

for the M

AN EXPERIMENTAL AND CRITICAL RESEARCH WITH REGARD
to

- 1, The bacterial significance of the skin glands and their secretions in the practice of aseptic surgery;
- 2, The circumstances determining the incidence and duration on the skin of germs other than the permanent epiphytes;
- 3, The nature of the permanent skin epiphytes from the surgical aspect;
- 4, The value of lanoline employed as a protection against infection of the skin of the operator;
- 5, The microscopical evidence as to the localisation of bacteria on the skin under varying circumstances,

by

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SECTION I.

The significance of the skin glands and their secretions in the practice of aseptic surgery.

Since the days when Listerism was first inaugurated the importance of the disinfection of the skin both of operator and of patient has always been prominent, and of all the problems involved in the practice of antiseptics this has proved the most troublesome of solution. It cannot in fact be said to be yet settled, if by disinfection of the skin is to be understood a production of a condition of absolute sterility in the bacteriologists' sense of the word. That a degree of sterility which allows of the primary union of the operation wound may be attained is of course a matter of every day experience, but the fact that, whatever method of disinfection be chosen, it is impossible for the surgeon to predict with certainty that he cannot infect the wound with his hands, is a state of affairs which it has been the attempt of innumerable workers to remedy. Nevertheless even in the many years that have now passed since antiseptic methods have been studied and carried out, no process of skin disinfection of mechanical or chemical nature, or of both combined, has yet been devised which by common consent is acknowledged as producing an absolute sterility. Numerous processes may and do attain to producing an almost entire absence of germs as far as can be demonstrated, but the bacteriologist may claim that so long as a single organism is still to be found in an active state the ideal result has not been arrived at. The surgeon may well perhaps question whether such an ideal is at all necessary or of importance in view of the probably small pathogenic activity of such germs as may ordinarily remain. The frequent possibility of the infection of the surgeon's skin with virulent microbes however is sufficient reason for aiming at the bacteriological ideal.

On consideration of the causes of this paramount difficulty in disinfection the reason is not far to seek. The skin presents a most irregular surface with countless

macroscopic and microscopic crevices and openings. Its surface is still further roughened by accidental abrasions or by the natural process of the casting off of epithelial scales as is constantly taking place, all of which afford to the minute microbe a comparatively enormous resting-place in which to lie and possibly reproduce itself.

If the presence of germs were quite superficial a thorough disinfection ought not be impossible; the fact that they must lie deeply and firmly imbedded in the skin however, has long been recognised as the main reason for the fallacies of a skin-disinfection. Moreover in recent years the belief has gained acceptance that the natural openings of the skin, the sweat-glands and hair-follicles furnish a ready nidus for the deposit of bacteria which, as might be expected, in such a situation are unaffected by antiseptics even of penetrating properties. This view is generally approved and mentioned by most recent writers on the subject, among others von Mickulicz 1), 2), Opitz 3), Schumacher 4), Kocher 5), Nagelschmidt 6), Döderlein 7), Schleich 8), 9). These authors, many of them foremost authorities on hand-disinfection, refer as a fact to the impossibility of a complete disinfection of the skin on account of the deeply seated position of skin germs in the sweat-glands and frequent mention is made of the likelihood of these germs again reaching the surface.

V. Mickulicz and Kausch 1) for instance in a recent textbook state that "the reason for the difficulty of sterilising the skin is on account of its containing, in the deeper layers of the epidermis and in the efferent ducts of the glands, bacteria which appear to be regular epiphytes (staph. albus). Disinfectant methods remove or kill the germs of at most the superficial layers. During the operation the bacteria of the deeper layers soon come to the surface. The skin is consequently in the most favourable cases only superficially and temporarily

sterile." V. Mickulicz had already in 1897 given full expression to this view 2). After remarking that "no one will expect a genuine disinfection of the hand through our present agencies who knows how deep in the gland ducts ... the regular epiphytes of the skin lie", he continues, "On a hand which as that of the surgeon is so often infected with virulent bacteria, into whose pores through rubbing and pressing manipulations, the germ-containing secretions are driven, the bacteria may often for a long time remain so fast that they resist the best methods of disinfection. Such a hand may be proved by cultures to be almost germ-free on its surface; in the course of a prolonged operation through pressing and rubbing, in a manner massaging, manipulations the concealed germs are again urged to the surface and so can produce an infection".

Schleich 8), mentions that Mickulicz and his pupils have pointed out how the bacteria which on every skin find hiding-places in the glandular apparatus cannot be got at. Döderlein 6) is even more explicit. He speaks of the "sebaceous and sweat glands whose canals give entrance to bacteria. They settle down in these ungetatable spaces, probably also increase in the recesses, defy disinfectants and in inoculation experiments are carried over to the culture medium in greater or less number according to the condition of the skin, or else remain behind in the skin sooner or later, depending on the external conditions, to again reach the surface: it is these skin-parasites which are deposited at operations in great number by the uncovered hands in the wound where we can always readily demonstrate them". He considers these skin-parasites however to be usually non-pathogenic, while virulent organisms accidentally brought to the skin do not settle down as constant parasites.

I might quote many others giving expression to similar opinions. Sarwey and Paul 10), whose work on hand-disin-

fection has been one of the most complete and comprehensive, accept this view, and say that if, as has been suggested, deep seated germs possibly lying in the wrinkles of the skin perhaps in the glands and hair-follicles are enabled to come to the surface by soaking of the hands, such hands cannot be considered sterile.

There is unfortunately in all these writings but scanty reference to the actual authorities for this belief and, as a matter of fact, of practical research to demonstrate it there has been extremely little. It is however so plausible a view that it finds ready acceptance and would seem to furnish an explanation to many experimentors on skin-disinfection of the impossibility of attaining ideal results. In so far as actual research is concerned I can quote but two or three writers whose work has dealt specially with this particular problem.

Haegler (11), in a very elaborate and painstaking monograph on hand-disinfection states that under certain circumstances he has found bacteria lying in the hair-follicles and sweat-gland-ducts., in sections of skin examined with the microscope. I shall refer to this more fully later.

