"On the Production of a Specific Gastrotoxic Serum.—Preliminary Communication." By CHARLES BOLTON, M.D., B.Sc., M.R.C.P., Research Scholar of the Grocers' Company. Communicated by Professor SIDNEY MARTIN, F.R.S. Received July 26, 1904.

(From the Pathological Laboratory, University College.)

[PLATES 8 AND 9.]

Within the last four or five years the study of cytotoxins has advanced rapidly, but this difficult subject is still involved in a vast amount of obscurity. Very few of the cytotoxins have been at all fully worked out, and, since it is becoming recognised that specificity is a term which should be applied to receptors and not to cells, the existence of many specific cytotoxins is doubted.

The present research was, therefore, undertaken from two points of view: (1) To add, if possible, any facts to our knowledge of cytotoxic action, (2) To throw some light upon the pathology of human gastric ulcer.

My first endeavour was to produce a hetero-gastrolytic serum by the injection of the mucous membrane of the stomach of the guinea-pig into the rabbit. Having succeeded in this direction, I then attempted to produce an iso-gastrolytic serum by the injection of the stomach cells of the rabbit into the rabbit and also of the stomach cells of the guinea-pig into the guinea-pig. In commencing the work I fully realised the difficulty of excluding bacterial infection and therefore have adopted strictly aseptic measures in the preparation of the mucous membrane for injection, making in addition bacteriological examinations of the injected animals.

Intraperitoneal injections are given, and in spite of the strictest precautions death occasionally results from bacterial infection; I have, however, succeeded in immunising 14 animals, and by injection of the serum obtained from them have produced lesions in the stomachs of over 50 animals, and have been able to show that the peritoneal cavity of immunised animals was sterile on bacteriological examination, whilst their blood serum was highly toxic for other animals.

The subject will be discussed under the following headings:—

(1.) Methods.
(2.) Effects of Injection of the Stomach Cells of the Guinea-pig into the Rabbit.
    (*a*) Symptoms following injection.
    (*b*) Changes in the rabbit's blood.
        Action of the serum on guinea-pig's cells *in vitro*.

Action when injected into the guinea-pig.
Symptoms following injection.
Post-mortem lesions in guinea-pig.
Nature of the gastrolytic serum.
Effects of heat.
Specificity of the serum.
Relation to hæmolysin.
(*c*) Changes in the rabbit's peritoneum.

(3.) Effects of Injection of the Stomach Cells of the Rabbit into the Rabbit.
Changes in the rabbit's blood.
Action on injection into the rabbit.
Action on injection into the guinea-pig.
Effects of heat.
Effects of previous treatment with guinea-pig's stomach cells.
Effects of previous treatment with rabbit's stomach cells.

(4.) Effects of Injection of the Stomach Cells of the Guinea-pig into the Guinea-pig.
Action on injection of the serum into the rabbit.

(5.) Action of Normal Rabbit's Serum on Injection into the Guinea-pig.

(6.) Action of Hæmolysin on Injection into the Guinea-pig.

(7.) Lesions in the Guinea-pig's Stomach due to Causes other than Hæmolysin or Gastrolysin.

(8.) Conclusions.

(1.) Methods.

Preparation of Mucous Membrane for Injection.—The instruments used for this purpose are a knife, a porcelain plate, a glass mortar, and a 10 c.c. syringe, all of which can be sterilized with the greatest ease.

A guinea-pig which has been previously starved for about 24 hours, so that its stomach is empty, is killed with chloroform and the stomach at once cut out. It is opened and thoroughly washed in a stream of sterilized 0·86-per-cent. salt solution, all the adherent mucus being completely removed. The mucous membrane is then scraped off and ground up into an emulsion with salt solution, which is then injected into the peritoneal cavity of a rabbit. I have also used subcutaneous injections and have obtained the same results, but as suppuration is so liable to occur I have discarded them.

In two cases a fresh filtered extract of the mucous membrane was used for intraperitoneal injection. In one of these cases a 0·1-per-cent. hydrochloric acid solution and in the other a 0·4-per-cent. sodium bicarbonate solution was used for making the extract. The mucous

membrane was ground up in a glass mortar with a small quantity of sterilized sand in order to break up the cells, and an emulsion made with one of the above solutions. The emulsion was then filtered through a Pasteur-Chamberland filter and the resulting filtrate used for injection.

The effects obtained on injecting the serum of these two rabbits into guinea-pigs were exactly the same as those which resulted from injection of the serum of rabbits which had received injections of stomach cells.

Collection of Blood.—The blood is collected by opening a vessel of the ear, if possible an artery, having first shaved and disinfected the skin. In this way from 30–40 c.c. of blood can easily be obtained, without injury to the animal, especially if a flask of hot water be placed in contact with the ear to promote vaso-dilatation. The blood is whipped and centrifugalised.

The resulting serum is examined: (1) With regard to its action upon the stomach cells of the guinea-pig *in vitro*; (2) With regard to its action upon the guinea-pig's stomach when injected into the peritoneal cavity of that animal.

Examination in Vitro.—My plan has been to take a scraping from a fresh guinea-pig's stomach and carefully tease out the stomach glands in salt solution, separating them from the submucous and other tissue mixed with them. This is quite easily done by using the low power of a microscope or a dissecting lens. A portion of these glands is then placed in the serum to be examined, and a similar portion in normal rabbit's serum, the latter being used as a control.

The glands are examined at various periods of time up to 24 hours in the fresh condition, and also after fixing with osmic acid and staining with picro-carmine. In several cases small portions of the stomach wall were immersed in the serum for various periods of time and finally hardened, cut into sections in paraffin, and stained with hæmatoxylin and eosin.

Preparation for Microscopical Examination.—Patches of necrosed tissue with surrounding healthy mucous membrane were cut into a series of sections by the paraffin method. Eight or ten adjacent sections were selected from each row and fixed on slides, from six to twelve slides being thus obtained from each block. The sections were stained with hæmatoxylin and eosin. The stomachs of twenty animals have been investigated in this manner.

(2.) Effects of Injections of the Stomach Cells of the Guinea-pig into the Rabbit.

