appendix and found it void of offence, and at the same time noting possibly the escape of a whiff of gas, and by the colour and odour of the peritoneal exudate that the lesion is of the stomach or duodenum, the latter are exposed by an incision splitting the upper right rectus. The escape of gas and gastric and duodenal contents are now usually obvious. The pylorus is pulled up, as in two cases out of three the perforation is within half an inch of this. If no perforation is found in this neighbourhood, the anterior surface of the stomach is explored manually up to the cardiac orifice, feeling for the induration around the perforated ulcer. Failing to find an ulcer on the anterior surface the stomach is pulled out with the transverse colon, and its posterior surface explored through an incision in the mesocolon.

A perforation is seldom of more than a quarter of an inch in diameter, though occasionally twice as large as this, and can be firmly occluded by the passage of one or two sutures. These sutures should secure a good wide grip through the whole thickness of the organ, since a small grip will easily tear out of the soft œdematous wall. The occluded ulcer should be invaginated where possible by a series of interrupted sutures taking up the serous and muscular coats. Invagination of the ulcer may, however, prove impossible if the ulcer and area of surrounding induration are very large, or in some instances where the ulcer is at the attachment of the duodenum to the posterior wall. In such cases the occluded ulcer is covered with a graft of detached omentum, or drainage is made down to the ulcer with a gauze pack (Corner [1]) in case the preliminary sutures cut out.

One must next consider whether a gastro-jejunostomy should be done. In most cases where the patient is not likely to die shortly we finish with a gastro-jejunostomy, especially where the ulcer is in the vicinity of the pylorus, since if this be done the patient can be fed after operation much more effectively, and there can be little doubt that many of these patients are suffering from malnutrition, the results of previous dyspepsia, which prevents healing taking place readily. This addition does not add greatly to the duration of the operation (the whole procedure from start to finish averages, we find, about 35 minutes) and improves the prospects of ultimate success. In the less usual cases where the ulcer is on the body of the stomach gastro-enterostomy is not so urgently needed, but nevertheless is advisable.

The Uses of Jejunostomy.

Where the patient's condition is extremely grave and every moment spent on the operation is of importance, we advise simply occluding the ulcer with one or two sutures, placing a gauze drain down to the site of perforation, and performing a jejunostomy for the purpose of feeding the patient early. Jejunostomy is performed on the invagination (Kader) principle, takes less than five minutes to perform, and has the advantage that fluid nourishment can be introduced to the most absorbent surface of the intestinal canal, in a situation where vomiting is impossible, and which, unlike the rectum, is unable to reject the proffered refreshment. The actual results of cases treated by this method were less good than were those of cases treated otherwise simply owing to the very grave condition of the patients; 1 recovered and 3 died. One of the latter, which had been perforated three days, lived four days after operation. Another lived 14 days, only succumbing at length to a slowly spreading peritonitis. We only regret that this method was not employed in some of our earlier severe cases which succumbed.

The following table shows the various operations adopted and their results:—

Operation.	Gastric ulcer patients. Lived.	Gastric ulcer patients. Died.	Duodenal ulcer patients. Lived.	Duodenal ulcer patients. Died.
Suture alone	1	3	2	2
Suture and gastro-jejunostomy ...	4	2	18	4
Suture and jejunostomy	1	2	0	1

My best thanks are due to my house surgeons for their notes on the above cases, and especially to Mr. W. S. Perrin, surgical registrar to the London Hospital, for his care in collecting and collating the histories.

Wimpole-street, W.

[1] THE LANCET, Jan. 10th, 1914, p. 101.

AN INVESTIGATION ON THE NATURE OF ULTRA-MICROSCOPIC VIRUSES.[1]

BY F. W. TWORT, L.R.C.P. LOND., M.R.C.S.

(*From the Laboratories of the Brown Institution, London.*)

DURING the past three years a considerable number of experiments have been carried out at the Brown Institution on filter-passing viruses. Many of these, previous to the outbreak of the war, were performed by Dr. C. C. Twort, and, unfortunately, circumstances during the present year have made it difficult to continue the work.

