats, A and B.
Blood-vessels of 1 foot of jejunum tied in each.
Two loops of each cat at caecal end injected with 5 c.c. of a 1 per cent. solution of β-i.

Time of absorption:

A, 2 hours.
B, 1 hour 45 minutes.

Result:

0·45 c.c. β-i = 0·85 c.c. A_1 = 1 c.c. A_2,
= 0·8 c.c. B_1 = 0·9 c.c. B_2.

Experiment 52 (p. 172).

Two cats, A and B.
Two loops in each cat injected.
Loops A_1 and B_1 at jejunal end of small intestine.
Loops A_2 and B_2 at caecal " " "
5 c.c. of 1 per cent. solution of β-i injected into each loop.

Time of absorption:

A, 2 hours 40 minutes.
B, 2 hours 35 minutes.

Result:

0·2 c.c. β-i = 0·4 c.c. A_1 = 0·6 c.c. B_1,
= 1·2 c.c. A_2 = 1·8 c.c. B_2.

Amount of β-i absorbed:

A_1 = 50 per cent., A_2 = 60 per cent.
B_1 = 84 per cent., B_2 = 90 per cent.

EXPERIMENT 134 (p. 172).

Two cats, A and B.
Two loops in each cat injected.
Two loops, A_2 and B_2 at caecal end.
A_1 and B_1 25 cm. above loops A_2 and B_2 respectively.
5 c.c. β-i 1 per cent. solution injected into each loop.

Time of absorption:

A, 1 hour 17 minutes.
B, 1 hour 18 minutes.

Result:

0·1 c.c. β-i solution = 0·2 c.c. A_1 = 0·22 c.c. A_2,
= 0·22 c.c. B_1 = 0·3 c.c. B_2.

Amount of β-i absorbed:

A_1 = 50 per cent., A_2 = 55 per cent.
B_1 = 55 per cent., B_2 = 66 per cent.

EXPERIMENT 123 (p. 173).

A. C. E.
Two cats.
A. No food.
B. Given 100 c.c. milk by mouth 4½ hours before β-i injected.
5 c.c. β-i 1 per cent. solution injected into loop of intestine at caecal end in each cat.

Time of absorption:

2 hours in each case.

Result:

0·4 c.c. β-i = 1·2 c.c. A = 0·8 c.c. B.

Amount of β-i absorbed:

A = 66 per cent.
B = 50 per cent.

EXPERIMENT 37 (p. 173).

Two cats.
A. No food.
B. Given milk by mouth 5½ hours before β-i injected.
5 c.c. β-i 1 per cent. solution injected into three loops of each cat.

Time of absorption.

A, 4 hours.
B, 3 hours 50 minutes.

Result:

0·5 c.c. A_1 = 1 c.c. A_2 = 1 c.c. A_3 = 0·2 c.c. B_1 = 0·2 c.c. B_2 = 0·6 c.c. B_3.

Amount of β-i absorbed:

Original solution not estimated, but it is evident that much more β-i has been absorbed from the intestine of the unfed cat than from that of the fed one.

EXPERIMENT 125 (p. 173).

Two cats, A and B.
A. No food.
B. 30 grm. meat eaten 1 hour 50 minutes before β-i injected.
5 c.c. β-i 1 per cent. solution injected into 1 loop of intestine in each cat.

Time of absorption:
$1\frac{1}{2}$ hours in each cat.
Result:
0·35 c.c. β-i = 0·65 c.c. A_1 = 0·55 c.c. B_1.
Amount of β-i absorbed:
A = 46 per cent.
B = 36 per cent.

EXPERIMENT 129 (p. 173).
Two cats, A and B.
A. No food.
B. 35 grm. meat $4\frac{1}{2}$ hours before β-i injected.
Loops A_2 and B_2 at caecal end of intestine.
Loops A_1 and B_1 25 cm. distance above loops A_2 and B_2 respectively.
5 c.c. β-i 1 per cent. solution injected into each loop.
Time of absorption:
$2\frac{1}{2}$ hours in each cat.
Result:
0·1 c.c. β-i = 0·35 c.c. A_1 = 0·5 c.c. A_2,
= 0·35 c.c. B_1 = 0·35 c.c. B_2.
Amount absorbed:
A_1 = 70 per cent., A_2 = 80 per cent. (control),
B_1 = 70 per cent., B_2 = 70 per cent. (meat).