(a) *Symptoms following Injection.*

The immediate symptoms in the rabbit are those of collapse, the temperature sinking to about 96°, the respiration being accelerated, and

the animal prostrate. This is simply a vascular phenomenon due to the shock of injection, and the condition is soon recovered from, the animal subsequently taking its food well, and seeming to be quite healthy. Sometimes there is a fall in body weight, but this is by no means constant.

I have not found that any toxic symptoms follow the injection, and if the animal dies it is the result of bacterial infection.

(b) *Changes in the Rabbit's Blood.*

After four or five injections at intervals of 10 days, the blood serum of the rabbit is found to possess toxic properties.

Action in Vitro.—Contrary to the results of several observers, who have found that many tissue cells become dissolved in their corresponding cytotoxic sera, I have never been able to detect any difference whatever between the glands which had been exposed to the gastrolytic serum and those which had been similarly treated with normal rabbit's serum, although a marked effect was obtained on injecting the former serum into guinea-pigs.

In those cases in which the glands had been exposed for long periods of time to the two sera, each showed an equal degree of maceration in proportion to the time of exposure.

Action on Injection into Guinea-pigs.

Symptoms following Injection.—In about half-an-hour after intraperitoneal injection of the serum the symptoms are well marked. The animal sits huddled up, with hair erect, and will not move. The temperature becomes subnormal.

If the dose of serum is small (1—5 c.c.), the animal will probably have quite recovered by the following day, but with large doses (10 c.c.) it becomes rapidly worse, and finally general twitchings commence, and the animal lies prostrate, death occurring generally within 24 hours.

In one case a guinea-pig weighing 280 grammes was killed by 5 c.c., and in another an animal weighing 140 grammes by 1 c.c., but, as a rule, the killing power of the smaller doses is uncertain. It may be stated that in most cases a dose of 10 c.c. is fatal for a guinea-pig weighing from 200—300 grammes within 24 hours.

All guinea-pigs which are killed by the serum invariably show lesions in their stomachs, and a large proportion of those which receive small doses and recover show similar lesions. Speaking generally, the larger the dose administered, the greater is the effect upon the stomach. The lesions always occur during the first 24 hours after injection.

Macroscopic Appearance of Lesions in Stomach.—The lesions consist of patches of necrosis in the mucous membrane, and hæmorrhage. All the following lesions are described from cases which survived and

were killed on the following day, *post-mortem* lesions being thus absolutely excluded.

The areas of necrosis appear in the form of black patches, round or irregular in shape, from the size of a pin's head to a large area, occupying a third or more of the surface of the stomach, and contrasting markedly with the surrounding mucous membrane which is normal. The black colour is due to altered blood pigment, a variable amount of which is usually found clinging with particles of food to the necrosed patches, from which it can be washed off, leaving the blackened and necrosed tissue plainly visible. This altered blood is present in very variable amount, sometimes being in considerable quantity, at other times scanty, with the necrosis greatly in excess.

The patches are found most commonly near or on one of the curvatures, but they may occur anywhere in the stomach. Their usual situation on the curvatures may be due to the fact that the main blood-vessels of the stomach run along the latter, and, therefore, these parts are the first to be exposed to the action of the toxin. Sometimes, also, the patches spread out into streaks, passing along the anterior and posterior walls of the stomach from the curvatures, as if they followed the distribution of the blood-vessels. In some places the necrosed tissue has disappeared, leaving ulcers with blackened edges and bases. After about 48 hours the black tissue apparently disappears, leaving a perfectly clean and sharply punched out ulcer.

Microscopic Appearance.—The necrosed patches are sharply marked off from the normal tissue, and do not extend beyond the muscularis mucosæ, the muscular coats and peritoneum being normal.

The cells of the glands in the necrosed patch, when they can be distinguished, are seen to be diffusely and very faintly stained by the eosin, their outlines are clouded, and they look shrunken; the nuclei are not stained, although towards the margin of the patch they can just be distinguished.

In a small patch, which is apparently just commencing, the cells, both oxyntic and central, have the above appearances, whilst the interstitial connective tissue cells take the stain well, and show up by contrast. In a farther advanced stage the whole patch is diffusely stained a faint pink colour or totally unstained, and the gland cells are irregularly arranged more or less in columns, but no definite idea of what structure had preceded this patch can be made out. A certain amount of altered blood pigment in the form of scattered brownish granules can be seen, but, as a rule, the necrosed tissue is singularly free from blood.

Towards the edge of the patch the normal tissue contains a few necrosed cells, and the edges and base are infiltrated with leucocytes. In many sections the bases of the glands do not appear to be so disintegrated as the inner ends. Sometimes the patch looks like a

brownish coagulated mass in which no structure can be made out; at other times strands of interstitial tissue alone can be seen, the gland cells having completely disappeared.

A definite ulcer is formed by the gradual disappearance of the necrosed tissue. The blood in the vessels is generally normal, and frequently so when they are situated inside the necrosed patch. In the other cases it is apparently hæmolysed, and in one case a large mass of hæmolysed blood was seen infiltrating the submucous tissue.

The remaining portions of the alimentary canal have invariably been found to be normal, and no lesion has been detected in any other organ of the body. The primary lesion in this necrotic process is, I think, undoubtedly the result of a direct action of the gastrotoxin upon the gland cells, with a subsequent digestion of this tissue by the gastric juice, the black colour being due to the action of the gastric juice upon the blood which is contained in the necrosed patch.

As no change as a result of the action of the gastrotoxin *in vitro* can be made out, the questions must be raised whether the serum can produce necrosis when it attacks a cell having its normal lymph supply and normal connections in the body, or whether it only produces a devitalisation of the cell, thus allowing of its digestion by the gastric juice, which in this way renders the lesions visible. Previous work upon other cytotoxins does not settle the question one way or the other, because in some cases necrosis has been stated to occur, as in the liver, whilst in other cases no change has been found whatever, as in the pancreas and suprarenal body.

Hæmolysis undoubtedly occurs to a variable extent in different cases, and may account for a part of the hæmorrhage, though usually no laking of the blood can be seen in the vessels of the stomach.