In 1900 Mohaupt (12) published an account of an experimental research, "Zur Frage der Bedeutung der Hautdrüsensecretion auf den Sterilisationseffect bei der Hautdesinfection". I may be excused if I refer to this in some detail as it is the most important experimental work so far which may be held to demonstrate that, as the result of germs lying in the sweat-glands being brought out with the stream of perspiration, a skin surface after a thorough disinfection apparently sterile can be again infected from within, quite apart from any external source of infection.

Mohaupt carried out four series of experiments. In the first a skin surface was shaved, cleaned with soap, hot water and brush and finally with aether. The skin was then

scraped with a piece of sterilised hard wood and a bouillon tube or Petri-dish inoculated with the cut-off end. The skin surface covered with sterilised protective-silk was again tested by scraping after a certain number of hours. It was found that rarely was the surface proved to be sterile, but that, while after the lapse of one hour hardly any difference was noticeable in the number of germs present, after some 24 hours a very great increase was observed. The writer attributed this increase chiefly among other possible causes to a growth of the germs under the sterile covering.

To determine the origin of the microbes a second series of experiments was carried out in which the skin was made to perspire by means of a steam bath at a temperature of 60 to 70° C. The hands were first scrubbed with soap and hot water for 10 minutes. The process of cleaning of the portion of skin chosen consisted in a scrubbing with sterile brush and soap and hot water for periods varying from 5 to 10 minutes, and immediately after the soap was rinsed off a rubbing with pads soaked in aether frequently changed during periods of 3 to 5 minutes. The skin was then covered as before with a piece of protective. Two successive inoculations were made with a piece of wood. The silk was then fixed with a bandage and the skin made to sweat in the steam bath for from 5 to 30 minutes. Inoculations were then twice made with wood as before and the 4 wood chips covered over in separate dishes with liquefied agar. Of 6 experiments 5 were on the sole of the foot and 1 on the forearm. Of the inoculations made before the sweating 3 showed the presence of microbes, on each respectively 1, 2, and 6 to 8 colonies. Of the inoculations after the sweating all showed growths varying from 1 to 50 colonies. In these experiments a maceration of the skin was observed as a result of the effect of the steam and perspiration.

Accordingly a third series of experiments was inaug-

urated, similar in all respects to the second, except that hot dry air at a temperature of about 120° C. was employed, and the skin was covered with a sterilised wadding compress so that the sweat was absorbed at once and the skin remained dry. Inoculations were again made with wood. 9 experiments were performed, all on the sole of the foot. The washing with water varied from 6 to 12 minutes and with aether from 3 to 5 mins. In two cases however alcohol was substituted for aether but used in similar fashion for 5 mins. Of the first inoculations 4 of the experiments showed growth of germs (3 plates infected), one "strongly infected" and the rest with from 1 to 15 colonies. The period of sweating lasted from 10 to 40 mins. Of the inoculations after this 4 were sterile (in one experiment both plates). The rest had colonies varying in number from 2 to 50. In one plate mould only was present. The case in which both plates remained sterile after the sweating, Mohaupt mentions was just the one ("accidentally perhaps") where the cleansing had been most prolonged. He also points out that on the occasions when alcohol was used the number of colonies after the sweating was relatively small.

Recognising however that it was impossible to exclude a certain even if slight degree of maceration of the skin by the sweat which would allow of the epidermal scales loosened in this way being more readily scraped off with the wood, the author performed a fourth series of experiments. In these the electric incandescent light was employed to produce the perspiration, and the exposure to it took place in a small mirrored cabinet or under a shade, the lamp and hand being surrounded with wetted muslin. Control experiments on the air showed only a small number or an absence of germs on Petri-dishes. laid out in the room for 15 mins.

The sweat was collected with a platinum loop, in

later tests with a piece of gauze. The skin experimented on was either the dorsum (11 times) or palm (5 times) of the hand. More profuse sweating was found to take place on the former, on which the sweat was sufficient to soak the gauze. On the palm the sweat only appeared as fine bright specks. In 16 experiments aether was used for the first 7, alcohol for the other 9. The washing was carried out for from 4 to 6 mins. followed by the use of aether for 2 to 4 mins. The alcohol, used alone, was rubbed on for 4 or 5 mins. In 4 cases a culture of *Bacillus Prodigiosus* was lightly rubbed over the skin surface before the cleaning with the point of the finger or the end of a test tube for 4 mins. In 3 cases the skin was moistened before hand and rubbed with the test tube without using any bacterial culture. In the majority of cases the first inoculation after the sweating was followed by another rubbing with aether or alcohol and a repetition of the sweating for varying periods of time. In the most prolonged experiment the sweating was produced 4 times successively lasting in all for 37 mins., never longer in any case than 10 mins. at one time.

35 inoculations before the sweating proved sterile and 16 inoculations after the sweating in these cases showed growths. In 2 cases there were present 9 and 5 colonies respectively; in the others one or at most two colonies. Of the 4 experiments on *Bac. Prodigiosus* out of 12 inoculations following the various periods of sweating 3 dishes showed a growth of the specific bacterium.

In the 5 experiments on the palm of the hand the 10 inoculations before the periods of sweating were sterile. After these periods growth was present in 4 dishes.

These results of the last three series might well be accepted as proving the truth of the already adopted belief in the constant presence of germs in the sweat-glands. As all of them had been carried out with an artificially produced sweating I was curious to know if the force of the stream in any way determined the readiness with which the germs were washed out and also their numbers, and I resolv-

ed to test the results with what may be called a normal degree of perspiration with a view to more definitely judging the practical significance of this for the operating surgeon. His hands are without doubt frequently moistened with perspiration but the conditions are in no way approximate to those involved in the artificial production of sweat by the aid of heat or light.

The methods adopted in my experiments differ considerably from those of Mohaupt and were chosen with a view to avoiding several obvious possible fallacies. The portion of skin made use of was the palm of the hand, chosen on account of its conveniently smooth surface, entire absence of hair follicles and glands, and because here the sweat glands are more numerous than in any other part. It is also the source from which possible infection from sweat is most likely to arise for the surgeon, as it sweats the most freely and so constantly comes in contact with instruments, gauze, or in fact anything held in the hand.