EXPERIMENT 145 (p. 173).
Two cats.
A. Given 20 grm. fat by mouth 6 hours before β-i injected.
B. No food.
Two loops in each cat injected—
A_1 and B_1 at duodeno-jejunal flexure.
A_2 and B_2 25 cm. distance above ileo-caecal valve.
5 c.c. β-i 0·5 per cent. solution injected into each loop.
Time of absorption:
1 hour in each cat.
Result:
0·35 c.c. β-i = 0·55 c.c. A_1 = 0·5 c.c. A_2,
= 0·6 c.c. B_1 = 0·6 c.c. B_2.
Amount of β-i absorbed:
A_1 = 36 per cent., A_2 = 30 per cent. (fat fed).
B_1 = 42 per cent., B_2 = 42 per cent. (control).

EXPERIMENT 146 (p. 173).
Cat A, no food.
Cat B, 20 grm. fat $4\frac{1}{2}$ hours before injection.
Loops A_2 and B_2 at caecal end.
Loops A_1 and B_1 50 cm. above loops A_2 and B_2 respectively.
Lacteals of B_1 and B_2 full of fat, especially B_1.
5 c.c. β-i 0·5 per cent. solution injected into each loop.
Fat in lacteals of loops injected in cat B disappeared within 3 minutes of injection.
Time of absorption:
A, 1 hour 50 minutes.
B, 1 hour 50 minutes.
Result:
0·4 c.c. β-i = 1·5 c.c. A_1 = 1·8 c.c. A_2,
= 1·2 c.c. B_1 = 1·5 c.c. B_2.

Amount of β-i absorbed:

$A_1 = 73$ per cent., $A_2 = 78$ per cent.
$B_1 = 66$ per cent., $B_2 = 73$ per cent.

EXPERIMENT 144 *a* (p. 174).

One cat.
Two loops injected.
Loop (2) at caecal end of intestine.
Loop (1) 25 cm. distance above loop (2).
Loop (1) contained 3 c.c. water + 5 c.c. β-i 1 per cent. solution.
„ (2) „ 3 c.c. olive-oil emulsion + 5 c.c. β-i 1 per cent. solution.

The water and olive oil were injected into the two loops respectively ½ hour before the β-i solution.

Time of absorption:

1 hour 20 minutes.

Result:

0·12 c.c. β-i = 0·4 c.c. A_1 = 0·3 c.c. A_2.

Amount of β-i absorbed:

Loop $A_1 = 70$ per cent.
Loop $A_2 = 60$ per cent.

EXPERIMENT 144 *b* (p. 174).

Chloroform.
One cat—two loops injected.
Loop (2) at caecal end of intestine.
Loop (1) 25 cm. distant above loop (2).
Loop (1) contained 3 c.c. water and 5 c.c. β-i 1 per cent. solution.
Loop (2) „ olive oil emulsion and 5 c.c. β-i 1 per cent solution.

The water and olive oil were injected into the two loops respectively 3 minutes before β-i solution injected.

Time of absorption:

1 hour 45 minutes.

Result:

0·12 c.c. β-i = 0·6 c.c. B_1 = 0·3 c.c. B_2.

Amount of β-i absorbed:

$B_1 = 82$ per cent. (control).
$B_2 = 60$ per cent. (olive oil).

EXPERIMENT 130 (p. 174).

Cat A, nothing.

Cat B, 25 c.c. 10 per cent. dextrose into small intestine (duodenum tied to prevent regurgitation) ¼ hour before β-i injected.

Time of absorption:

A, 1 hour 30 minutes.
B, 1 hour 30 minutes.

Result:

0·175 c.c. β-i = 0·5 c.c. A_1 = 0·7 c.c. A_2,
= 0·3 c.c. B_1 = 0·4 c.c. B_2.

Amount absorbed:

$A_1 = 65$ per cent., $A_2 = 75$ per cent. (control).
$B_1 = 42$ per cent., $B_2 = 56$ per cent. (dextrose).

EXPERIMENT 57 (p. 174).

Two cats, A and B.
Two loops in each injected.
Loops A_1 and B_1 at jejunal end of intestine.
Loops A_2 and B_2 at caecal „ „

Cat A, 40 minutes before β-i solution injected into loops, 60 c.c. of Ringer solution were injected into intestine at duodeno-jejunal flexure.