In my opinion the lesions are not primarily due to hæmorrhage, because the appearance of a hæmorrhagic erosion is different from the necrotic patches described above; there are no hæmorrhages to be seen in the mucous membrane of the stomach, such as one would expect, and none occur in any other organ of the body. There is, moreover, no reason to suppose that the capillary endothelium of the stomach is sufficiently special in constitution to evoke the formation of a specific endotheliolysin.

Infarction can also be excluded by the limitation of the lesion to the mucous membrane, by the very irregular shape of the large patches, which sometimes contain islands of normal tissue, and by the fact that no clots can be found in the vessels either of the stomach or any other organ.

In some cases where the hæmolytic factor is more than usually in evidence, there is no doubt that some of the erosions may be the result of such a factor, but in my experience these erosions present quite a different appearance from the necrotic patches described above, the

tissue being obviously broken up by the blood. It is a remarkable fact that evidence of hæmolysis should be found in the stomach and not in the other organs of the body in these cases.

Nature of the Gastrolytic Serum.—The experiments which I have made indicate that the gastrotoxin consists of an "immune body" or "amboceptor," which is newly formed in the blood as a result of the injection of the stomach cells, and a "complement" which is contained in the normal blood serum.

Effects of Heat.—If the serum be heated to from 55—60° C. for 1 hour its action on injection into the guinea-pig is destroyed, although the control animals which are injected with unheated serum show extensive necrosis of the stomach. On the other hand, if heated serum be added to an equal volume of normal rabbit's serum, the action is restored when the mixture is injected into the guinea-pig, but the resulting lesions are not quite so extensive as in the case of the unheated serum.

The guinea-pig's complement does not appear to possess a haptophoric affinity corresponding to the complementophile affinity of the gastrolytic amboceptor. The action of a hæmolysin, formed by injecting a rabbit with guinea-pig's corpuscles, is, on the contrary, not destroyed by heat; this agrees with the test-tube phenomenon that guinea-pig's normal serum will reactivate the heated hæmolytic serum of a rabbit.

Specificity of the Gastrotoxin.—This property has been tested by mixing various cells with gastrolytic serum previous to its injection into the guinea-pig, in order to determine whether guinea-pig's stomach cells alone or whether any other cells could extract the amboceptor.

The cells were allowed to remain in contact with the serum for 1 hour at laboratory temperature, and were then centrifugalised off and the supernatant serum injected. Control animals were in each case injected with untreated serum.

Admixture of Guinea-pig's Stomach Cells.—Whether the stomach cells are washed free from blood or not they anchor the amboceptor and carry it down on centrifugalisation, with the result that the serum is rendered inactive on injection into the animal.

Guinea-pig's Liver Cells.—The liver cells washed free from blood fail to extract the amboceptor, and necrosis of the stomach results when the serum is injected.

Guinea-pig's Red Blood Corpuscles.—The experiment was done in two ways, and in each case the red corpuscles failed to extract the amboceptor. In one case the gastrolytic serum was cooled to 0° C. and mixed with the corpuscles; the mixture being kept at 0° C. for 1 hour, was afterwards centrifugalised. In another case the serum was heated to 60° C. for an hour; excess of red corpuscles was then

added to it and the mixture kept at laboratory temperature for 1 hour. The corpuscles were then centrifugalised off and normal rabbit's serum added to the supernatant fluid. On injecting this mixture into a guinea-pig stomach lesions resulted.

It was proved that the corpuscles had extracted the hæmolytic factor, because on adding to the heated serum some guinea-pig's corpuscles and normal guinea-pig's serum, complete laking resulted, whereas on adding the corpuscles and normal serum to the gastrolytic serum, which had been treated with red corpuscles previously, no laking or only a slight discoloration of the serum immediately above the deposited corpuscles was visible after 24 hours.

Rabbit's Stomach Cells.—These cells also failed to extract the gastrolytic amboceptor. As will be seen later, this experiment stands on a different footing from the last three, and the fact of the cells failing to extract the amboceptor does not prove that they do not possess an affinity for such amboceptor.

Relation to Hæmolysis.—The specificity of the gastrotoxin was also tested by immunising a rabbit against washed guinea-pig's stomach. The washing was carried out by passing a cannula into the thoracic aorta, cutting the inferior vena cava above the diaphragm, and then allowing a stream of sterilised salt solution to flow through the cannula until the blood was completely washed out of all the abdominal organs. This experiment is quite easily accomplished.

The serum of this rabbit showed a great increase in its hæmolytic power after it had received three injections of stomach cells, and the serum on injection into a guinea-pig caused necrosis of the stomach. This experiment affords confirmation of the fact, which was first demonstrated in the case of spermatozoa by Moxter and in the case of ciliated epithelium by Von Dungern, that different kinds of cells may contain similar receptors.

In the present instance the gastric cells not only contain specific receptors peculiar to themselves, but they also contain receptors similar to those of red blood corpuscles. Whether other cells of the body may contain receptors similar to those which I have stated as being peculiar to the gastric cells the above experiments do not conclusively prove. So far as they go the experiments prove that, at any rate if present in other cells, the gastric receptors are not so abundant in them as in the gastric cells themselves; other cells may contain a few of those receptors, but not sufficient to extract all the immune body contained in the serum.

At any rate, in the present state of our knowledge, these experiments tend to show that the gastrotoxin not only contains a specific amboceptor, which has an affinity for the receptors peculiar to the gastric cells, but also an amboceptor having an affinity for the receptors which the gastric cell possesses in common with red blood

corpuscles. The term "specificity" used strictly should not, therefore, be applied to the gastrolytic serum itself, but to that part of it which has a specific affinity for the receptors peculiar to the gastric cells.

(c) *Changes in the Rabbit's Peritoneum.*

If an animal which has received several injections be killed a few weeks after the last one, the peritoneum will show slight opacity and thickening in various places, or slight adhesions between adjacent coils of intestine may be seen, or small pedunculated nodules may be present. The remaining portions of the peritoneum are quite normal.

The nodules on section are found to be composed of concentric layers of fibrous tissue, generally inclosing within them a group of epithelioid cells, and occasionally a giant cell, the nuclei of which are arranged in the centre. Whether these nodules are formed during the absorption of the stomach cells, as they were not seen in the cases in which an extract of stomach cells was injected, or owing to the presence of isolated bacteria, I am not prepared to state. At any rate, a bacteriological examination of such a peritoneum shows that it is sterile.

