apprehension of the future often return to hospital in a more neurasthenic state than when they went. I have noticed also a number of cases amongst officers and men who have come to our hospital, in which there has been a mental conflict and preoccupation regarding their honour and reputation as soldiers, and having no visible evidence to justify their return from active service, they are apprehensive lest their comrades should regard them as shirkers. If they had got a wound which disabled them there would be something to show, and they would have no fear or apprehension regarding estimation of their honour or justification for being invalided home. Owing to their neurasthenic condition they are unable to see things in their proper light.

Again, financial worry, domestic difficulties, and other fears of their future may add to the factors of mental preoccupation and neural exhaustion and hinder recovery. In respect to special organs being affected, the cardiac functions are most often disarranged; indeed, the majority of cases of what is called D.A.H. (disordered action of the heart) should be termed neurasthenia, for the knowledge of a neurotic individual that the diagnosis has been made of disordered action of the heart is liable to set up an obsessional preoccupation, causing præcordial anxiety, palpitation, and rapid action, which, reacting, intensifies the emotional disturbance and excites many of the distressing symptoms of this neurosis.

MANIFESTATIONS OF EFFECTS ON THE ENDOCRINE GLANDS.

Owing to limitation of time I cannot do more than briefly allude to the manifestations of effects on the endocrine glands in certain individuals. Many of the cases of neurasthenia have exhibited definite evidence of affection of the endocrine glands by the fact of increased blood pressure; rapid-acting heart; enlargement of the thyroid; von Graefe's sign; Moebius' sign and fine tremor. According to Crile, there is normally an inter-relation of the thyroid, the adrenals, and the brain, which constitutes the master-key to the automatic action of the body.

CONCLUSION.

In conclusion, while admitting that, from commotion or prolonged stress of war, a stable nervous organisation may develop an acquired emotivity; nevertheless, it seems that the psychogenic or auto-suggestive factor, acting upon a pre-war emotive brain, is the chief determining cause of war psycho-neurosis, and the neurotic patient's protest of anxiety lest he be boarded out of the service, or not allowed to go to the front, is really in many cases an indication of an exhausting conflict which has been going on continually in his mind between the self-conservative instinct and the moral obligation of duty and patriotism, which has its roots in the social herd instinct.

New Methods

FOR

THE STUDY OF EMIGRATION AND OF THE BACTERICIDAL EFFECTS EXERTED IN THE WOUND BY LEUCOCYTES.

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SOME ten years ago attention was drawn by Ponder¹ to the fact that when blood is allowed to clot in a cell built up with plasticine upon a microscopic slide the leucocytes emigrate and attach themselves to the glass. Later Emery observed that when a capillary pipette filled with blood is placed in the incubator the leucocytes come out of the clot and attach themselves to the walls. And I have shown² that when we centrifuge blood in "emigration tubes" the leucocytes make their way out from among the red corpuscles and climb up into the trellis of fibrin formed in the supernatant plasma. I have spoken of this emigration—which is clearly independent of chemiotaxis—as *eleuthero-tropism* or impulsion to free wandering. But clearly we might in these cases also conceive of the leucocyte as having an impulse to get into contact with, and attach itself to, any foreign body.

Our particular problems here, however, are problems not of "epistemic" but of "pragmatic science"—problems of how to go to work to sterilise war wounds by leucocytic agency.

Our first task must be to find out how to promote emigration from the blood. We want for this, in the first place, a technique for operating with equal volumes of blood disposed upon equal areas of glass surface. Shallow cells of fixed area for the reception of blood can be made by two methods. 1. We can paint very hot paraffin wax on to a moist glass slide; punch circles with a cork-cutter into the paraffin while it is still plastic, and prise out the discs when the paraffin has set hard (Fig. 4), afterwards polishing up the floor of the cells with a drop of xylol applied on a clean handkerchief. 2. If we want standard volumes of blood imposed on a standard area we provide ourselves with some squared paper—and the squares may conveniently measure 5 mm. each way; we cut up a strip of this paper into separate squares, letting these fall as we cut them upon a wet glass slide. Then draining off the excess of water we range the squares in a line, dry the slide with blotting-paper, and then press the paper squares down firmly upon its surface. In the meantime we have melted some paraffin, heating it till it

just commences to smoke. Then, applying this with a rapid stroke of a large camel's hair brush, we cover up our paper squares. When the paraffin is firmly set we turn up the

FIG. 1.

edge of our squares with the point of a knife and remove them, exposing the clear underlying surface of glass (Fig. 1 A.)