The whole of both hands and wrists were given in each case for fully five mins. a preliminary scrubbing with soap and cold, or at most tepid, water and nail-brush, but during this washing the palm to be experimented on was not scrubbed. A space of five mins. was then given to scrubbing in similar way the palm alone and its immediate neighbourhood using all possible energy. The soap used was ordinary white household soap containing no added antiseptic. In all cases the dishes used for washing, the brush and the water itself, etc., were thoroughly sterilised. The soap foam was then rinsed off with sterile water and an other five mins. were devoted to soaking and rubbing the palm with gauze swabs in a fresh dish containing plain cold sterilised water. The object of this last washing was principally to remove as far as possible any traces of soap that might still remain and possibly interfere with the culture experiments. A Petri dish with agar was thereupon inoculated along numerous

strokes with a sterilised knife with which the whole palm surface was scraped with sufficient force to remove countless epithelial cells. The skin surface was wiped carefully to remove any loosened scales still left behind by the scraping. A sterilised double fold of muslin, about two square inches in size, was then laid on the palm, the fingers covered over with another piece of gauze in several folds - were then clasped over the palm and the whole hand wrapped round with sterile lint. By this method it may be seen the muslin at once soaks up any sweat as soon as it is excreted, any maceration is completely avoided and the sterile gauze covering over the fingers as far as the web in front and behind effectually protects the muslin square from contamination by any other part of the skin. Provided that the gauze is sterile and the skin surface likewise, the only possible infection of the muslin square can come from the sweat. All friction or rubbing of the surface was avoided so that no epithelium could be brought away with the gauze as is the case with the wood-scraping method. After fixed periods of time varying in each experiment 5 mins., 10 mins., etc. during which the gauze was held in the hand, it was then removed to a Petri dish, the palm then carefully wiped clean with a moist swab and a fresh piece of muslin clasped on the palm. Liquefied agar was finally poured over the gauze squares in the various dishes.

It may be objected that the perspiration thus produced was insufficient to give results of value. It was found however by previous experiment that with the hand clasped and kept warm in this way a ready and quite obvious flow of sweat resulted within two or three minutes, and this effect could be seen in any of the subsequent observations in which the skin of the palm, except where in contact, with the gauze was found covered with moisture. It may be accepted, I think, that this does not inadequately represent the condition of the surgeon's hands during an

S E R I E S I.

Number	Scraping from skin.	1st piece of gauze.	2nd. Piece.	3rd. Piece.	4th. Piece	5th. Piece.	6th. Piece	7th. Piece.	8th. Piece.	Periods of sweat- ing each	Total duration of exper- iment	Remarks
1.	<u>Sterile</u>	3 cols. staph. alb.	<u>Sterile</u>	1 col. staph. alb.	<u>Sterile</u>	<u>Sterile</u>	2 cols. staph. alb.	1 col. B. subt.	<u>Sterile</u>	5 mins.	52 mins.	Palm of left hand used in all experiments.
2.	<u>Sterile</u>	2 cols. B. subt.	<u>Sterile</u>	2 cols. B. subt.	1 col. B. subt.	1 col. B. subt.				10 mins.	58 mins.	All cultures upon Agar. Temp. of room 82.18 to 20 C.
3.	<u>Sterile</u>	<u>Sterile</u>	2 cols. E. subt.	<u>Sterile</u>	<u>Sterile</u>					15 mins.	67 mins.	
4.	3 cols. E. subt.	1 col. E. subt.	1 col. E. subt.	<u>Sterile</u>						20 mins.	68 mins.	
5.	<u>Sterile</u>	<u>Sterile</u>	<u>Sterile</u>							30 mins.	62 mins.	

NOTE:- In these and all succeeding experiments an interval always of at least 48 hours, usually several days, was allowed to elapse between each, in order to enable the hand skin to resume its normal condition in case that had been distributed by the washing processes.

operation.

The results expressed in tabular form are shown on a separate page (series I).

It might well be thought at first sight that these experiments confirm to some extent the results obtained by Mohaupt. I could not however feel so convinced of this, in view of the very great possibility of air infection having occurred, either during the manipulations or previously, of the accessories used in them. The presence in the first experiment of the cocci which never re-occurred in any subsequent one was probably due to a too short preliminary disinfection of the various articles. Control tests with agar plates which were regularly made showed after 5 or 10 minutes exposure to the air a growth of anything up to eighteen colonies, and always the presence of bacillus subtilis, which it will be noted was the microbe present in all infected experiments. It was also once found that the simple pouring of sterilised agar from its tube into a Petri dish resulted in a growth of 4 colonies of this bacillus, and gauze taken direct at the end of an experiment from the box containing it and flowed over with agar showed similar growth. In such an atmosphere of a bacteriological laboratory and with so numerous opportunities of accidental infection of the gauze, etc. in the complicated manipulations and having only the rather inadequate assistance of a laboratory servant, I could not but have doubts that these bacteria should have originated from the skin itself. It is also generally recognised that the natural constant tenant of the skin is a staphylococcus albus (Lauenstein 13).

It was then decided to carry out a further comparative series of experiments, this time with an abnormal production of perspiration. The method employed was in all respects similar to that described, but, after the gauze had been clasped in the hand, the arm up to the elbow was laid in a laboratory "inspissator" an open shallow rectangular copper jacket, containing water which was kept at a

S E R I E S II.

Number	1.	2.	3.	4.	5.
Scraping from skin after cleansing.	1 col. B. subtt.	sterile	sterile.	sterile	sterile
Result on Gauze	sterile	sterile	1 col. B. subtt.	sterile	sterile
After profuse sweating for	30 mins.	25 mins.	20 mins.	15 mins.	10 mins.
Remarks: -	Temperature of inspissator, ca. 90°-75°C. Temperature of room usually 20°C. All experiments on palm on right hand. Cultures on Agar at 37, 5°C.				

temperature of between 90° and $75, 5^{\circ}$ C. The top was covered over with a thick piece of felt and the arm was packed round with a towel. In this hot air bath the forearm within a few minutes was dripping with perspiration, the palm however was kept dry as the gauze at once soaked up all moisture and no maceration of the skin resulted.

The preparations (washing etc.) were carried out in an other room of the laboratory from that first used. It was determined by control agar plates that the air here was distinctly freer of germs. In addition to this the experience gained both by the assistant and myself in carrying out the manipulations, and some minor improvements in the details, such, for instance, as keeping the muslin slips in a separate Petri dish apart from the other gauze, all tended to diminish the risk of an air infection.