Cat B, no Ringer injection.

5 c.c. β-i 1 per cent. solution injected into loops.

Time of absorption:

A died after 1 hour 40 minutes.

B killed after 1 hour 35 minutes (respiration nearly knocked out).

Result:

0·1 c.c. β-i = 0·15 c.c. A_1 = 0·15 c.c. A_2,

0·2 c.c. B_1 = 0·9 c.c. B_2.

Amount of β-i absorbed:

A_1 = 33 per cent., A_2 = 33 per cent. (Ringer in intestine).

B_1 = 50 per cent., B_2 = 89 per cent. (no Ringer).

EXPERIMENT 58 (p. 174).

Two cats, A and B.

Two loops in each injected.

Loops A_1 and B_1 at jejunal end of small intestine.

Loops A_2 and B_2 at caecal „ „ „

Cat A, 25 c.c. Ringer solution injected into intestine between loops A_1 and A_2 5 minutes before β-i injection.

Cat B, no Ringer.

5 c.c. β-i 1 per cent. solution injected into all loops.

Time of absorption:

A killed after 2 hours 40 minutes.

B died after 2 hours 30 minutes.

Result:

0·3 c.c. A_1 = 0·6 c.c. A_2,

0·35 c.c. B_1 = 1·4 c.c. B_2. Original solution not estimated.

Amount of β-i absorbed:

It is clear that more β-i has been absorbed from B_1 than A_1 and more from B_2 than A_2.

EXPERIMENT 105 (p. 175).

Three loops of cat's intestine injected.

Loop (3) at caecal end.

Loop (2) 25 cm. distance above loop (3).

Loop (1) „ „ „ (2).

Loop (1) 1 c.c. cat's own bile + 5 c.c. β-i 1 per cent. solution.

Loop (2) no bile 5 c.c. β-i „ „ „

Loop (3) 1 c.c. cat's own bile + 5 c.c. β-i „ „ „

Time of absorption:

2 hours 30 minutes.

Result:

1 c.c. β-i = 2 c.c. (1) = 3 c.c. (2) = 1·5 c.c. (3).

Amount of β-i absorbed:

Loop (1) 50 per cent., with bile.

Loop (2) 66 per cent., no bile.

Loop (3) 34 per cent, with bile.

EXPERIMENT 106 (p. 175).

One cat.

Two loops injected.

Loop (1) and loop (2) at caecal end, ½ inch between each.

Loop (1) contained ½ c.c. bile + 5 c.c. β-i 1 per cent. solution.

Loop (2) contained 5 c.c. β-i 1 per cent. only.

Time of absorption:
2 hours 40 minutes.
Result:
0·08 c.c. β-i = 0·2 c.c. A_1 = 0·8 c.c. A_2.
Amount of β-i absorbed:
Loop (1) 60 per cent., with bile.
Loop (2) 90 per cent., no bile.

EXPERIMENT 108 *a* (p. 175). Bile duct tied.
One cat.
Loop (1) at jejunal end of small intestine.
Loop (2) ½ inch below loop (1).
Loop (1) contained 5 c.c. β-i 1 per cent. solution.
Loop (2) „ „ „ „ „ + ¼ c.c. bile-salt solution.
Bile-salt solution contained 6 per cent. sodium glycocholate,
2 per cent. sodium taurocholate.
Time of absorption:
2 hours 30 minutes.
Result:
0·6 c.c. loop (1) = 0·5 c.c. loop (2).
Amount of β-i absorbed:
β-i solution was not estimated, and therefore it is not possible to say how much was absorbed.
It is clear, however, that more was absorbed from loop (1), which contained no bile salts.

EXPERIMENT 108 *b* (p. 175). Bile duct tied.
One cat.
Loop (2) at caecal end of intestine.
Loop (1) ½ inch above loop (2).
Loop (1) contained 5 c.c. β-i 1 per cent. solution.
Loop (2) „ „ „ „ „ + ¼ c.c. bile-salt solution.
Bile-salt solution contained 6 per cent. sodium glycholate,
2 per cent. sodium taurocholate.
Time of absorption:
2 hours 30 minutes.
Result:
1·7 c.c. loop (1) = 0·7 c.c. loop (2).