¹ THE LANCET, Dec. 12th, 1903, p. 1746.

² THE LANCET, 1915, i., 787.

FIG. 2.

FIG. 3.

We have now on our slide a row of paraffin cells, each with a superficies of 25 square millimetres. This done, we take an ordinary capillary pipette, calibrate it with mercury delivered from "an automatic pipette,"³ and then, drawing blood from the finger, impose upon each cell 25 c.mm. of blood, carefully leading that blood into all the four corners of the square. (Fig. 1 B.) Our slide is then placed in an incubator in a warm, moist chamber made by flooring a Petri dish with three or four folds of very wet filter paper. After 20 to 40 minutes the slide is withdrawn, the clots are gently removed, the escaped red blood corpuscles are washed

FIG. 4.

off from the floor of the cells with warm salt solution, or the preparation is simply held under the tap. This done, we fix with corrosive sublimate and stain with methylene blue.

The floor of the cell seen under a hand lens then presents the appearance shown in Fig. 2; and under the microscope we see the very interesting moving forms depicted in Fig. 3. To see these at their best we must catch the leucocytes at the stage of most active emigration; we must avoid washing in cold water; and the fixative must be applied to our specimen as quickly as possible

METHODS FOR STUDYING THE EFFECT OF CHEMICAL REAGENTS IN THE BLOOD UPON THE EMIGRATION OF LEUCOCYTES.

In studying the effect upon leucocytic emigration of chemical agents added to the blood we shall do well to employ the "method of washes." We fill our capillary pipette with blood up to the fiducial mark upon the stem; we then draw in an air-bubble; then fill up to the fiducial mark with our reagent; expel this; and then, by sucking in and blowing out, mix our blood with the wash left upon the walls. Such a wash will correspond to about 1/25th of the unit volume of the fluid. When less than a wash of our solution is to be added, we make graduated dilutions; and by operating with these obtain corresponding fractions of the original wash. The results obtained by adding to 25 c.mm. of blood two washes, one wash, and a series of fractions of a wash of N/3—i.e., approximately 5 per cent. solution of sodium carbonate—are shown in Fig. 1 C. It will be seen that there was obtained with 1/8th wash of our

solution (i.e., by the addition of 1/8th of 1/25th of a 1/20 solution = 1 in 4000 of carbonate of soda added) an emigration very much greater than that obtained in the control blood. This result is obtained with quantities of sodium carbonate ranging down to 1 to 6000 to 1 in 8000, and the result is, as experiments by my colleague, Captain L. Colebrook, have shown, obtained also when we inject intravenously in man.

With 1/6th of a wash of saturated (i.e., 36 per cent.) sodium chloride solution (i.e., by the addition of 1/6th of 1/25th of 36/100 = a 36/15,000 or 0.24 per cent. solution) we obtain also a very striking increase of emigration.

METHODS FOR STUDYING THE CHIMOTACTIC EFFECTS EXERTED BY BACTERIAL CULTURES.

For investigating the chimiotactic effects exerted by microbes we take our paraffin cells, and impose upon them graduated dilutions of bacterial culture, or, as the case may be, of a vaccine, spreading out in each case 10 c.mm. upon the 25 square millimetres of glass surface. We then evaporate to dryness in the incubator. Upon the cells thus prepared we impose our 25 c.mm. of blood, and proceed as before. A typical series of results are shown in Fig. 1 D. We have here, beginning at the left hand of the specimen, an implantation of 2 million streptococci per square millimetre

FIG. 5.

of surface; and then towards the right progressively smaller implantations; and on the extreme right, as controls, unimplanted cells. In the left hand cell we have no emigration; we have then from left to right more and more emigration; until, in the cell implanted with 10,000 streptococci per square millimetre, we achieve emigration equal to the controls; and in the cell implanted with 3300 streptococci per square millimetre, augmented emigration

³ Vide the author's "Technique of the Teat and the Capillary Glass Tube," Constable, London.