The results are again shown in a table (series II).

As will be seen the only infection present on the knife strokes or gauze was in two cases a growth of *B. Subtilis*. This rather tends to indicate the origin of the microbe from an outside source, as the possibilities of the air infection of the gauze had been considerably less in this second row of experiments — there was not the same frequent opening and closing of the gauze--box as was inevitable in the former series, every time that the palm was wiped, and again when each fresh piece of muslin was required. The table however on the other hand fails to demonstrate that the sweat, profuse though it was and lasting for from 10 to 30 mins., contained any organisms brought from the depth of the sweat-glands, and this though in all cases the water used for washing was cold to avoid as far as possible any anticipatory perspiration and flushing of the glands.

To more certainly arrive at the truth a third series of tests was made in all of which was attempted an artific-

S E R I E S III.

Number	Growth of B. Prod. from skin after cleansing.	from Gauze	after sweating for	following rubbing in of Bacillus	Special prepar- ation of skin before rubbing in active bouillon culture.
1.	None	None	20 mins.	12 hours before	None
2.	None	None	15 mins.	6 hours before	None
3.	None	None	20 mins.	3 hours before	With ether, alcoh- ol, soap, water.
4.	None	None	15 mins.	1 hour before	With soap, water.
5.	None	None	15 mins.	immediately before	With alcohol, ether, soap, water.
6.	None	None	15 mins.	immediately before	With alcohol, soap, water.

Remarks:-- All experiments on palm of left hand. Agar at 22° C. used for all cultures.
Temperature of inspissator ca. 90° to 75° C. Temperature of room 20° C.

ial infection by rubbing in to the skin for a considerable time a pure and active culture of a specific bacterium. The organism chosen for several reasons was the bacillus predigiosus. It is of very prolific growth, its colonies and microscopic appearances are characteristic allowing of ready identification, it grows most actively at a temperature of about 21° at which other organisms that might be introduced from the air do not grow so well, it is extremely small and may be accordingly expected to find access to gland openings more easily and in greater number than a larger bacillus, and what is of importance, its presence had never been observed in any of the numerous control experiments on the air and on the undisinfected skin. An active culture in bouillon was rubbed with all firmness for ten minutes into the skin of the palm using either the end of a test tube or a finger of the other hand protected with a rubber stall. This was done immediately before or else at a definite time previous to the cleansing. The culture was never permitted to dry on the skin anywhere, and immediately after the rubbing the hand was thoroughly rinsed under a cold water tap for about a minute. In some instances the skin was previously washed with soap, alcohol or aether, or combinations of these, with the idea of removing fatty matter as far as possible from the skin which might oppose the entrance of the microbes but on each occasion a free douching with cold running water followed to remove in turn these various agents.

In all other respects the processes as regards cleansing etc. were the same as in series II. The results are given in table III.

These results show that the even prolonged and profuse sweating failed to bring out from sweat-glands any of the organisms which had been rubbed so thoroughly over their orifices, and the question must naturally arise from this as to whether any had ever entered the ducts; for it is hardly possible to conceive that not a single

bacterium which may have found entrance to the gland canals should fail to be brought out with the flow of perspiration.

I think it is therefore desirable to review in general the evidence on which this belief has been founded, to endeavour to arrive at what is the most probable truth in the matter. And first of all the experiments of Mohaupt are open to several serious fallacies which must be considered. These had not struck me so forcibly previously to my own experiments, but my own experience had shown me more clearly the enormous possibilities of error in a work of this nature. His first series of tests is avowedly no direct proof of the infectivity of the sweat, and we may pass at once to the others. The method of testing the sweat used in series II and III is eminently unsatisfactory as the plates were inoculated with the epithelium itself brought away by scraping the skin after, as the writer himself says, it had been macerated by the sweat. This is an insuperable objection to my mind, although a satisfactory enough proof perhaps of the possibility of an apparently sterile skin surface becoming infected after a time, which is quite a different question however.

The maceration of the epithelial surface was all but prevented, the author says, in the third series by the use of wadding which absorbed the sweat and kept the skin comparatively dry. But unfortunately it was not the wadding which was tested but a scraping from the skin from which the sweat had thus been removed. That the preliminary scraping of the skin surface so often proved sterile is a result which may well have been affected by the use of ether, and (in the latest tests) of alcohol, immediately prior to the scraping, and the removal of which is not stated to have been done. Deeper lying germs loosened by the softening of the skin with the sweat would not be so much influenced either by the direct action of the

agent, or by bits being carried over on to the culture medium, diluted as it would also be by the skin moisture.

But more significant still is the influence of aether and alcohol in drying and hardening the skin surface which interferes with a satisfactory inoculation by means of a scraping method, and any one who has studied such experiments knows that the results of successive inoculations from first dry, and then, moistened skin are absolutely different. This action of alcohol has already been shown to be a source of fallacy in the apparently favourable results in its use as a disinfectant (14), and would similarly account for the relatively small numbers of colonies following its use to which Mohaupt refers, while of course its hardening effect would be diminished after sweating had taken place for some time. On the other hand if the germs had come from the skin pores a larger number should be expected in this case as it produces an opening out of their orifices.*

The experiments in series IV more nearly approached a satisfactory test as here the sweat was collected without presumably removing any epithelium (a wiping of the surface following the wood-scraping is not however mentioned), and here there is a very striking difference in the numerical results as I have above described. The enormous number of colonies seen in the earlier results are absent - numerous tests remain sterile. Mohaupt explains this partly by the smaller areas of skin tested, and their condition being better suited physically for a cleansing process, and further suggests that the greater and more constant pressure on the sole of the foot in walking more frequently introduces organisms here than else where. I refer more fully later to this question of the effect of pressure.

In this series there is an other important fallacy, namely that the majority of the experiments were made on the dorsum of the hand, a surface studded with hair follicles which afford, theoretically at least, a still larger

* Haegler; loc. cit. p.97.

and better nidus for the deposit of germs.

It is also to be regretted that the author has not stated the nature of growth in the air control tests and more fully of those in the cultures from the hands.