A few days after an injection the peritoneum is in places smeared over with a thin layer of cheesy material, and masses of the same material may exist between the coils of the intestine; elsewhere the peritoneum is normal. Microscopically, this substance consists of proliferated peritoneal epithelium and exuded leucocytes. This substance is also sterile on cultivation.

Last year, after I had commenced the present research, a paper was published by Théohari and Babès on a gastrotoxin. This is the only paper, so far as I am aware, dealing with the subject, and, as the authors obtained different results from mine, I will here briefly enumerate them.

The mucous membrane of the dog's stomach was subcutaneously injected into the goat, and, on injection of the goat's serum into the dog, they obtained the following results :—

(1) Pronounced hypersecretion of the stomach cells in the case of the weak gastrotoxic serum.

(2) Rapid death, with intense hyperæmia of the stomach, and especially of the intestine in the case of the stronger serum.

(3) In small doses, excitation of gastro-intestinal peristalsis and intestinal hæmorrhage. The chief cells in the stomach show functional changes; the marginal cells show degenerative changes. No change in the pyloric region of the stomach or the large intestine.

(4) The serum, specific against the peptic region of the dog's stomach, produces, moreover, material alterations in the small intestine, whilst the large intestine remains normal.

(3.) Effects of Injection of the Stomach Cells of the Rabbit into the Rabbit.

The same initial symptoms, and the same changes in the rabbit's peritoneum, are observed as in the case of injection of guinea-pig's stomach cells.

Changes in the Rabbit's Blood.—I have been unable to produce any stomach lesion, or demonstrate any toxic action, on injecting the serum into a rabbit.

Action on Injection into the Guinea-pig.—That the blood has acquired toxic properties, however, I have shown by injecting the serum into guinea-pigs. Identical lesions are obtained by such injections with those described above.

Effects of Heat.—My experiments so far tend to show that, on heating the serum to 50—60° C. for 1 hour, the action is destroyed if the serum has only a low toxicity, but that some action may remain in the case of the more toxic sera. According to these results, the guinea-pig serum may to some extent complement this amboceptor.

Effects of Previous Treatment with Guinea-pig's Stomach Cells.—The stomach cells extract the amboceptor, and render the serum inactive, as described in dealing with the serum obtained by injecting the rabbit with guinea-pig's cells.

Effects of Previous Treatment with Rabbit's Stomach Cells.—In this case the rabbit's stomach cells entirely fail to anchor the amboceptor, and, on injecting the serum thus treated into a guinea-pig, necrosis of the stomach results. The same result was seen in the serum of rabbits immunised with guinea-pig's cells.

From these experiments I think it may be inferred that the rabbit can produce a gastrolytic serum when rabbit's stomach cells are injected into its body. And also, that the immune body thus produced has two cytophilic affinities: (1) for the receptors of the rabbit's stomach cells, since it is formed in response to their injection; (2) for the guinea-pig's stomach cells, since it causes necrosis of them.

Another alternative would be to assume that two separate immune bodies are formed. Whichever of these two views be accepted, the conclusion follows that the failure of the rabbit's stomach cells to anchor the immune body for which they have an affinity points to the hypothesis that this junction is prevented by the interposition of an anti-immune body, this anti-immune body having presumably been formed by the rabbit to protect its own stomach and prevent autolysis.

If we accept the view that two separate immune bodies are formed, we must also conclude that the rabbit's stomach cell has receptors similar to the guinea-pig's, as well as those peculiar to itself; this view presents the difficulty that we cannot explain why the guinea-

pig's cells should anchor the immune body, whilst the rabbit's cells will not. On the other hand, if we accept the view that there is one immune body only, but that it has two cytophilic affinities, that corresponding to the rabbit's receptor being saturated by an anti-immune body, but that corresponding to the guinea-pig's receptor being free, the above difficulty vanishes.

(4.) Effects of Injection of the Stomach Cells of the Guinea-pig into the Guinea-pig.

The serum in this case was shown to be toxic by injecting it into a rabbit, when typical necrosis resulted. A difficulty encountered here is due to the fact that a sufficient amount of blood cannot be obtained from the guinea-pig in order to inject an amount corresponding to the weight of the rabbit which would lead to a result as marked as that obtained in the guinea-pig's stomachs described above.

This experiment also shows that when an animal absorbs cells which have been taken from the body of an identical animal its blood becomes toxic.

(5.) Action of Normal Rabbit's Serum on the Guinea-pig.

The blood serum of the rabbit is to some extent hæmolytic for the guinea-pig's red corpuscles. On injection of the serum into a guinea-pig, even in large does (20 c.c.), no lesions are found in the stomach or alimentary canal such as result from hæmolysis. In one case a patch of engorged vessels was seen in the stomach, but no hæmorrhage or necrosis was to be observed.

(6.) Action of Strongly Hæmolytic Serum.

After a rabbit has received several injections of guinea-pig's red corpuscles a marked effect is observed on injecting the rabbit's blood serum into a guinea-pig. The lesions produced consist of hæmorrhage with distortion and solution of the blood corpuscles. The hæmorrhages are best seen in the alimentary canal. They occur in the stomach and lead to erosions of the mucous membrane as a secondary consequence, but patches of necrosis in the mucous membrane are not seen. Hæmorrhages are also seen in the lymphatic follicles of the intestine, especially the colon. There is intense engorgement of the organs and hæmorrhages may occur all over the mesentery.

Effects of Heat.—On heating the hæmolytic serum to 50—60° C. for an hour, its action is not destroyed, the guinea-pig's serum being able to complement the hæmolytic amboceptor. The differences between the action of a hæmolytic and a gastrotoxic serum are thus seen to be very considerable.

(7.) Lesions in the Guinea-pig's Stomach due to Causes other than Hæmolysis or Gastrolysis are occasionally seen.

If the stomachs of a large number of normal guinea-pigs are examined regularly, there may occasionally be seen patches of congestion in the mucous membrane; sometimes hæmorrhages are present, and in two cases I have found ulcers. What the pathology of this condition is does not seem quite clear, but probably the ulcers are of hæmorrhagic origin.