Fig. 1E shows a method of procedure in some respects better—the method of subdivided cells. Here before constructing our paraffin cells we have drawn a line with a glass-cutting knife longitudinally down the middle of the slide, and have pencilled in this line with a coloured grease pencil. Upon this we range our paper

FIG. 6.

squares, disposing them so that exactly bisect them. The the glass floors of our paraffin-way divided into two panes by a confine fluid. Upon the upper panes we now impose in series graduated dilutions of bacterial culture or vaccine, keeping the lower panes clear. We use, of course, for the implantation of our half-squares only 5 c.mm. of fluid, and when we have dried off in the incubator we fill into each cell, as before, 25 c.mm. of blood. Besides providing us with better positioned controls, this method brings into view

the line shall windows, i.e., cells, are in this line which will

FIG. 7.

“negative” and “positive induction effects.” In the cell on the extreme left, emigration is restrained not only over the implanted half-square, but also over the whole of the other half. In the next cell we have again a negative induction effect, but the restraining influence extends only a little distance into the lower half-square. Further along the line of cells, we have what we may call “the reaction of indifference”—i.e., emigration is equal on the implanted and unimplanted half-squares. And, finally, in the upper half-square of the fifth cell we have positive induction. That is, leucocytes which would in the ordinary course have emigrated on to the unimplanted half-square have here diverted to the implanted half-square.

It will be plain to consideration that we have here a method for procuring information with regard to the chemiotactic quality—i.e., toxicity—of vaccines; and the chemiotactic response—i.e., the attacking power—of a patient's leucocytes.

The “method of subdivided cells” points the way to another development—the *Method of Ratlines*. Here we rule upon a microscope slide with a glass writing pencil a system of parallel longitudinal lines; and then intersect these with vertical lines in such a way as to form a wide centre ladder-way, flanked on either side by a narrower one. We now impose upon the compartments of the central ladder-way from above downwards progressive dilutions of a vaccine or bacterial culture—sucking these off with a capillary pipette (drawn out into a filiform end) so as to leave behind only a wash of the fluid. This done, we divide the centre ladder-way into two narrower

ladder-ways. We use for this purpose a dissepiment made by rolling out a small pellet of plasticine between a microscope slide and the laboratory bench. Finally, we bridge over the double ratline thus obtained, converting it into a double cell. We use as the body of our bridge a large cover-glass or a piece of glass slide; we support it on either side upon a *thick* cover-glass fastened down with vaseline; and we use as our central support and partition our roll of plasticine. One of the two compartments we now fill in with the patient's blood; and the other with our own to serve as a control (Fig. 6), and then we incubate in a warm, moist chamber for from $\frac{1}{2}$ to $\frac{3}{4}$ hour.

In Fig. 7 we have a comparison instituted by this method. It is a comparison between the author's blood as a control, and the blood of a colleague who had received an inoculation of staphylococcus vaccine for the treatment of boils. Upon the left is the blood of the patient, upon the right the control blood; and in each case, as explained above, we have upon the external ladder-ways the emigration on the unimplanted compartments; and upon the internal ladder-ways the emigration on the implanted compartments—the bacterial implantation being lighter as we proceed from above downwards. It will be seen, in connexion with the normal blood upon the right, that we have an inhibition of emigration into the more heavily implanted compartments, and a negative induction effect in the adjoining unimplanted compartments. On the left in the patient's blood, we have positive chemiotaxis into the upper more heavily implanted compartments. Further, we see on comparing the two

FIG. 8.

external unimplanted stairways, that there is more emigration from the patient's than from the control blood. This stands in relation with the circumstance that the patient had a leucocytic count of 10,900 per c.mm.

In Fig. 8 we have a comparison between the blood of a wounded man suffering from heavy staphylococcus septicæmia and the blood of a healthy laboratory worker. The patient's blood is on the right, the control blood on the left—the disposition being in all other respects the same as in the last case. It will be seen at a glance that we have

FIG. 9.

enormously greater emigration from the patient's blood. The leucocytes of his blood counted 20,000 per c.mm. Further, we see when we compare the emigration of the patient's blood into the implanted compartments with the emigration of the normal blood into the corresponding implanted compartments, that the patient's leucocytes have confronted a much heavier implantation.