I must make reference here shortly to the experiments of Genevet (15) as well. He held one of his fingers which had been vigorously mechanically disinfected in a suitable vessel with an aperture in the side, and through another aperture he poured bouillon medium into the glass leaving it in contact with the finger for one minute. The fluid was abstracted and fresh bouillon poured in. The hand and the vessel were then introduced into a hot chamber with a view to producing a flow of sweat which would pass into the bouillon. This was continued for 5 minutes. The two portions of broth were incubated, and in a series of some twenty experiments it was found that while the first bouillon was sterile the second showed germ-growth. To these experiments the best answer is provided by an investigation of Haegler's (loc. cit., p.92) in connection with alcohol disinfection. He found incidentally that a hand after alcohol treatment, apparently sterile, if bathed in sterile serum at the body temperature invariably gave off germs in constantly increasing number the longer the soaking lasted, and this had already commenced after 5 minutes had elapsed. The fallacy of Genevet's method is obvious then, and, as the cultures were made in bouillon, a single germ was sufficient to produce a positive result, and it is hardly surprising that the first portion of bouillon which was only in contact with the newly disinfected finger for one minute - as contrasted with five minutes at a considerably higher temperature in the second instance - should prove sterile. The remarks I have made above in criticism of Mohaupt's third series apply equally well here.

If I were to sum up the conditions necessary for a satisfactory test of the infectivity of the sweat and the sweat alone, I would postulate a method which collects the

sweat free from any epithelial cells, which is proved on a skin surface free from all except sweat glands, a surface by preference which affords a criterion for the surgeon, which avoids the use of all antiseptics for the cleansing process or at any rate removes these again as completely as possible; and further that the air infection must be tested in all cases to compare with the germs obtained from the hand. As of interest it may be mentioned that the only experiments of Mahaupt on the palm of the hand after rubbing in B. prod. showed no growth of this organism.

But to turn from this to other evidence. So long ago as 1875 Eberth (16) and in more recent times Brunner (17), v. Eiselsberg (18) and Gärtner (19) had found bacteria in sweat. That cocci should be capable of demonstration by these authors in the sweat of pyaemic or septicaemic patients has little practical significance for the operating surgeon. In corresponding way it may be mentioned Geisler (20) found typhoid bacilli present in a severe case of that disease. As regards normal skin Ziegeler (21) carried out an experiment demonstrating the increase of bacteria on the skin surface after free perspiration. His experiments which were widely published at the time in the lay press were as follows. A bath of 200 Ltr. water was found to contain 240 germs per ccm, i.e. 48 millions in the whole. After one person had used it, the germ-count showed 80 millions. Another bather, freely perspiring as the result of a hot air bath, entered the water for the same period and thereafter the germ count amounted to 144 millions. The sweating skin had therefore given off double the number of germs given off by the previously dry skin. Such a demonstration however is hardly convincing, but the writer argues from it the desirability of a sweat bath for all parts of the body to be operated on and also for its prophylactic value in freeing the body from microbes.

Petruschky 22) found that from smooth skin surfaces*, cleaned with aether and alcohol and then shaved, the epidermal scales removed rarely gave growth on agar, only exceptionally a growth of large cocci and never streptococci or staph. pyogenes. Lauenstein 18), examined bacteriologically in 147 cases patients' skin in healthy and inflamed, undisinfected and disinfected conditions. He found the staph. pyog. albus to be the microbe most frequently present. He attributes the difficulty of disinfecting the skin to the hair fat; the palm and fingers are therefore much easier to disinfect. He inclines to the view that the sweat glands have little influence on disinfection as he noticed no difference in the frequency of microbes in different parts of the skin, even though varying so greatly in the number of contained sweat glands.

Samter 23) at the same time published the result of an examination of the skin removed from the site of operation following a disinfection with soap, alcohol, aether, carbolic acid and sublimate. Out of these only 20 proved sterile. In 12 cases E. prodig. was rubbed in after the use of soap alcohol and aether. On 6 occasions cultures showed the bacillus. Once nothing was present and in the other 5 cocci alone.

* My foregoing experiments show the possibility of a merely mechanical sterilisation of a smooth skin surface being achieved. They cannot of course be taken as a criterion for a similar sterilisation of the whole hand.

Schumacher 4) more recently has published some similar investigations. He starts with an already formed belief that "ducts of the fat and sweat glands allow the entry of microbes and their long duration there. To a staphylococcus of $.7\mu$ nothing stands in the way of its entering into the comparatively wide cavities in the skin and wandering into the subcutaneous tissue." Such germs are beyond the reach of all disinfectants. He cleaned the field of operation in patients with spirit soap for 5 minutes, then applied a sterile compress for one and a half hours, and finally shaved the epidermic scales over an area of 5 x 2 cm. The shavings were placed in an agar-tube. Then a portion of skin $\frac{1}{2}$ cm. was taken from the incision and minced up with a knife in a Petri dish. Control plates showed as a rule only a small number of colonies from the air, mostly moulds. Out of 30 cases the surface cells gave a growth of staph. pyog. albus in 6 cases, the rest remaining sterile; the deeper skin examined was only 5 times sterile, the remainder showed staph. albus 21 times and in others staph. aureus and citreus and sarcinae. The incisions healed well, 2 out of 22 showing stitch abscesses. The author concludes that the cleansing of the skin surface loses its importance so long as germs can lie concealed, deeply seated and able at any time to be brought to the surface with the stream of out flowing secretion. But while these demonstrate the difficulty of a deep disinfection the actual situation of bacteria in the skin is still undecided. Obviously only a microscopic demonstration is of value.

Schimmelbusch 24) dealt with this in an important research on the cause of furunculi. He found by experiment that folliculitis might be produced by rubbing a growth of staph. pyog. aureus into the skin. Two things were necessary for its production, the presence of casual microbes and a process of rubbing. Cocci merely laid on the skin produced no result. Microscopically the hair follicles and glands showed numerous microbes, the sweat glands never.

Mohaupt thinks this is due to their having been washed out afterwards as the examination took place 24 hours after the inoculation. But similar examinations made by Schimmelbusch on freshly amputated skin showed cocci in the hair follicles only, nowhere else. He therefore refuted the previously accepted view of boils being a sweat gland inflammation and held that the alterations of these glands as observed by Longard 25) were only of secondary occurrence.