In the first instance attempts were made to demonstrate the presence of non-pathogenic filter-passing viruses. As is well known, in the case of ordinary bacteria for every pathogenic micro-organism discovered many non-pathogenic varieties of the same type have been found in nature, and it seems highly probable that the same rule will be found to hold good in the case of ultra-microscopic viruses. It is difficult, however, to obtain proof of their existence, as pathogenicity is the only evidence we have at the present time of the presence of an ultra-microscopic virus. On the other hand, it seems probable that if non-pathogenic varieties exist in nature these should be more easily cultivated than the pathogenic varieties; accordingly, attempts to cultivate these from such materials as soil, dung, grass, hay, straw, and water from ponds were made on specially prepared media. Several hundred media were tested. It is impossible to describe all these in detail, but generally agar, egg, or serum was used as a basis, and to these varying quantities of certain chemicals or extracts of fungi, seeds, &c., were added. The material to be tested for viruses was covered with water and incubated at 30° C. or over for varying periods of time, then passed through a Berkefeld filter, and the filtrate inoculated on the different media. In these experiments a few ordinary bacteria, especially sporing types, were often found to pass through the filter; but in no case was it possible to obtain a growth of a true filter-passing virus.

Attempts were also made to infect such animals as rabbits and guinea-pigs by inoculating two doses of the filtered material, or by rubbing this into the shaved skin. In other cases inoculations were made directly from one animal to another in the

[1] This investigation was made on behalf of the Local Government Board.

hope of raising the virulence of any filter-passing virus that might be present. All the experiments, however, were negative.

Experiments were also conducted with vaccinia and with distemper of dogs, but in neither of these diseases was it found possible to isolate a bacterium that would reproduce the disease in animals. Some interesting results, however, were obtained with cultivations from glycerinated calf vaccinia. Inoculated agar tubes, after 24 hours at 37° C., often showed watery-looking areas, and in cultures that grew micrococci it was found that some of these colonies could not be subcultured, but if kept they became glassy and transparent. On examination of these glassy areas nothing but minute granules, staining reddish with Giemsa, could be seen. Further experiments showed that if a colony of the white micrococcus that had started to become transparent was plated out instead of being subcultured as a streak then the micrococci grew, and a pure streak culture from certain of these colonies could be obtained. On the other hand, if the plate cultures (made by inoculating the condensation water of a series of tubes and floating this over the surface of the medium) were left, the colonies, especially in the first dilution, soon started to turn transparent, and the micrococci were replaced by fine granules. This action, unlike an ordinary degenerative process, started from the edge of the colonies, and further experiments showed that when a pure culture of the white or the yellow micrococcus isolated from vaccinia is touched with a small portion of one of the glassy colonies, the growth at the point touched soon starts to become transparent or glassy, and this gradually spreads over the whole growth, sometimes killing out all the micrococci and replacing these by fine granules. Experiments showed that the action is more rapid and complete with vigorous-growing young cultures than with old ones, and there is very little action on dead cultures or on young cultures that have been killed by heating to 60° C. Anaerobia does not favour the action. The transparent material when diluted (one in a million) with water or saline was found to pass the finest porcelain filters (Pasteur-Chamberland F. and B. and Doulton White) with ease, and one drop of the filtrate pipetted over an agar tube was sufficient to make that tube unsuitable for the growth of the micrococcus. That is, if the micrococcus was inoculated down the tube as a streak, this would start to grow, but would soon become dotted with transparent points which would rapidly extend over the whole growth. The number of points from which this starts depends upon the dilution of the transparent material, and in some cases it is so active that the growth is stopped and turned transparent almost directly it starts. This condition or disease of the micrococcus when transmitted to pure cultures of the micrococcus can be conveyed to fresh cultures for an indefinite number of generations; but the transparent material will not grow by itself on any medium. If in an infected tube small areas of micrococci are left, and this usually happens when the micrococcus has grown well before becoming infected, these areas will start to grow again and extend over the transparent portions, which shows that the action of the transparent material is stopped or hindered in an overgrown tube; but it is not dead, for if a minute portion is transferred to another young culture of the micrococcus it soon starts to dissolve up the micrococci again. Although the transparent material shows no evidence of growth when placed on a fresh agar tube without micrococci it will retain its powers of activity for over six months. It also retains its activity when made into an emulsion and heated to 52° C., but when heated to 60° C. for an hour it appears to be destroyed. It has some action, but very much less, on staphylococcus aureus and albus isolated from boils of man, and it appears to have no action on members of the coli group or on streptococci, tubercle bacilli, yeasts, &c. The transparent material was inoculated into various animals and was rubbed into the scratched skin of guinea-pigs, rabbits, a calf, a monkey, and a man; but all the results were negative.