Again, another procedure for the study of emigration is illustrated in Fig. 5. We have here upon a microscope slide a series of paraffin islands, made by conveying small loopfuls of melted paraffin to the slide and then

melting. Round each of the islands is a circle—imprinted from paraffined paper with a cork-cutter—and disposed after the fashion of an atoll. The atolls of islands 1, 2, 4, and 5 are implanted by the same technique as in the case of the ratlines with a wash of a suspension of microbes—the thick suspension employed being, of course,

FIG. 10.

progressively diluted from 1 to 5. The atoll of island No. 3 is left unimplanted to serve as a control. The marginal area of the slide is then brushed over with melted paraffin, leaving the central part free. This is then flooded with blood—this coming right up to but not overflowing the paraffin islands.

The pattern of emigration obtained is shown in Fig. 5 B. All that area of the slide which was flooded with blood and lies outside the atolls is covered in with an even deposit of leucocytes. In the interior of the atolls the conditions are in each case different. In the first the glass is completely bare of leucocytes; in the second we have a narrow zone round the island completely bare and the rest covered sparsely with leucocytes. In the third—i.e., in the control—atoll, we have a similar zone of bare glass immediately round the island; and, in the outlying portions of the atoll, the same amount of emigration as outside. In the fourth, we have in the atoll rather more leucocytes than outside, but the narrow zone immediately round the island is still visible. In the fifth, we have a heaping up of leucocytes, and these come right up to the shore of the island.

The new fact brought into prominence by this method is that emigration is checked wherever the blood comes into contact with the air. We have here, it would appear, not inhibition caused by the exposure to oxygen, but inhibition caused by desiccation. For when we repeat our experiment under anaerobic conditions we obtain the same amount of marginal inhibition round the paraffin islands. The same zone of marginal inhibition is seen also when we impose blood upon paraffin cells, and fail to make proper provision against desiccation. And marginal inhibition is always more pronounced when we make to the blood any addition which brings up its salt content. The practical lesson we learn from these experiments is that the emigration of leucocytes from wound surfaces will be inhibited as soon as these begin to get dry.

METHODS FOR STUDYING THE CHIMIOTACTIC EFFECTS EXERTED BY SOLUBLE SALTS.

For the study of chimiotactic effects exerted upon leucocytes by chemical agents which are readily

soluble, other methods than those described above will be required. When working with readily soluble chemical agents we must guard against direct mechanical admixture with the blood, and must arrange for a gradual diffusion of the agent into the clot. We can achieve this by using agar as an excipient, and filling our blood into cells cut out in tablets of this agar.

A convenient procedure is the following: We mix our reagent with a 2 per cent. water agar, and then cast this agar, into tablets of standard thickness, in moulds constructed out of a pair of microscopic slides held apart at the ends by pieces of slide measuring 1, 1.5, or 2 mm. in thickness. We push the slide which forms the lid of this mould a little away from us, pour in our agar, and then bring the lid back into position. When the agar has set we withdraw the lateral supports and prise off the lid. We now, using for this purpose a cork-cutter or a piece of very thin-walled glass-tubing, cut circles in this agar and remove the included discs. Into the cells thus obtained we fill in our blood.

In Fig. 10 are shown emigration patterns obtained upon the glass floor of cells 7 mm. in diameter cut out in agar containing 2.8, 1.4, 0.7, and 0.35 per cent. of NaCl,

FIG. 11.

and water agar respectively. It will be seen in Fig. 10 A that with 2.8 per cent. salt agar we have sparse emigration, and emigration confined to the central area of the cell. With the 1.4 per cent. salt (Fig. 10 B) we have much more emigration, and emigration over the whole floor of the cell. With 0.7 per cent. salt (Fig. 10 C) we have still heavier emigration, and here the leucocytes are lined up in the form of a concentration ring round the margin of the cell. With 0.35 per cent. of salt agar (Fig. 10 D) and water agar (Fig. 10 E) we have again less emigration.