EXPERIMENT 137 (p. 176).
One cat.
Two loops injected. Loop (2) at caecal end of intestine.
Loop (1) 25 cm. distance above loop (2).
Loop (1) contained 2 c.c. water + 5 c.c. 0·5 per cent. β-i solution.
Loop (2) contained 2 c.c. 20 per cent. $MgSO_4$ + 5 cc. β-i 0·5 per cent. solution.
Time of absorption:
2 hours.
Result:
0·35 c.c. β-i = 0·8 c.c. loop (1) = 0·5 c.c. loop (2).
Amount absorbed:
Loop (1) 56 per cent.
Loop (2) 30 per cent.

EXPERIMENT 138 (p. 177).
Two cats, A and B.
Two loops of each injected. A_2 and B_2 at caecal end of intestine; A_1 and B_1 25 cm. distance above A_2 and B_2.

A_1 contained 5 c.c. β-i 0·5 per cent. solution + 2 c.c. 20 per cent. $MgSO_4$.
A_2 " " " " " + 2 c.c. water.
B_1 " " " " " + 2 c.c. water.
B_2 " " " " " + 2 " "

The $MgSO_4$ solution and the water were injected into the loops about 3 minutes before the β-i solution.

Time of absorption:

A, 2 hours.
B, 2 hours.

Result:

0·4 c.c. β-i = 0·6 c.c. A_1 = 1·7 c.c. A_2,
= 1·2 c.c. B_1 = 1·3 c.c. B_2.

Amount absorbed:

A_1 = 33 per cent., A_2 = 76 per cent.
B_1 = 66 per cent., B_2 = 69 per cent.

EXPERIMENT 136 (p. 177).

Two cats, A and B.
Loops A_1 and B_1 at jejunal end of small intestine.
Loops A_2 and B_2 at caecal " " "
Loop A_1 contained 1 c.c. water + 5 c.c. β-i 1 per cent. solution.
Loop A_2 " " + " " "
Loop B_1 " " + " " "
Loop B_2 contained 1 c.c. $MgSO_4$ 5 per cent. solution + 5 c.c. β-i (i.e. 0·83 per cent. $MgSO_4$).

Time of absorption:

A, 1 hour 48 minutes.
B, 1 hour 22 minutes.

Result:

0·25 c.c. β-i = 0·3 c.c. A_1 = 0·5 c.c. A_2,
= 0·5 c.c. B_1 = 0·6 c.c. B_2.

Amount absorbed:

A_1 = 17 per cent.
A_2 = 50 per cent.
B_1 = 50 per cent.
B_2 = 58 per cent. $MgSO_4$ loop 0·83 per cent.

EXPERIMENT 134 (p. 177).

Two cats, A and B. β-i injected into two loops of each.
A_2 and B_2 at caecal end of small intestine.
A_1 and B_1 25 cm. distance above A_2 and B_2.

Into each loop, A_1, A_2, B_1, and B_2, 5 c.c. β-i 1 per cent. solution were injected.

Between loops A_1 and A_2, 5 minims of liq. morph. hydrochlor. were also injected.

Into B, no morphia.

Time of absorption:

A, 1 hour 18 minutes (died), morphine.
B, 1 hour 18 minutes (killed), no morphine.

Result:

0·1 c.c. β-i = 0·2 c.c. A_1 = 0·22 c.c. A_2,
= 0·22 c.c. B_1 = 0·3 c.c. B_2.

Amount of β-i absorbed by each loop.

A_1 = 50 per cent., A_2 = 55 per cent., morphine.
B_1 = 55 per cent., B_2 = 66 per cent., no morphine.

EXPERIMENT 133 *b* (p. 178).

One cat.

Two loops were injected with 5 c.c. β-i 1 per cent. solution.

Loop (2) at caecal end.

Loop (1) 25 cm. distance above loop (2).

1 c.c. of liq. morphinae hydrochlor. was injected into the intestine above loop (1).

Time of absorption:

1 hour 10 minutes.

During the last 40 minutes, the respiratory centre worked badly, after which the cat died of respiratory failure.

Result:

0·375 c.c. β-i = 0·6 c.c. loop (1) = 0·625 c.c. loop (2).

Amount of β-i absorbed by the loops of intestine:

Loop (1) 37·5 per cent.