In two cases of septic peritonitis in guinea-pigs I have found hæmorrhage into the stomach and small intestine. On microscopic examination they present a very typical appearance, the blood being extravasated into the substance of the mucous membrane, and exhibiting quite a different appearance from patches of necrosis.

(8.) Conclusions.

(*a.*) On either intraperitoneal or subcutaneous injection of the stomach cells (or a fresh extract of them) of the guinea-pig into the rabbit, the blood serum of the latter becomes highly toxic for guinea-pigs.

(*b.*) The serum leads to death on injection into the guinea-pig and causes necrosis of the mucous membrane of the stomach, leading to ulceration and hæmorrhage.

(*c.*) The toxin contained in the serum consists of at least two factors: (1) A specific Gastrolysin, which leads to necrosis; (2) A Hæmolysin which assists in producing hæmorrhage. The hæmolytic factor can be removed, leaving the gastrolytic, which still produces stomach lesions.

(*d.*) The gastrolysin is a specific cytotoxin and consists of an immune body and a complement.

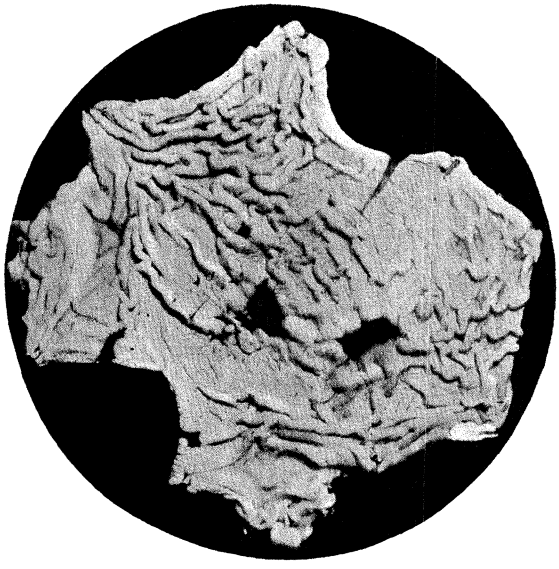
(*e.*) The gastrolysin does not visibly affect the cells *in vitro*.

(*f.*) The gastrolysin does not produce necrosis in the stomach of the animal which has elaborated it, possibly owing to the concomitant formation of an anti-immune body.

(*g.*) By injection of the stomach cells of the rabbit into the rabbit a gastrolysin is formed which causes necrosis in the guinea-pig's stomach; it therefore possesses at least two cytophilic affinities.

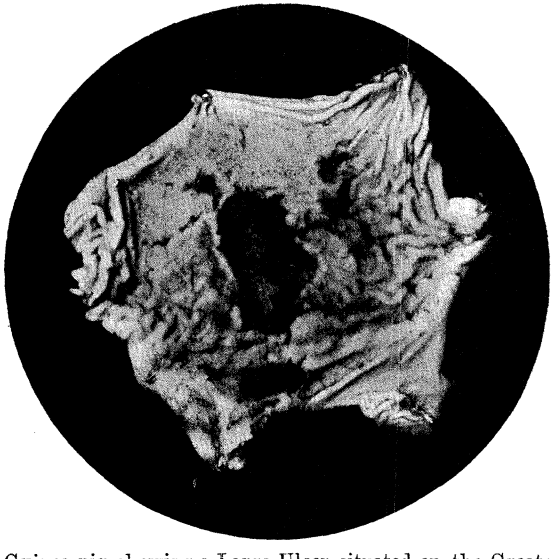
(*h.*) By injection of the stomach cells of the guinea-pig into the guinea-pig a gastrolysin is formed which causes necrosis in the stomach of the rabbit; this gastrolysin is probably of a similar nature as the preceding.

(*k.*) The importance of the above conclusions with regard to the pathology of human gastric ulcer lies in the fact that an animal can elaborate in its blood by the absorption of the cells of a similar animal,



Stomach of Guinea-pig showing Two Ulcers. The animal received an injection of 12 c.c. of a weak gastrotoxic serum, and after 24 hours it was killed.

The serum was prepared by injecting a rabbit with fresh *extract* of guinea-pig's stomach cells.

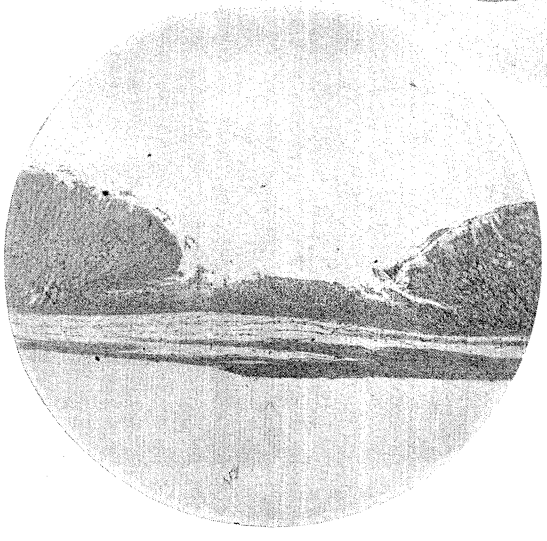


Stomach of Guinea-pig showing a Large Ulcer situated on the Greater Curvature and several Smaller Ones. The animal received an injection of 12 c.c. of gastrotoxic serum, and was almost dead after 24 hours, when it was killed.

The serum in this case was prepared by injecting a rabbit with *rabbit's* stomach cells ; in the other four cases *guinea-pig's* stomach was used for injection.

Section of Stomach Wall of Guinea-pig showing a large area of Necrosis in the Mucous Membrane. The animal received an injection of 4 c.c. of gastrotoxic serum, and was killed 24 hours afterwards.

Section of Stomach Wall of Guinea-pig at the edge of a Necrotic Patch. A blood-vessel containing normal red corpuscles can be seen in the patch. The animal received 5 c.c. of gastrotoxic serum, and was killed the next day.



Section of Stomach Wall of Guinea-pig showing an Ulcer. The animal received 10 c.c. of weak gastrotoxic serum, and was killed 48 *hours* afterwards.

and therefore presumably of its own, a toxin which would cause necrosis in the mucous membrane of its own stomach were it not for some deterring influence, probably the concomitant formation of an anti-immune body.