Wasmuth 26) after similar researches came to a like conclusion. He believes that the hair glands (though not the follicles) and the sweat glands do not afford an entrance for germs.

We now come to the most important work of Haegler II) who in 1900 published a most comprehensive and thorough work on hand disinfection and cognate questions. Chapter II is devoted to a consideration of the localisation of the micro-organisms on the hands, and I must review it in some detail especially as he is, in the few references of writers on this sweat gland question, almost the only authority quoted, and, if I may say it, frequently misquoted. Haegler attributes the little work done on the subject of germ localisation to the great difficulty and tediousness of an investigation involving, as it does, a careful examination of hundreds of sections. His first hundred sections of cleaned and uncleaned skin showed only surface organisms, passing no deeper than under any raised up or loosened epidermic lamellae. As the detection of organisms, was found, however to be unsatisfactory, he employed Indian ink, whose minute particles are readily seen with a microscope, and the preparations were carried out on living skin previous to the amputation of various extremities. He found that if the ink was painted with a brush on the skin, or if ink soaked compresses were kept moist on the skin up to 24 hours, the sections showed the particles never deeper than the outer epidermic layers, lying in any crevices of the skin but never in glands or follicles.

If the ink was rubbed in with the point of the finger for 2 - 3 minutes, the sections were identical except that the epidermis being perhaps slightly roughened by the rubbing admitted the ink particles "one cell - layer deeper". The hair follicles also showed particles as far as to about the opening of the sebaceous glands; in the sweat glands they were found in the first, at furthest, in the second spiral. The numbers present were as nothing compared to those found in an accidental microscopic injury of the skin. Haegler attributes this to the resistance offered by the fatty nature of the glandular secretions. With microorganisms similar results were obtained but the penetration was rather deeper in the ducts. In one section depicted from a working man's hand, the cocci had reached the fourth spiral, which the author says was exceptional. Having now acquired experience in the technic, Haegler made some further examinations - over 500 - of "unprepared" skin. In the hair follicles germs, usually cocci, were to be found sparingly in numerous preparations but quite superficially and in the sweat glands they were never found.

Haegler discusses the question of possible multiplication of the organisms in the hair follicles. He considers the fatty secretion an unfavourable medium for their growth, but to test it experimentally he examined skin on which organisms had been rubbed in as before, 72, 48, and 24 hours before its removal from the body. In all cases the sweat ducts were found to be empty, and in the hair follicles the organisms were found to be much more superficial and more infrequent (often entirely absent) as compared with control preparations from same cases (rubbing in immediately before the removal). That the organisms on the surface of the skin do not grow down into the depths, he demonstrated by keeping virulent cultures of staph. aureus. in contact with the skin for several days, after which no inflammation resulted, and also by microscopical

examination of sections of skin covered with the whole contents of a Petri dish of staphylococcus culture for 24 hours. In these not once were the germs found to have penetrated the peripheral ends of the hair follicles.

The summing up of these important investigations, which I may be allowed to quote as literally as possible, Haegler states as follows: "that the germs on our hands are present only relatively seldom and sparingly in the hair follicles and here only in the peripheral portions; that they are normally absent from the sweat glands and into all these natural skin openings do not grow but are brought there from outside by rubbing; that once there they do not under normal circumstances develop but - though through the stream of secretion likely - are again driven out; that the accidental skin openings, the minute injuries, present a difference, in that germs can be regularly found there and it may be accepted that they can increase in the depth as also grow in from the skin surface towards the bottom of the small wound-canals."

It is somewhat difficult to know perhaps what conclusions rightly to draw from the whole of this review. Based on Haegler's and my own results my belief inclines to the idea that the whole significance to the surgeon of sweat gland infection has been grossly exaggerated if it is not even entirely fallacious. Its general acceptance by workers on hand disinfection is probably a welcome explanation of the impossibility of obtaining complete sterility by any of their methods. In any case most confusing are the views announced.

I have already mentioned Mohaupt's assumption of the effects of pressure in introducing organisms into the sweat ducts, and also the similar views of other writers. As a matter of fact a simple test of this point can be made. If a normal hand, most conveniently the hypothenar eminence carefully wiped dry from all apparent moisture is stroked slowly and firmly with, say, a glass slide, it is found that a considerable quantity of moisture is brought away

from the skin. It may really be accepted that ordinarily the glands are in constant activity and constantly full of secretion. The "insensible perspiration" is a recognised fact, but apparently often forgotten in this connection; and to a statement such as that of Schumacher 4) already quoted that nothing stands in the way of a minute coccus entering a duct and wandering into the subcutaneous tissue, I would unhesitatingly reply that a constant stream of fluid of comparatively enormous volume stands in its way, not to mention a very tortuous course to be followed were it even able to stem the tide. If this stream as is so often said is able to wash out the supposedly deep-lying cocci during an operation, it can as well do so at any other time and to my idea is always doing so, or more accurately is continually opposing their entrance. Some writers will almost have us to believe, if we accepted their rather random statements, that the bacteria lie in wait till the surgeon has commenced his operations and then only come out with the now flowing stream, on purpose to upset his best laid aseptic plans. On the other hand experimental investigations and common sense would seem to indicate that the germs do not naturally find an inlet. The nature of the sweat secretion is also important. Its fatty contents are recognised, (Ziegler 27) and afford a natural resistance to the entrance of germ-containing fluid, and it is also questionable whether its re-action is favourable to germ-life, (Heim 28). * The contents of the larger sweat glands it may be noted are only semi-fluid containing numerous solid particles.

That under some abnormal conditions the germs may find access to the glands one can hardly deny in the face of Haegler's work, yet it is surprising that in my own experiments, after an energetic rubbing ⁱⁿ of *B. prodigiosus*, not one could be recovered from something like 6000 glands

* Tests with litmus paper during my own experiments showed a distinct acid re-action of the normal sweat secretion from the palm.

in full activity (in the palm of the hand Krause estimates the number of sweat glands at about 2800 to the square inch), and this too though the fat had been removed to some extent from the skin by alcohol etc.