Loop (2) 40·0 per cent.

EXPERIMENT 133 *a* (p. 179).

Two loops at caecal end injected with β-i 1 per cent. solution.

Loop (2) at caecal end.

Loop (1) 25 cm. distance above loop (2).

Loop (1) contained 5 c.c. β-i 1 per cent. solution + 1 c.c. of water.

Loop (2) „ „ „ „ + 1 c.c. 5 per cent. Na_2CO_3.

Time of absorption:

2 hours 30 minutes.

Result:

0·375 c.c. β-i = 1 c.c. loop (1) = 1·2 c.c. loop (2).

Amount absorbed:

Loop (1) 62·5 per cent.

Loop (2) 69·0 per cent.

EXPERIMENT 120 (p. 178).

Alkali at jejunal end of small intestine.

Two loops injected with 5 c.c. β-i 0·5 per cent. solution.

Loop (1) at duodeno-jejunal flexure.

Loop (2) immediately below loop (1).

Loop (1) contained 1 c.c. of 5 per cent. Na_2CO_3 in addition to the β-i solution.

Time of absorption:

2 hours 34 minutes.

Result:

0·3 c.c. β-i = 1·7 c.c. loop (1) = 0·6 c.c. loop (2).

Amount absorbed:

Loop (1) 82 per cent.

Loop (2) 50 per cent.

EXPERIMENT 140 (p. 180).

Two cats, A and B. A much larger than B.

Two loops of intestine injected with 5 c.c. β-i 1 per cent. solution.

A_2 and B_2 loops at caecal end of intestine.

A_1 and B_1 25 cm. above A_2 and B_2.

From A, 23 c.c. blood withdrawn from carotid 4 minutes before β-i injected.

B, 17 „ „ „ „ 19 „ after „ „

Time of absorption:

A, 2 hours.

B, 2 hours.

Result:

0·2 c.c. β-i = 0·4 c.c. A_1 = 0·6 c.c. A_2,

= 0·8 c.c. B_1 = 0·1 c.c. B_2.

Amount of β-i absorbed:

$A_1 = 50$ per cent., $A_2 = 66$ per cent., blood withdrawn.

$B_1 = 75$ per cent., $B_2 = 80$ per cent., normal.

A did not die in this case, but whereas the fall in blood-pressure in B was only 40 mm. Hg, in A it fell 68 mm. Hg.

In other words, A was much more affected by the smaller amount of β-i absorbed.

EXPERIMENT 141 (p. 180).

Two loops of each cat injected with 5 c.c. β-i 1 per cent. solution.

Loops A_2 and B_2 were at caecal end of small intestine.

Loops A_1 and B_1 were 25 c.m. distance above A_2 and B_2.

From A, 15 c.c. blood were withdrawn 3 minutes before β-i was injected.

B, normal.

Time of absorption:

A, 30 minutes, died.

B, 30 minutes, lived.

Result:

0·2 c.c. β-i = 0·25 c.c. A_1 = 0·3 c.c. A_2,
= 0·3 c.c. B_1 = 0·325 c.c. B_2.

Amount of β-i absorbed:

$A_1 = 20$ per cent., $A_2 = 33$ per cent. (bled cat), died.

$B_1 = 33$ per cent., $B_2 = 39$ per cent. (normal cat).

In A, the blood-pressure fell rapidly from 90 to 48 mm. Hg.

In B, „ „ „ only from 160 to 138 mm. Hg.

EXPERIMENT 56 (p. 181).

Cat B, bled 30 c.c. before injecting two loops of intestine with 5 c.c. β-i 1 per cent. solution each.

Injected 2.25 p.m.
Died 2.40 p.m. } 15 minutes.

Cat C, bled 20 c.c. before injecting β-i into intestine.

Injected with β-i at 5.40 p.m.
Died 5.50 p.m. } 10 minutes.

Cat A, control, i.e. not bled.

Injected 2.20 p.m.

Died 5.45 p.m., i.e., not till 3 hours 15 minutes had elapsed.

EXPERIMENT 50 (p. 182).

Two loops of each cat injected with 5 c.c. β-i 1 per cent. solution.

Loops A_2 and B_2 at caecal end of intestine.

Loops A_1 and B_1 at jejunal end „

Into A 200 c.c. Ringer were injected into external jugular vein at 3.50 p.m.
100 „ „ „ „ „ „ 4.40 p.m.
100 „ „ „ „ „ „ 5.50 p.m.