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"On the Action exerted upon the *Staphylococcus pyogenes* by Human Blood Fluids, and on the Elaboration of Protective Elements in the Human Organism in Response to Inoculations of a Staphylococcus Vaccine." By A. E. WRIGHT, M.D., late Professor of Pathology, Army Medical School, Netley, Pathologist to St. Mary's Hospital, Paddington, W., and STEWART R. DOUGLAS, M.R.C.S., Captain, Indian Medical Service. Communicated by Sir J. BURDON SANDERSON, Bart., F.R.S. Received July 26, 1904.

(From the Pathological Laboratory of St. Mary's Hospital, London, W.)

The subject matter with which we have here to deal may be distributed under the following headings:—

(1) Determination of the nature of the action which is exerted upon the *Staphylococcus pyogenes* by normal human blood fluids, and by the blood fluids of patients who have been inoculated with a staphylococcus vaccine.

(2) Comparison of the phagocytic power of the subjects of staphylococcus invasion with the phagocytic power of normal individuals.

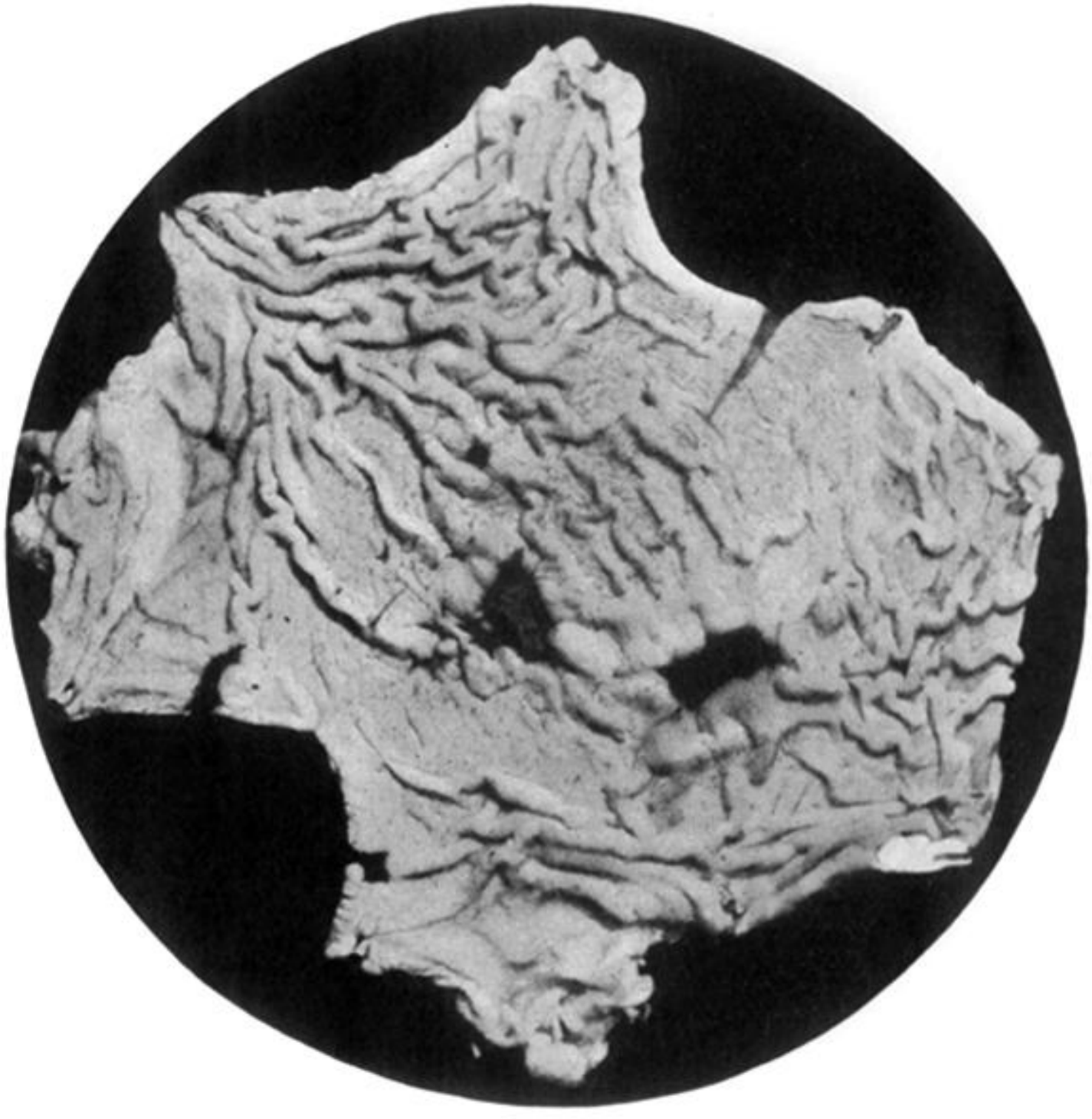
(3) Distribution in the infected organism of the opsonins which here come into consideration.

(4) Determination of the question as to whether the opsonins are present in the blood of the infant at birth.

(5) Determination of the course of the reaction of immunisation which supervenes upon the inoculation of a staphylococcus vaccine.