From these results it is difficult to draw definite conclusions. In the first place, we do not know for certain the nature of an ultra-microscopic virus. It may be a minute bacterium that will only grow on living material, or it may be a tiny amœba which, like ordinary amœbæ, thrives on living micro-organisms. On the other hand, it must be remembered that if the living organic world has been slowly built up in accordance with the theories of evolution, then an amœba and a bacterium must be recognised as highly developed organisms in comparison with much more primitive forms which once existed, and probably still exist at the present day. It is quite possible that an ultra-microscopic virus belongs somewhere in this vast field of life more lowly organised than the bacterium or amœba. It may be living protoplasm that forms no definite individuals, or an enzyme with power of growth.

In the vaccinia experiments described above it is clear that the transparent material contains an enzyme, and it is destroyed at 60° C. It also increases in quantity when placed on an agar tube containing micrococci obtained from vaccinia, and this can be carried on indefinitely from generation to generation. If it is part of the micrococcus it must be either a stage in its life-history which will not grow on ordinary media but stimulates fresh cultures of the micrococcus to pass into the same stage, or an enzyme secreted by the micrococcus which leads to its own destruction and the production of more enzyme. The fact that the transparent portion cannot be grown except on the micrococcus makes it impossible to obtain any definite evidence on these points. There is this, however, against the idea of a separate form of life: if the white micrococcus is repeatedly plated out and a pure culture obtained, this may give a good white growth for months when subcultured at intervals on fresh tubes; eventually, however, most pure strains show a transparent spot, and from this the transparent material can be obtained once again. Of course, it may be that the micrococcus was never quite free from the transparent portion, or this may have passed through the cotton-wool plug and contaminated the micrococcus, but it seems much more probable that the material was produced by the micrococcus. Incidentally, this apparent spontaneous production of a self-destroying material which when started increases in quantity might be of interest in connexion with cancers. In any case, whatever explanation is accepted, the possibility of its being an ultra-microscopic virus has not been *definitely* disproved, because we do not know for certain the nature of such a virus. If the transparent portion were a separate virus, it might be vaccinia or it might be some contaminating non-pathogenic ultra-microscopic virus, for it is conceivable that whereas a non-pathogenic variety might grow on micrococci or bacilli, a pathogenic variety might grow only on the animal it infects.

As the animal experiments were negative there is no evidence that it is vaccinia, although such a virus might lose its virulence when grown outside the body. On the other hand, no evidence was obtained that it was a non-pathogenic contaminating ultra-microscopic virus. On the whole it seems probable, though by no means certain, that the active transparent material is produced by the micrococcus, and since it leads to its own destruction and can be transmitted to fresh healthy cultures, it might almost be considered as an acute infectious disease of micrococci.

In view of the results obtained with vaccinia similar experiments were carried out with other material. It will not be necessary to describe all these in detail; it will suffice to note that similar, though not such definite, results were obtained with a micrococcus and a member of the coli-typhoid group of bacilli which were obtained from the intestinal mucous membrane of a dog suffering from acute distemper, and there is some evidence that the difficulty often experienced in isolating certain known pathogenic micro-organisms may be due to the same cause. Experiments carried out with tuberculous pleural fluids and tubercle bacilli gave negative results.

More recently, that is when the investigation of infantile diarrhœa and vomiting was continued during the summer and autumn of this year (1915), similar experiments were carried out with material obtained from the intestinal tract. The general results of this investigation will be published later, and it will be sufficient here to note that after certain difficulties had been overcome it was found that in the upper third of the intestine, which contained numerous bacilli of the typhoid-coli group, some larger bacilli were also present. In some cases they grew in far larger numbers than the coli types of bacteria; but this was only so when precautions were taken to eliminate the action of a dissolving substance which infected the colonies so rapidly that they were dissolved before attaining a size visible to the eye. Here, then, is a similar condition to that found in vaccinia, and the greatest difficulty was experienced in obtaining the bacilli free from the transparent dissolving material, so rapidly was the infection increased and carried from one colony to another. Finally, cultures were obtained by growing the bacilli with certain members of the typhoid-coli group for a few generations and then plating out. From the colonies cultures were obtained on ordinary agar. Some of these cultures being slightly infected with the dissolving material rapidly became transparent and were lost, while a few grew well. The bacillus has several curious characters, and these are now being investigated. It is in no way related to the typhoid-coli group. The relation of this bacillus and the dissolving material to infantile diarrhœa has not yet been determined, but probably it will be found also in cases of dysentery and allied conditions; and I greatly regret that I have not been afforded an opportunity of investigating the dysenteric conditions in the Dardenelles to determine this and other points.