In Fig. 10 F we have an emigration pattern similar to that in C, but differing in that there is much more marked concentration of leucocytes round the margin. This pattern was obtained by using an agar cell impregnated with 1 per cent. sodium chloride and 0.1 per cent. sodium carbonate. And it is to be remarked in this connexion that the time factor, the temperature of the cell when the blood is filled in, the diameter and depth of the cell, and no doubt the density of the agar will all influence the pattern of emigration obtained. To obtain constant results we must keep to

FIG. 12.

a definite time limit, must warm up the cell beforehand in the incubator, and must work with a cell of standard dimensions made out of the same agar.

We can vary our procedure by conducting our experiments in "emigration boxes" (Fig. 9). We build these up out of three elements—a slab of agar, 1 mm. thick, for the floor; a middle, 2 mm. thick, perforated agar or paraffin slab for the body of the box; and a 1 mm. thick slab of agar—or, as the case may be, a piece of glass slide—for a lid. For filling in with blood we dispose the lid as shown in the figure, and as we fill in gradually sliding it over the mouth of the cell.

It will be found, on taking to pieces such a cell and examining the slabs of agar under the microscope, that leucocytes emigrate after incubation quite freely on to the surface of agar—provided, of course, that this contains an appropriate quantity of salt. By employing for the roof and floor of our emigration box slabs of agar with different salt concentrations we can find out which is the more congenial to leucocytes. The naked-eye appearances are, however, deceptive, for emigrated leucocytes very easily come from an agar surface, and there remains behind a film of fibrin which shows up to naked-eye observation.

We pass now from methods which have reference only to the emigration of leucocytes to methods for measuring the bactericidal effects exerted by the emigrated leucocytes. Here two points must be kept clearly in view. First, that—to avoid confusing results due to a bactericidal action of the serum with results due to the bactericidal action of the leucocytes—we must go to work with definitely serophytic bacteria, such, for instance, as streptococci and staphylococci. Secondly, we must make provision for the effacement of all dead spaces—i.e., all hollow spaces in which microbes would be safely out of reach of the leucocytes. It will be obvious with respect to this last requirement that it can be fulfilled only by disposing our leucocytes and microbes upon apposed flat surfaces. One method will be to impose a slab of implanted nutrient agar upon a glass surface covered with living leucocytes. The other—and this more closely imitates the bringing together of the surfaces of a wound—is to take two surfaces covered with living leucocytes, one of glass and the other of nutrient agar; and then, having infected both, to bring the surfaces together so as to combat the infection by leucocytic attack from the two sides.

If we employ the former method, we fill our blood into open agar or paraffin cells; incubate; and then, after lifting off the framework of the cell and then the blood clot, impose upon the leucocyte-covered floor and surrounding surface a slab of agar implanted with microbes. We employ, of course, a series of such agar slabs implanted with successive dilutions of our culture. We then, after ringing round the edges of our slabs with melted vaseline or paraffin, and covering in with a slide, incubate over-night, and compare the number of colonies which come up in the central and peripheral areas of the field. A typical result is shown in Fig. 11.

Where two layers of leucocytes are to be brought to bear upon the bacterial infection we collect our leucocytes upon the glass floors and agar lids of emigration boxes; or we use in pairs open boxes floored the one with glass and the other with agar. In either case before inverting our agar slabs upon our glass slides—leucocyte-covered on leucocyte-covered area, and bare upon bare area—we wash off red blood corpuscles and make our microbial implantation by irrigation with serum to which we have made graduated additions of microbes.

Instead of using, as just described, the outlying portions of our field to serve as controls we can, as in Fig. 12, use separate agar slabs.

Finally, attention may be drawn to an all-important principle to which all the foregoing leads up. When the surgeon seeks information from the bacteriologist as to whether the wound is making progress in sterilisation, and whether secondary suture may or may not be undertaken, the bacteriologist is wont to take the pus and plant it out in such a manner as to secure the survival of the microbes and the destruction of the leucocytes. In other words, the bacteriologist is wont to make what I may call a "*neuro-pyo-culture*." And then the surgeon, when he undertakes the secondary suture, very often obtains a result much more favourable than the findings of the bacteriologist would appear to warrant.