Time of absorption:

A injected with β-i solution, 4.30 p.m. B injected with β-i, 4.25 p.m.
A killed 6.20 p.m. B killed 6.20 p.m.

Result:

0·15 c.c. β-i = 0·2 c.c. A_1 = 0·23 c.c. A_2,
= 0·25 c.c. B_1 = 0·5 c.c. B_2.

Amount of β-i absorbed:

$A_1 = 25$ per cent., $A_2 = 35$ per cent., Ringer.

$B_1 = 40$ per cent., $B_2 = 70$ per cent., without Ringer.

EXPERIMENT 52 (p. 182).

Two loops of intestine in each cat injected with 5 c.c. β-i 1 per cent. solution.

Loops A_1 and B_1 at jejunal end of intestine.

Loops A_2 and B_2 at caecal „ „

Ringer injected into external jugular vein of A 200 c.c. at 11.10 a.m.
100 c.c. at 11.45 a.m.
100 c.c. at 12.45 p.m.
100 c.c. at 2.10 p.m.

Time of absorption:

A injected with β-i, 11.40 a.m. B injected with β-i, 11.35 a.m.
A killed 2.20 p.m. B died 2.10 a.m.

Result:

0·2 c.c. of β-i = 0·4 c.c. A_1 = 1·2 c.c. A_2,
= 0·6 c.c. B_1 = 1·8 c.c. B_2.

Amount of β-i absorbed:

A_1 = 50 per cent., A_2 = 82 per cent., with Ringer.
B_1 = 66 per cent., B_2 = 89 per cent., no Ringer.

EXPERIMENT 110 (p. 182).

Two loops of intestine in each cat.
All the loops at caecal end of intestine.

Into A, 10 c.c. of Ringer solution were injected at intervals of about 15 minutes from 2.20 to 5 p.m.

B no Ringer.

Time of absorption:

A injected with β-i, 2.25 p.m. B injected, 2.40 p.m.
A killed 5.2 p.m. B died 5.2 p.m.

Result:

0·4 c.c. β-i = 0·8 c.c. A_1 = 1·5 c.c. A_2,
= 2·0 c.c. B_1 = 2·5 c.c. B_2.

Amount of β-i absorbed:

A_1 = 50 per cent., A_2 = 73 per cent., Ringer.
B_1 = 80 per cent., B_2 = 84 per cent., no Ringer.

EXPERIMENT 143 (p. 183).

5 c.c. β-i 1 per cent. solution were injected into two loops in each cat, A and B.
Loops A_2 and B_2 at caecal end of intestine.
Loops A_1 and B_1 25 cm. distance above A_2 and B_2.

In B, at 11.45 p.m., 25 c.c. of 100 per cent. solution of dextrose were injected into the external jugular vein.

A no dextrose.

A, β-i injected 11.50 p.m. B, β-i injected 11.45 p.m.
A killed 12.54 p.m. B died 12.47 p.m.

Result:

0·25 c.c. β-i = 0·4 c.c. A_1 = 0·45 c.c. A_2,
= 0·45 c.c. B_1 = 0·7 c.c. B_2.

Amount of β-i absorbed:

A_2 = 37 per cent., A_2 = 55 per cent. (control).
B_1 = 55 per cent., B_2 = 64 per cent. (dextrose).

EXPERIMENT 115 (p. 184).

Two loops of each cat (A and B) injected with 5 c.c. β-i 1 per cent. solution.
Loops A_2 and B_2 at caecal end.
Loops A_1 and B_1 25 cm. distance above loops A_2 and B_2 respectively.

Ringer solution containing 0·5 per cent. $MgSO_4$ was injected into the external jugular vein of B as follows:

25 c.c. of solution at 3.2 p.m.
25 „ „ „ 3.32 p.m.

A, β-i injected 2.40 p.m. B, β-i injected 3.10 p.m.
A died 3.45 p.m. B killed 4.15 p.m.

Result:

0·35 c.c. β-i = 0·75 c.c. A_1 = 0·7 c.c. A_2 (control),
= 0·75 c.c. B_1 = 1·3 c.c. B_2 ($MgSO_4$).