If for the sake of argument however it be admitted that germs lie in the outermost parts of the efferent duct at certain times, I am positive that their duration there can be but short, and as soon as the skin resumes its normal activity the germs must be driven out. How otherwise would Haegler, Schimmelbusch and others have failed to find cocci in the sweat glands of normal skin after examinations of scores of sections? The absence of primary (i.e.) starting from the skin surface) inflammatory affections of these glands is a clinical fact not otherwise explainable. Contrast this with the frequency of hair gland and follicle suppuration, but at the same time keeping in view the preliminary abnormal state of affairs necessary to produce this latter condition and the comparatively slow and feeble flow of secretion from the sebaceous glands. As skin pathology throws some interesting side-lights on this question I may with advantage quote here certain opinions expressed by one of its foremost exponents. Unna (29, p. 348), speaking of the Deep Inflammations of the Epidermis, says "while the immigration of bacteria and fungi from the surface into the entrance of the hair follicle has been proved to demonstration, clinically, experimentally, and histologically, one cannot yet regard Spiradenitis, (the primary inflammation of the coil glands), as a definitely established fact..... Here the anatomical nature of the entrance, and the relatively long narrow canal, which, in contrast to the hair follicle, is constantly washed by a centrifugal stream, and therefore presents the greatest resistance to a centripetal entrance of organisms, opposes not a few difficulties. For we see, that these very organisms, which are exceptionally well adapted to the acid re-action and other peculiarities of sweat, the flora of the flexures

and the genitals, show no tendency to wander into the sweat pores, against the sweat stream." Again (loc. cit. p. 257) speaking of Impetigo Staphylogenes he makes the striking observation, "There was never in these cases any penetration of the cocci into the sweat pores, and I was never so fortunate to be able to follow, even from the densest layers of cocci on the surface, a single one into the sweat pores".

To turn to the practical surgeon's point of view is it not the case that the scrubbing and the hot water are the very things to produce a hyperaemia and consequent glandular activity and flushing out of the gland ducts? I will not deny that the results obtain in my experiments with B. prod. are not due to this action, though by the use of cold water as described it was avoided as far as possible, but on the other side it is quite as justly to be claimed that the germs never did find entrance, as that they did enter and were subsequently removed in the washing.

Much still remains to be learned regarding the bacteriology of the skin and the complicated problems involved in the question of the "abstinence time" necessary for the operator to observe after handling infectious material. No one will presume to deny that virulent germs may find a resting place on the skin and in spite of all disinfection still remain capable of infecting fresh wounds in which the surgeon may unhappily deposit them. Here again the experimental evidence is slight, and clinical observation is the chief source of knowledge. The important question which one naturally wishes an answer to is where do the germs find their nidus. Is it in the glandular structures or on the outer epithelial surface layers?

I have dealt above with the significance of the sweat-gland and ducts and the sweat gland secretion on an experimental basis. There remain the hair structures—the hair follicle and the sebaceous gland secretion—to be

S E R I E S I V.

Number	Scraping with knife from small portion of skin, previously shaved.	Hairs cut off.	Hairs removed by forceps with roots.	Previous rubbing in of active culture of	for	following of cleansing skin with
1.	1 colony	1 colon. B. subt.	Hairs all sterile	---	---	---
2.	2 colonies staph.	Hairs all sterile	Hairs all sterile	B. Prod.	5 mins.	Soap water.
3.	Sterile	1 Colon.	Hairs all sterile	B. Prod.	5 mins.	Aether alcohol water
4.	1 colony	Hairs all sterile	Hairs all sterile	B. Prod.	10 mins.	Aether alcohol water

Remarks: - All experiments on left forearm. Sterilisation with soap and cold water and brush 5 mins. with plain cold water and gauze 5 mins. All growths on Agar at 21°C. In each case from 8 to 16 hairs tested.

considered. It is obvious that this hardly lends itself as does the sweat to the experimental method, as the flow of the sebum is comparatively so sluggish, and cannot be stimulated by any known suitable means. It is doubtful therefore if it need weigh much in the surgeon's calculations even if it be assumed to be heavily laden with germs, since it is in all probability not excreted sufficiently rapidly to reinfect during the space of an operation a previously disinfected skin surface. Still it is of interest, to estimate to what extent germs are harboured by the normal hair-follicles and the contained sebum. One might a priori suppose on pathological and microscopical evidence that this retention of organisms is considerable.

I wished first however before drawing any conclusions to test the bacterial incidence in the hair follicles, or at the same time perhaps determine their facility for being sterilised by mechanical means, by means of a cultural examination of the follicular portions of the normal hair, also after if that were possible, an artificially produced infection of the follicle. Experiments were accordingly made as shown on the table, Series IV. The radial aspect of the forearm was chosen as presenting a stronger hair growth than any other convenient area of skin. In three cases an active Bouillon culture of *B. Prodigiosus* was rubbed in after the surface had been cleansed in various ways with the idea of removing fatty matter which might impede the entrance of the bacilli into the mouths of the follicles. The culture was then rinsed off from the surface under the cold water tap. In the first case no such preparation was made. The washing processes were the same as in the other experiments already detailed, except that the tenderness of the skin prevented so free a use of the brush as on the palm of the hand. As controls, a scraping was made from the epithelium over a small area previously shaved, and also hairs were cut off above the surface. The results appear on the table.(IV).

The incidence of colonies on the knife strokes and on the hairs cut off above the surface is a good indication most likely of the difficulty of sterilising these by a purely mechanical process, which as just mentioned could not be so vigorously carried out as on a tougher portion of skin.

Heagler in referring to experiments on the presence of germs on hairs pulled out from their sheath (loc.cit.p.15) alludes to the possible fallacy that such have merely been collected by the hair as it passed the surface layers; but the disinfection and the control tests on the skin avoid this, though in my own results it is negligible as such hairs proved to be sterile. One is entitled to assume, I think, that these extracted hairs would in at least some instances have brought with them germs already lying in the hair follicle or introduced into them by the artificial infection with the pure culture.

One is forced then to the conclusion that bacilli had either not entered the follicle or had reached only so far as to allow of being removed in the washing.

In any case it furnishes the final answer to the question above propounded regarding the nidus of the germs on the skin of the operator, that is to say the real source of danger for infection lies wholly in connection with the epithelial scales themselves.