If he wants to be a help to the surgeon, the bacteriologist should change his procedure. Instead of making films and *neuro-pyo-cultures*, and reporting the number of microbes seen or rescued alive from the pus; he should make *bio-pyo-cultures*—i.e., provide in his cultures conditions favourable to phagocytosis, and should then report whether the leucocytes on the wound surface—assuming that the surgeon gives them a fair chance by effacing dead spaces and surface desiccation—are competent to get the better of the remaining bacterial infection.

In conclusion, I have to express my indebtedness to my fellow-workers—Captain Alexander Fleming and Captain Leonard Colebrook—for much valuable assistance in the working out of these methods, and to Sergeant A. K. Maxwell for much help in preparing the illustrations.

A METHOD OF SPLINTING GUNSHOT FRACTURES.

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THE following series of apparatus for the treatment of fractures of war have been used by us for some considerable time, and the results obtained by their use have been so satisfactory that we now publish them in the hope that others may find them of value.

The various splints have been made in the ordinary workshop of a general hospital from bars of aluminium which are supplied from base medical stores.

The principles adopted have been those of Thomas and Jones, and a few of the splints are modifications which we have found advantageous of the Jones's models. In constructing them we have aimed at the following points:—

1. Wide and easy access to the wound and fracture area.
2. Continuous immobilisation of the fracture area.
3. A splint enabling the patient to be up and about whenever his general condition permits.
4. A splint by means of which the alignment will be such as to give the best functional and anatomical result, and in several cases a starting-point for after-movements.

Several fractures necessitate the limb being in the abducted position, and to obtain this in an ambulatory type of splint some form of body grip is necessary from which to build up the support for the arm. The one we have used consists of an aluminium bar shaped round the shoulder of the affected side and coming down to the anterior superior spine of the opposite side in front and to a corresponding point behind. The ends are then joined to a semi-girdle round the same side by means of rivets. Besides being the basis for construction this grip forms the medium for counter-extension.

A short description of the method of construction and application is given under each splint. In their application it is, of course, necessary for any bar coming in contact with the body to be well padded.

Upper Arm Splint for High Open Fractures of the Humerus.

A hoop aluminium is fitted closely over the wounded shoulder, lying into the side of the neck, and coming down to the opposite anterior superior spine in front, and it is carried down to a corresponding point behind. The free ends of the loop are joined together by a semi-circular band encircling the same side and gripping it just above the iliac crest. This constitutes the "counter-extension grip," and the rest of the apparatus is built up from it. From the points of the greatest convexity of this grip (the upper arm being carried out from the body) bars are carried to a point about 4 inches beyond the elbow tip, and there joined by a vertical upright. To this out-structure a forearm rest is built, as in the accompanying diagram, and supported by a stay to the medial end of the abdominal hoop. (Fig. 1.)

Application.—The forearm, by means of supports and bandages, is firmly fixed in the forearm rest, which acts as a fulcrum, against which extension acts. This is applied (1) directly from the lower end of the shaft of the humerus by adhesive, &c.; or (2) from the upper third of the forearm by suitable methods. The prolongation beyond the elbow constitutes the end of the splint upon which the pull is always taken. It will here be observed that the whole of the shoulder—the axilla—and the upper of the chest wall are absolutely exposed and unencumbered in any way. Room is allowed for placing counter-incisions in those places where, with this injury, pocketing and tracking of pus usually occur—i.e., in the axilla, down the side of the chest wall, or at the posterior border of the deltoid.

Apparatus for Open Fractures of the Lower and Middle Thirds of the Humerus (Modification of Jones's Arm Splint).

This splint is used only in cases where there are no wounds in the axilla or around the shoulder. It differs from the Jones's splint in the following respects: (1) The bars coming from the axillary counter-extension grip are placed anterior and posterior relative to the upper arm and not medial and lateral; (2) the upper arm bars are placed about

HOSPITAL SUNDAY FUND AT PLYMOUTH.—Oct. 21st, 1917, was observed in the "Three Towns" as Hospital Sunday, and as a result the sum of £590 has been handed to the South Devon and East Cornwall Hospital, Plymouth.