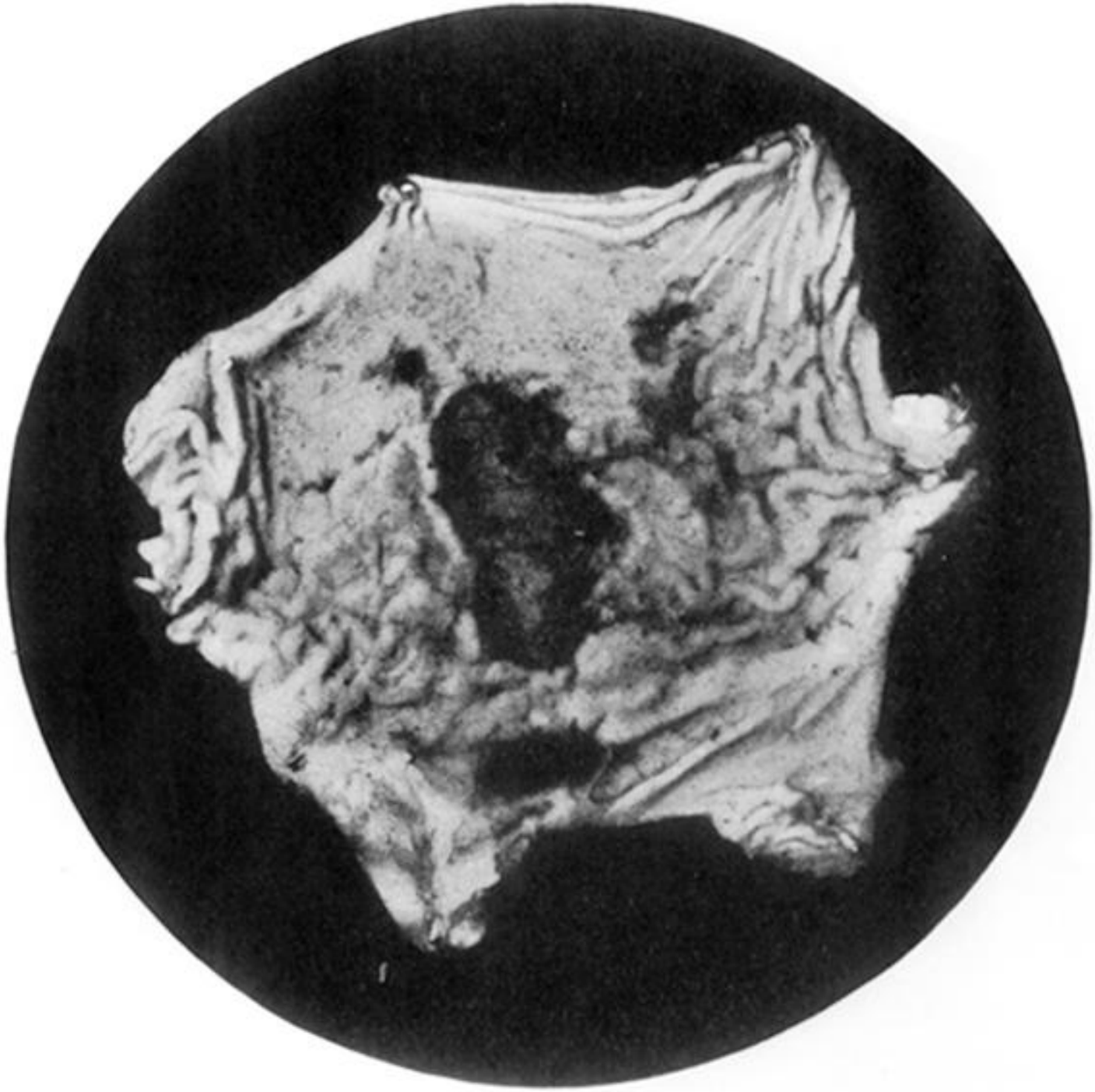
(1) *Nature of the Action which is exerted upon the* Staphylococcus pyogenes *by Normal Blood Fluids, and by the Blood Fluids of Patients Inoculated with a Staphylococcus Vaccine.*

Bactericidal Action.—It was shown in the course of the classical researches on the bactericidal power of the blood which were conducted



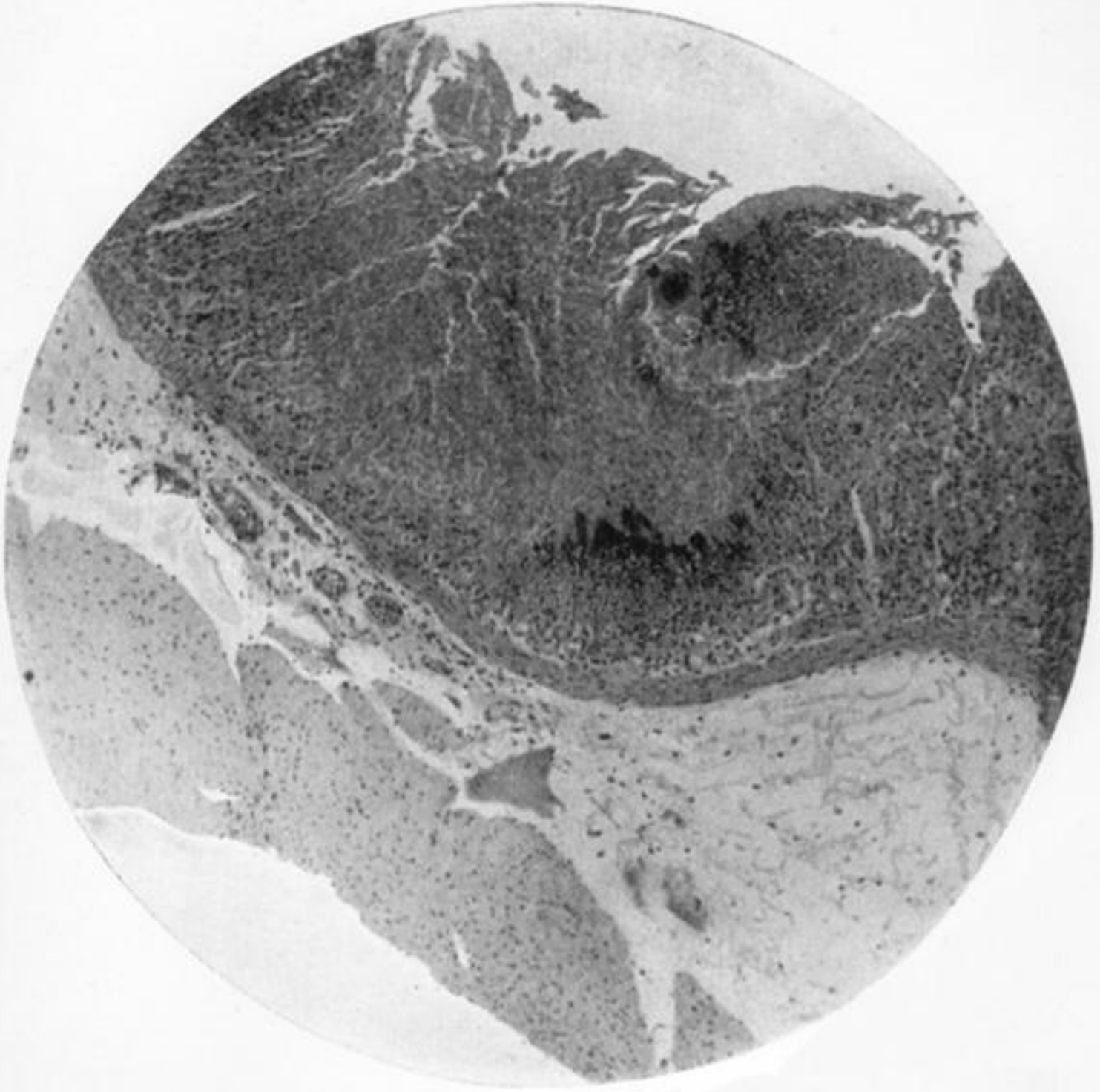
Stomach of Guinea-pig showing Two Ulcers. The animal received an injection of 12 c.c. of a weak gastrotoxic serum, and after 24 hours it was killed.

