of Dr. Luys, and the practical fact that Cathelin's urine separator can be used with the patient lying down has a distinct advantage over that of Dr. Luys. Since Dr. Bickersteth's interesting paper appeared¹ Luys's instrument is becoming well known in this country.

Cathelin's urine separator is shaped like an English lithotrite, and has a small catheter on each side of its stem and beak. Before the instrument is introduced into the bladder, the partitioning diaphragm is withdrawn into the stem, until the mark "o" on the carrying rod indicates that it is completely shielded; the other figures, graduated from 10 to 300, indicate that a globe that would hold one of the specified quantities in grams would be bisected through its middle by the diaphragm projected to that index. For example, when one leaves 40 grams of water in the bladder, the rod is pushed into the stem until the figure 40 is about to disappear, then the diaphragm fills and bisects the bladder into lateral halves.

In my practice I have had some very striking illustrations of the efficacy of the instrument in keeping unmixed the two urines in the bladder. In our consulting rooms I consider the use of the urine separator is less objectionable to females than that of the cystoscope. Ten to twenty minutes is long enough to collect sufficient urine for diagnosis. In one of Dr. Herbert Vachell's cases I left the instrument in a male patient's bladder for one hour and twenty-five minutes, with very little discomfort. This patient had pains—back-ache on the left side—and the left kidney secreted but one-fifth the quantity of the right kidney. I performed decortication of the left kidney with satisfactory results.

In this paper I do not propose to give reports of cases, but would venture to refer to one that is a most striking example of perfect partitioning of the bladder. In another patient of Dr. Herbert Vachell's at the Cardiff Infirmary, who had haematuria with a large palpable tumour of the right kidney, Cathelin's instrument drew highly-coloured bloody urine from the right side, and clear urine containing albumen from the left. The coloured right urine was alkaline in reaction, whilst the left clear urine was acid. I was inclined to think the albumen on the left side might be due to a trace of the right urine escaping by the diaphragm. Five weeks ago I removed the kidney with the tumour (carcinoma); since then the urine has remained clear, but still contains albumen, as it did before removal of the diseased kidney.

For demonstration purposes urines can be fixed in test tubes by means of gelatine and formalin, and I have been much struck with the difference in colour of normal urine from the two kidneys; in some cases one kidney seems to possess greater affinity for extracting colouring material than the other.

I have once been saved from removing a tumour of the only functional kidney. An old lady had a large right renal tumour; I took her into the infirmary for operation. As I had just received Cathelin's instrument I used it, but to my surprise I could only get urine from the diseased side. I thought that this might possibly be due to the diaphragm not being placed in the middle of the bladder, and that it pressed on the left ureteral orifice. I made a second examination with the same result. A third examination under an anaesthetic gave exactly the same result, and I also found by cystoscopic examination that the left ureteral orifice was a thin U-shaped slit with no peristaltic movements. I injected water coloured with potassium permanganate through the left catheter, and it returned the same colour without affecting the urine from the right side. The mere fact of anaesthetizing this old lady gave rise to serious uraemic symptoms for a fortnight. In cases of renal calculus with albuminuria I have found non-albuminous urine on the sound side. From the above observations it seems clear we have at present reliable urine separators, and it is scarcely possible to bring forward a better illustration than the one briefly referred to, where the urine was alkaline on one side and acid upon the other, to show that we possess an instrument of very great value for diagnostic purposes.

REFERENCE.
Lancet, March, 1904.

THE LIGHT TREATMENT IN FLORENCE.—A committee, of which Professor Pellizzari is the chairman, has been formed to promote the establishment of a light institute for the treatment of lupus and other forms of cutaneous tuberculosis at Florence. The King of Italy has intimated his approval of the scheme, towards the execution of which he has contributed £120.

THE LEISHMAN-DONOVAN BODY IN ULCERATED SURFACES;

A POSSIBLE ROUTE OF ITS ESCAPE FROM THE HUMAN BODY.

BY

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IN the *BRITISH MEDICAL JOURNAL* of May 28th we described the presence of Leishman-Donovan bodies in the lymphatic glands of the mesentery. In later work it has been found that the distribution of the parasites in the glands is not uniform, some containing none, some a few, others enormous numbers.

A careful examination of the intestine of the case before mentioned revealed a small ulcer about 12 in. above the ileo-caecal valve, three small punched-out ulcers at the splenic flexure, and two areas of atrophied mucous membrane, one 15 in. or thereabouts above the caecum, the other in the upper part of the ileum.

The mesentery presented numerous small nodules, somewhat resembling minute tubercles; similar nodules were present on the peritoneal surface of the small intestine, and also at the base of the small intestinal ulcer.

Microscopically the intestine exhibited areas of small-celled infiltration between the mucous glands, and in the sub-mucous coat, the atrophied areas showing similar though more advanced changes, most of the mucous membrane having disappeared.