Amount of β-i *absorbed:*

$A_1 = 53$ per cent., $A_2 = 50$ per cent. (control).
$B_1 = 53$ per cent., $B_1 = 73$ per cent. ($MgSO_4$).

EXPERIMENT 47 (p. 184).

5 c.c. of β-i 1 per cent. solution, were injected into two loops of each cat (A and B).

Loops A_1 and B_1 were at the jejunal end of small intestine.
Loops A_2 and B_2 „ „ caecal „ „ „

Secretin was made in the ordinary way from the mucous membrane of the duodenum of another cat and injected into A as follows:

30 c.c. of secretin into A at 2.25 p.m.
15 „ „ „ 2.57 p.m.
15 „ „ „ 3.30 p.m.
15 „ „ „ 4.0 p.m.
15 „ „ „ 4.45 p.m.
15 „ „ „ 5.15 p.m.

A, β-i injected 2.55 p.m. B, β-i injected 2.45 p.m.
A killed 5.55 p.m. B died 5.0 p.m.

Result:

0·2 c.c. β-i = 1 c.c. A_1 = 1 c.c. A_2,
= 1·2 c.c. B_1 = 0·8 c.c. B_2.

Amount of β-i *absorbed:*

$A_1 = 80$ per cent., $A_2 = 80$ per cent., secretin.
$B_1 = 83$ per cent., $B_2 = 75$ per cent., no secretin.

EXPERIMENT 71 (p. 185).

5 c.c. β-i 1 per cent. solution injected into two loops.
Loop (1) at caecal end of small intestine.
Loop (2) including all large intestine.
β-i injected 3.40 p.m.
Cat died 5.35 p.m.

Result:

0·3 c.c. β-i = 0·5 c.c. B_1 = 0·3 c.c. B_2.

Amount of β-i *absorbed:*

$B_1 = 40$ per cent.
B_2 = none.

EXPERIMENT 132 (p. 185).

Three loops of intestine injected with 5 c.c. β-i 1 per cent. solution.
Loop (1) 25 cm. distance above loop (2).
Loop (2) just above ileo-caecal valve.
Loop (3) large intestine.
β-i injected 2.45 p.m.
Cat died 4.50 p.m.

Result:

0·15 c.c. β-i = 0·3 c.c. (1) = 0·3 c.c. (2) = 0·22 c.c. (3).

Amount of β-i *absorbed:*

Loop (1) = 50 per cent.
Loop (2) = 50 per cent.
Loop (3) = 30 per cent., large intestine.

EXPERIMENT 144*c* (p. 185).

5 c.c. of a 1 per cent. β-i solution were injected into a loop of the large intestine, the mesentery of which was tied to prevent absorption.

Time of absorption: 2 hours 20 minutes.
Result: 0·4 c.c. β-i = 0·6 c.c. of loop contents.
Amount disappeared: 33 per cent.

EXPERIMENT 110 (p. 189).

Two cats.

A, Ringer injected in 10 c.c. at intervals of 10 minutes. 110 c.c. injected altogether.

Blood-pressure A.
Before injecting β-i.
2.21 p.m. 110 mm. Hg.
After injection.
2.27 p.m. 64 mm. Hg.
2.43 „ 60 „
3.0 „ 64 „
3.20 „ 58 „
3.50 „ 62 „
4.10 „ 68 „
4.30 „ 67 „
4.55 „ 86 „
and up to 100 mm.

B, no Ringer injected.

Blood-pressure B.
Before injecting β-i.
2.25 p.m. 120 mm. Hg.
After injection.
2.45 p.m. 70 mm. Hg.
3.0 „ 76 „
3.20 „ 84 „
3.50 „ 78 „
4.10 „ 74 „
4.30 „ 75 „
4.55 „ 64 „
and lower till death at 5 p.m.

Amount of β-i *absorbed:*
A = 61 per cent.
B = 82 per cent.

EXPERIMENT 141 (p. 191).

A, 15 c.c. blood withdrawn from carotid artery, 12.39 p.m.
β-i injected 12.42 p.m.
Blood-pressure.
Before injection.
12.40 p.m. 90 mm. Hg.
After injection.
12.50 p.m. 66 mm. Hg.
12.55 „ 48 „
down to 40 „
and died 1.10 p.m.

B, normal.