The serum was prepared by injecting a rabbit with fresh *extract* of guinea-pig's stomach cells.



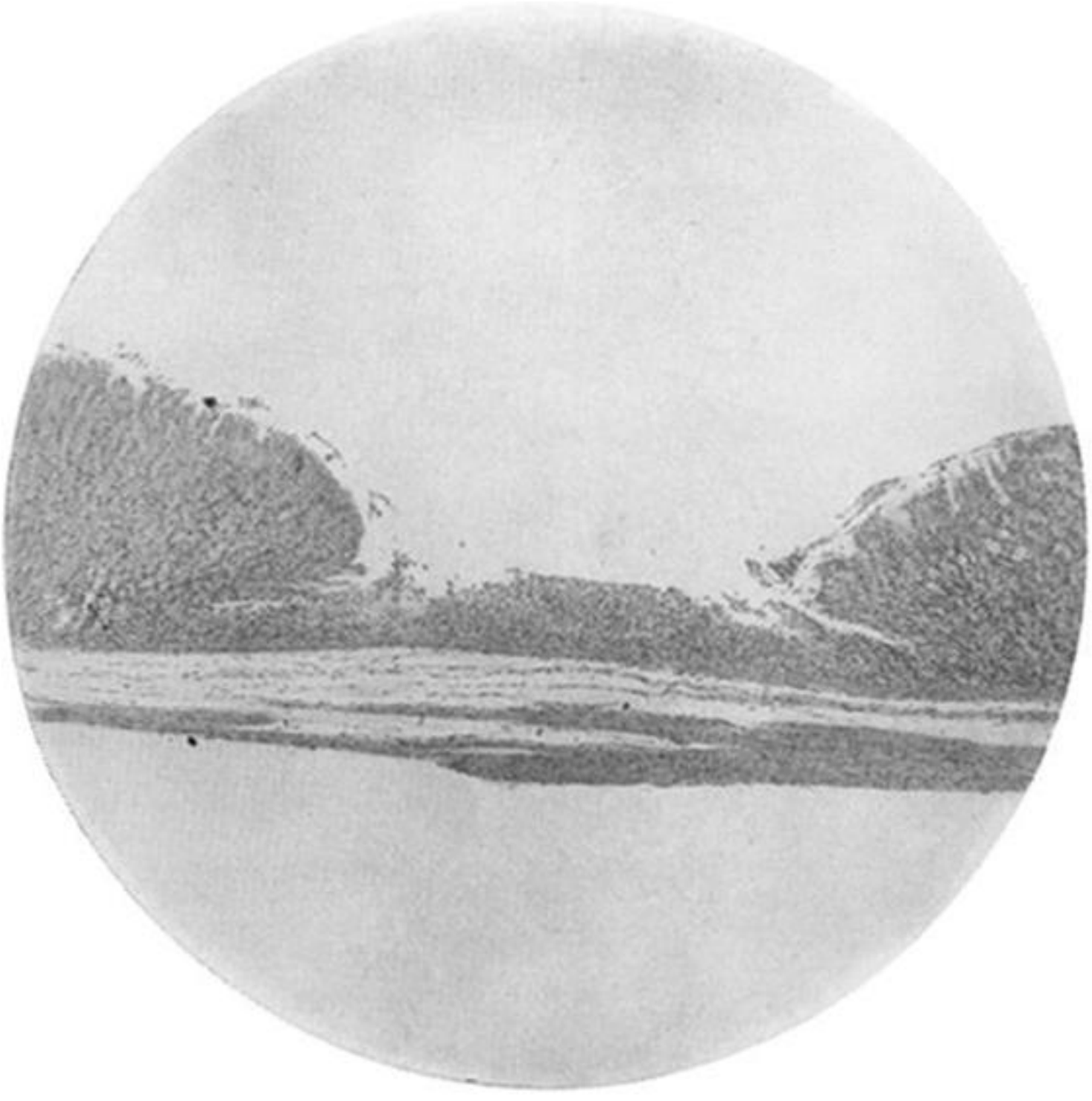
Stomach of Guinea-pig showing a Large Ulcer situated on the Greater Curvature and several Smaller Ones. The animal received an injection of 12 c.c. of gastrotoxic serum, and was almost dead after 24 hours, when it was killed.

The serum in this case was prepared by injecting a rabbit with *rabbit's* stomach cells; in the other four cases *guinea-pig's* stomach was used for injection.



Section of Stomach Wall of Guinea-pig showing a large area of Necrosis in the Mucous Membrane. The animal received an injection of 4 c.c. of gastrotoxic serum, and was killed 24 hours afterwards.

Section of Stomach Wall of Guinea-pig at the edge of a Necrotic Patch. A blood-vessel containing normal red corpuscles can be seen in the patch. The animal received 5 c.c. of gastrotoxic serum, and was killed the next day.



Section of Stomach Wall of Guinea-pig showing an Ulcer. The animal received 10 c.c. of weak gastrotoxic serum, and was killed 48 *hours* afterwards.