In the infiltrated areas of the small intestinal ulcer, and in one of the ulcers from the large intestine, Leishman-Donovan bodies, few in number, it is true, but unmistakable, were found; they were enclosed in cells. In the other areas no parasites were detected, but as the bodies were so few in the ulcers it is possible that they may yet be found here also.

In sections of liver, spleen, and lymphatic glands the position of those bodies is undoubtedly, in the majority of instances, intracellular. In many cases the protoplasm of the cell is swollen, the nucleus staining faintly or having disappeared.

We have never seen the parasites in the red blood corpuscles in sections or otherwise. Wright and James have found the Leishman-Donovan, or a similar organism, in ulcers of the integuments (Oriental sore), Christophers in intestinal ulcers. The foregoing note again records their occurrence in ulcers of the intestinal mucosa, and this is interesting in view of the fact that dysenteric ulceration is a very usual termination for kala-azar.

Failing the discovery of any other means of exit from the body, may it not be that the normal route of escape for the parasite is by an ulcerated surface? With this idea in view we lately produced by croton oil and liquor epispasticus slight ulcerating dermatitis in a patient with Leishman-Donovan bodies in his viscera, but failed to find the parasites in the discharges. This experiment cannot, however, be considered conclusive, as we were afraid, in the interests of the patient, to produce anything like deep ulceration.

The above observations confirm, then, the work of Christophers recently published in India, and are of interest in showing that these parasites may escape by the intestinal canal.

PRELIMINARY NOTE ON THE CULTIVATION OF ANAEROBES.

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Most of the instruments devised for the cultivation of anaerobes are faulty in design, in so far as they do not allow of the organism being examined from time to time, and while it is growing, without disturbing the whole anaerobic arrangements; another drawback to their use is the uncertainty of obtaining typical surface cultures. As in the excellent instrument planned by my assistant, Mr. A. R. Laing, M.B., Ch.B. (*Lancet*, February 20th, 1904, p. 515) their construction is generally based on the principle of substituting an atmosphere of hydrogen for air.

The method I am about to describe aims at the exclusion of any atmosphere whatever, and has proved remarkably efficient in the large experience I have now had of its employment. By means of it, cultures of anaerobes, such as the

bacillus tetani, can be started in a few minutes; samples of the organism can be withdrawn from time to time without endangering the anaerobic conditions; and surface cultures can be obtained with the greatest facility and certainty.

The majority of the anaerobic organisms which are pathogenic in the animal body pass through a sporing stage in their life-history, and this renders their isolation a matter of comparative ease. The greater the number of its spores, the greater is the chance of isolating a particular anaerobe from surrounding impurities. There are many aerobic organisms and a few anaerobic, such as that of tetanus, which will spore on almost any medium. The members of a group of anaerobes with which I have been experimenting lately, and which I shall describe in a future publication, are not in this case. They usually fail to spore on any of the known media; they grow luxuriantly upon many of these, but by fission, even when the morbid liquid or tissue from which they have been derived is rich in spores. The means which I have found to be never-failing in encouraging sporulation, is to incubate (37.38° C.) the organism in the liquid upon which it is found growing naturally within the animal body. Thus supposing the organism to be growing naturally upon the lymph of the tissues, and to be in the bacillary phase of its life-history, all that is necessary in order to enable it to spore is to draw up the liquid into a Pasteur's pipette, to seal this off, and to place the tube in the incubator for from twenty-four to forty-eight hours. By this time every second or third bacillus will have developed a spore, and multitudes of free spores will probably be found in the surroundings. The drawing up of a little air into the pipette along with the liquid, curiously, does not seem to prohibit the tendency to sporulation. I do not think sufficient importance has been attached to this procedure as a means of bringing about such a very desirable result.

Having obtained the organism in the sporing state, its isolation is a matter of comparative ease, for it is my experience that where one pathogenic anaerobe which spores is present in excess, it has a decided influence in destroying or inhibiting the growth of any others having a like tendency.

The medium I employ for the purpose of obtaining a pure culture is glucose-peptone-beef-tea. For the growth of members of the group of organisms just referred to, its reaction must be distinctly alkaline to phenolphthalein. Mere alkalinity to litmus is not sufficient; indeed, a frequent cause of failure to obtain a culture is to be traced to the medium being neutral or only faintly alkaline. It must be boiled and filtered until precipitation of phosphates ceases. If precipitation of these does occur on boiling, it ought to be allowed to stand for twenty-four hours, and be filtered when cold. If this method be followed, it is seldom that precipitation occurs a second time, provided that the alkaline reaction is such as to give a slightly pinkish tint with phenolphthalein. The reaction ought always to be tested immediately before use, as the boiling may affect the result.

The medium thus prepared is decanted into test-tubes or flasks, according to the amount of culture required for any particular purpose. Test-tubes should be filled to about three-fourths of their depth and flasks to the commencement of the neck. Sterile olive oil is next poured over the medium to a depth of one and a-half centimetre. The olive oil seems to constitute a perfect atmospheric valve.