When possible, experiments should be conducted to determine the relative toxicity of cocci and bacilli when free from and when associated with the dissolving material, and vaccines prepared with the transparent material should be tested.

I regret that financial considerations have prevented my carrying these researches to a definite conclusion, but I have indicated the lines along which others more fortunately situated can proceed.

A METHOD OF DROP-MEASURING LIQUIDS AND SUSPENSIONS.

(ROUGH PRACTICAL NOTES.)

BY R. DONALD, B.SC. N.Z., D.P.H. OXF.

(*From the London Hospital Bacteriological Laboratory*—Professor W. BULLOCH, M.D. ABERD., F.R.S.)

THE following article gives some practical details of a method of measuring liquids in uniform drops of any standard size by means of ordinary simply drawn pipettes quickly and accurately gauged. The apparatus was briefly described in a communication[1] to the Royal Society in 1913. A more detailed account was in preparation when the outbreak of war restricted work to the immediately necessary. Under this category the method has been brought by the increased importance of an accurately quantitative form of the Widal test.

For the measurement of small quantities graduated pipettes have hitherto been largely used. But although the worker proposes to deliver small volumes from a graduated capillary pipette, and although he displaces the liquid column downwards the exact amount proposed, yet nature disposes the outflowing liquid in drops, and if the last portion expelled does not form a complete drop and fall spontaneously at its complete phase, then the incomplete drop clings with more or less pertinacity to the pipette point. Even when allowable, touching the pipette point on the side of the receiving vessel is uncertain[2] in its result.[3]

But just in the very conditions where surface-tension or capillary force hinders us most if we try to work independently of it, there that very force will act in our favour if we give it play. Thus, if instead of difficultly and uncertainly aborting drops at various phases of their formation-cycle we allow the rhythmic delivery of mature drops, those drops will be of wonderfully uniform size. Indeed, for delivering successive small equal quantities drop-measuring is superior in quickness and in accuracy to any other method. And the smaller the quantities concerned the greater is this superiority. This may have been more or less generally admitted. The problem hitherto has been how to get easily a supply of reliably uniform pipettes. That problem may be solved very simply.[4]

[1] Also various published articles have dealt with some applications of the method:—Donald, R.: Proceedings of the Royal Society, B, vol. lxxxvi., 1913, pp. 198-202. Idem: A Comparison (of two Wassermann methods), THE LANCET, June 29th, 1912, p. 1752. Idem: A Method of Counting Bacteria in Water, THE LANCET, May 24th, 1913, p. 1447. McIntosh, J., and Fildes, P.: The Wassermann Reaction and its Applications to Neurology, Brain, vol. xxxvi., November, 1913, pp. 213, 227. Benians, T. H. C.: The Resistance of Various Bacteria to the Disinfecting Action of Toluol, &c., Zeitschrift für Chemotherapie, Or., ii., 1913, p. 32. Donald, R.: A Method of Estimating Numerically and Qualitatively the Cells in Permanent Preparations of Cerebro-spinal Fluid, Folia Hæmatologica, Or., B, vol. xvii., 1913, pp. 139-166. Harrison, Major L. W.: Wassermann Test, Technique, Journal of the Royal Army Medical Corps, vol. xxii., 1914, p. 615. Donald, R.: Drop-methods of Counting the Cells of Cerebro-spinal Fluid; the Relation of the Cell-count to the Wassermann Reaction, Review of Neurology and Psychiatry, August, 1914, pp. 333-369. Head, H., and Fearnsides, E. G.: The Clinical Aspects of Syphilis of the Nervous System in the Light of the Wassermann Reaction and Treatment with Neosalvarsan, Brain, vol. xxxvii., September, 1914, p. 2.

[2] Even greater uncertainty may be found in (1) the amount removed and in (2) the amount delivered as a "loopful" of liquid taken by a platinum loop—usually for qualitative but sometimes for quantitative purposes.

[3] For *delivering* an exact amount of a non-wetting liquid such as mercury, Wright's ingenious capillary 5 c.mm. pipette is excellent. Also for measuring, without delivering, the quantity *contained*, capillary pipettes, such as Wright's multiple dilution pipettes and the hæmocytometer pipette, have not the weakness above mentioned. Even this weakness may be overcome by the skill and care of an experienced worker. As regards the hæmocytometer pipette, its 101 mark ought to be not below the narrowest point of the narrowed bore, as the upper meniscus is vigorously drawn up to that point by capillary force.

[4] This brief article omits the extensive bibliography of the subject and the discussion of the physical phenomena connected with drop-formation.