β-i injected 12.48 p.m.
Blood-pressure.
Before injection.
12.32 p.m. 160 mm. Hg.
After injection.
12.51 p.m. 138 mm. Hg.
1.0 „ 130 „
and remained so.
killed 1.16 p.m.

In the bled cat the blood-pressure came straight down to 48 mm., the control cat being very little affected. Yet the control cat absorbed 10 per cent. more β-i than the bled cat in the same period.

EXPERIMENT 139 (p. 192).

A, no food.
β-i injected 2.50 p.m.
Blood-pressure.
Before injection.
2.47 p.m. 125 mm. Hg.
After injection.
2.59 p.m. 106 mm. Hg.
3.10 „ 96 „
3.20 „ 82 „
3.40 „ 90 „
4.0 „ 96 „
4.25 „ 102 „
4.45 „ 124 „

B, given 30 grm. of meat at 8.0 a.m.
β-i injected 2.53 p.m.
Blood-pressure.
Before injection.
2.52 p.m. 134 mm. Hg.
After injection.
3.0 p.m. 115 mm. Hg.
3.10 „ 122 „
3.20 „ 122 „
3.35 „ 123 „
3.47 „ 120 „
4.12 „ 125 „
4.45 „ 150 „
4.53 „ 150 „

Amount of β-i absorbed:
$A_1 = 50$ per cent., $B_1 = 50$ per cent.
$A_2 = 73$ per cent., $B_2 = 66$ per cent.

EXPERIMENT 146 (p. 193).

A, no food.
β-i injected 3.45 p.m.
Killed 5.34 p.m.
Blood-pressure.
Before injection of β-i.
3.40 p.m. 160 mm. Hg.
After injection.
3.53 p.m. 128 mm. Hg.
3.57 „ 94 „
4.7 „ 88 „
4.20 „ 104 „
4.35 „ 115 „
4.50 „ 110 „
5.0 „ 112 „
5.20 „ 119 „
5.30 „ 128 „

B, 20 grm. of fat at 11 a.m.
β-i injected 3.48 p.m.
Killed 5.37 p.m.
Blood-pressure.
Before injection of β-i.
3.40 p.m. 124 mm. Hg.
After injection.
3.53 p.m. 98 mm. Hg.
3.57 „ 112 „
4.7 „ 140 „
4.20 „ 146 „
4.35 „ 144 „
4.50 „ 150 „
5.0 „ 132 „
5.20 „ suddenly down to 50.

Amount of β-i absorbed:
$A_1 = 73$ per cent., $A_2 = 78$ per cent., no fat.
$B_1 = 66$ per cent., $B_2 = 73$ per cent., fat.

EXPERIMENT 95 (p. 193).

A, fat eaten 11.15 a.m.
Injected with β-i at 2 p.m.
Blood-pressure.
Before injection.
1.55 p.m. 90 mm. Hg.
After injection.
2.7 p.m. 110 mm. Hg.
3.10 „ 134 „
3.30 „ 140 „
3.55 „ 116 „

B, no food.
Injected with β-i at 2.30 p.m.
Blood-pressure.
Before injection.
2.25 p.m. 135 mm. Hg.
After injection.
3.7 p.m. 122 mm. Hg.
3.30 „ 100 „
3.35 „ 78 „
4.0 „ 90 „
4.15 „ 84 „
4.30 „ 82 „

Amount of β-i absorbed:
$A_1 = 50$ per cent., $A_2 = 33$ per cent.
$B_1 = 50$ per cent., $B_2 = 50$ per cent.

EXPERIMENT 145 (p. 193).

A, given 20 grm. of fat at 11 a.m.
β-i injected 4.47 p.m.
Blood-pressure.
Before injection.
4.36 p.m. 110 mm. Hg.
4.45 „ 150 „
After injection.
4.57 p.m. 150 mm. Hg.
5.30 „ 140 „
5.42 „ 138 „

B, no food.
β-i injected 4.53.
Blood-pressure.
Before injection.
4.50 p.m. 120 mm. Hg.
After injection.
4.57 p.m. 76 mm. Hg.
5.15 „ 68 „
5.30 „ 72 „
5.44 „ 76 „

Amount of β-i absorbed:
Loops $A_1 = 23$ per cent., $A_2 = 23$ per cent.
Loops $B_1 = 33$ per cent., $B_2 = 38$ per cent.

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