In order to sterilize the medium, with its covering of oil, and at the same time to drive out any oxygen which may be in solution, the tubes or flasks are boiled after the usual fashion for three-quarters of an hour on the first day, and for a quarter of an hour on the second and third. They will remain anaerobic and ready for inoculating after this for weeks at a time, or, I believe, even longer, but in order to make perfectly certain, I generally boil them again for a quarter of an hour immediately before use, and cool them down rapidly thereafter in a running stream of water.

The ingrafting of the spores is accomplished by means of a pipette. A small quantity of the liquid in which they are contained is drawn up in a capillary Pasteur's pipette, and being plunged deeply into the beef-tea, a particle of liquid is blown out, care being taken to avoid emptying the tube, as air might thus find access to the medium.

The cotton plug is readjusted, and the inoculated beef-tea is heated to 80° C. in a water-bath for twenty minutes, in order to kill off any non-sporing contaminations. In the case of the organisms I have been working with, it is seldom, if ever, that I have failed thus to get a pure culture.

If the organism be contained in a tissue, a small piece of

the latter is clipped off with sterile scissors and forceps and dropped into the tube or flask.

After retaining the medium at 80° C. for the specified time, it is cooled down again rapidly in a running stream of water, and incubated. Within twenty-four hours, as a rule, germination is in full activity, and any gas which escapes, of course, has free egress through the layer of oil on the surface. It is not necessary to cover the mouth of the test-tube or flask with a caoutchouc cap; indeed, I have generally found it preferable not to do so, for the gas generated may, if pent up by this means, burst the cap or even the vessel itself. The gas-bells sometimes accumulate as a froth on the surface of the oil, but this is unusual, and occurs only where the evolution of gas is excessive.

Should it be desired to examine the culture microscopically, a sterile pipette is introduced to the necessary depth and a little of the medium sucked up. Any oil which adheres to the outside of the pipette may be removed with blotting-paper before the drop required for examination is blown out on the slide.

Such is the method of obtaining the organism in pure culture. In making a surface growth, the medium which I have found most suitable is glucose-peptone-agar, but any other solid medium, not coagulated by heat, may be substituted. I do not make surface growths in test-tubes, but in round capsules having an inside diameter of seven centimetres, with a depth of two and a-half centimetres. They are provided with a flat ground flange one and a-half centimetre broad. The cover is made of plate-glass, and extends outwards to half the breadth of the flange, so as to leave a margin uncovered. It is ground to fit closely on to the flange.

The medium is introduced into the capsule for a depth, say, of one centimetre, and over this, as in the test-tube preparation, is poured sterile olive oil nearly up to the rim. The cover is adjusted, and the whole capsule and its contents are sterilized by boiling, any air contained in the latter being thus also effectually removed. The capsule is afterwards allowed to cool down.

The medium is inoculated with a sterile Pasteur's pipette, which, immediately before use, is drawn through a Bunsen flame so as to sterilize the outside. A small quantity of a pure culture on glucose beef-tea, made as before described, is drawn up into it, and with the point one or more streaks are scratched upon the medium below the oil. The plate-glass cover is readjusted, and the capsule placed in an incubator. The growth is usually abundant within twenty-four to forty-eight hours, but time should be allowed for it to reach the maximum.

Should it be desired to preserve the culture permanently, the procedure is as follows:—When the organism has grown to the necessary extent, the olive oil is carefully poured off. It is sometimes desirable to pipette off the superfluity of the oil, should the surface of the culture be easily disturbed, but the remainder is got rid of by inverting the capsule over blotting paper. In twenty-four hours the oil will be almost entirely removed by so doing.

The culture must now be killed either with formalin vapour, or, simply, by pouring the solution of formaldehyde into the capsule and allowing it to remain in contact for a day or more. The formalin is afterwards run off, and the capsule again inverted over blotting-paper, usually for thirty-six to forty-eight hours. By this time all superfluous liquid will have been absorbed, and the medium with the culture upon it now presents a comparatively dry surface and is ready for mounting.

Of all mounting liquids, as well from its optical properties as from its preservative action, I have found refined castor oil to be far the best. It is evidently an excellent preservative, when the organism has been killed by formalin, and is so transparent that instead of obstructing the view of the object, it enhances the lucidity of its demonstration. The oil is simply poured gently into the capsule, the lid adjusted, and the air driven out. The lid is thereafter luted with any appropriate cement, but before applying this, I generally run some pure collodion round the margin so as to absorb any castor oil which may be present.

GERMAN PUBLIC HEALTH ASSOCIATION.—The German Public Health Association will hold its annual meeting this year at Danzig from September 14th to the 17th. Among the subjects proposed for discussion are diarrhoea and its prevention; the technique of refrigeration for sanitary purposes; and the improvement and better organization of the personnel of attendants on the sick.