It should therefore be the surgeon's object to avoid all maceration or softening of the hand during the operation. The sweat is of some significance in this connection, that is to say in so far only as it may cause loosening of the epidermic squames. The hands should accordingly be kept dry and if washing in water at intervals is done, the surface should be carefully wiped dry thereafter. The use of spirit for this washing may be a useful substitution but apt to prove uncomfortable.

The conclusion drawn from the foregoing I may finally sum up in the following manner:-

1. There is no evidence to show that the sweat glands or their ducts of normal skin afford a resting place for germs.
2. Experimental evidence to prove this is unsatisfactory, and microscopic examination has always been negative.

3. There is every reason for supposing that the stream of secretion, its fatty nature, and its constant flow, afford a complete protection against the entrance of germs.

4. Under certain abnormal conditions, for instance, on a very dry rough skin wanting in the usual fatty material, and by rubbing in of organisms the gland ducts at their orifices may perhaps be made to admit a few such, but these very rapidly are eliminated.

5. The surgeon can afford to disregard the sweat secretion as being itself a source of re-infection of the skin.

6. It is comparatively more easy for organisms to enter the superficial parts of the hair follicles but not the hair glands. Here also an elimination takes place and the contained secretion is inimical to germ-growth.

7. The process of rubbing considerably influences the presence of germs in these follicles.

8. There is little if any evidence to show that the secretion from the hair follicles is of practical significance to the operating surgeon.

The experiments described in this investigation were carried out in the laboratory of Dr. Piorkowski, Luisenstrasse, Berlin. It gives me much pleasure to record here my grateful thanks to him for the active interest he has all along taken in the course of the work and for his always ready and valuable expert advice.

SECTION II.

The circumstances determining the incidence and duration on the skin of other than the permanent epiphytes.

It can hardly fail to strike anyone who has read through the literature on the subject of the disinfection of the skin that, despite the enormous amount of work that has been brought to bear on this question, practically nothing has been done to elucidate the problems of primary importance connected with the circumstances which determine the incidence and existence on the skin of the bacteria whose complete and certain removal or destruction no process yet devised has attained to. Yet it must be obvious that to arrive at a satisfactory method of disinfection a proper understanding of the nature of the infection of the skin is desirable. The difficulty of settling the matter, as so many different factors are at work, is possibly one reason for its neglect, but any further progress towards the ideal must rest on a better knowledge of the factors to be contended with.

Döderlein 7) has given expression to a similar view in an admirable review of "The present position of the hand disinfection question and the next problems of the same," which merits quoting at some length. Speaking of the germs which probably become indigenous in the follicles and the sweat ducts, he says, "of what nature these skin bacteria are, what sorts are here present, whether certain hands cultivate particular varieties or if rather we also here, as on other parts of the body, as for instance the vagina, grow special kinds through eclectic selective breeding from the variegated mixture which we day by day catch up, these are all unsolved questions which await a grateful but difficult investigation. But one might be induced to accept, a priori, that these normal and ineradicable inhabitants of the skin are, as a rule at least, not of pathogenic nature, for the dream of antiseptics has indeed been profoundly disturbed; we never operate free from germs and have never operated germ free. If we then--and there is no rea-

son presumably to doubt it--hold fast to the belief in the evil results of introducing pathogenic germs and in spite of this infection of the hands observe as a rule quite faultless healing of wounds, then the fact is evident that it must be parasites, relatively harmless, and easily dealt with by the body and its tissues which we give off during the so-called aseptic operations. From these "normal" tenants of the skin we must strictly separate the foreign but pathogenic germs at times brought on to the skin of our hands, such as those the physician and midwife pick up from occasional contact with poisonous materials. To what extent now can such strangers become indigenious? How deep are they capable of penetrating into our skin, into its secret recesses and gland ducts? Can they increase in our skin, can they really become ineradicable skin dwellers? Here also we hit upon a purely open questions which admit of experimental solution. Practical experience allows us here also to anticipate and leads us to the conclusion that such foreign bacteria are not able to become permanently indigenious, else had we medical men never had non-infective hands, especially at that time when one did not yet so strictly aim at avoiding infection of the hands as now with every reason the watch-word is. But of the greatest importance now for the practical activity of surgeons is before everything the question, how long such pathogenic germs accidentally caught up adhere to our skin; The difficult question of the obstetrical abstinence after septic contact demands urgently an experimental investigation of the adherence of pathogenic germs to the skin; the avoidance of the touching of septic matter has indeed become a serious question of conscience from the moment when the non-disinfectibility of the hands became dogma."

It was with somewhat similar ideas in mind that the following work was undertaken. The question of the absti-

nence time as is evident depends on an extended knowledge of the duration on the skin of other than the regular epiphytes. A considerable amount has been written on this subject of abstinence based chiefly on purely clinical experience, many writers actually decry the value of any experimental evidence, and naturally as the clinical observations of each have varied the result has been quot homines, tot sententiae. Curiously the matter has been principally taken up by the obstetricians and gynaecologists, though it must be of equal importance to all engaged in operative work. Probably this is because in the practice of obstetrics the sources of any infection are more readily observed and controlled than is possible to the general surgeon who so frequently has to come in contact with septic matters, and again the obstetrician is usually in a better position to observe an abstinence time than the surgeon who is perhaps required daily to undertake operative measures. The consequence has been that, while surgeons have come to place a very great reliance on methods of disinfection, there has arisen a considerable amount of scepticism of their value among obstetricians, especially those in charge of lying-in institutions attended by students. For many cases of puerperal infection have been traced to examinations made by students who in spite of the use of disinfectants have evidently failed in ridding themselves of infection. In the case of students this is no doubt very largely due to an inadequate acquaintance with the best methods of disinfection or a lack of that conscientiousness which the responsible physician must feel, but nevertheless it has been recognised by many that it is not permissible to rely on a single disinfection of the hands.

The term abstinence time itself suggests the idea that time can accomplish what disinfectants fail to do. But some authorities have come to regard it rather as merely implying an extended period in which to carry out repeated disinfectant procedures. It will be as well here to briefly quote the most authoritative expressions of opinions which have been published.

Góth 30), writing in 1883, looks upon the hands as the most dangerous source of infection of women in child-bed. While cleansing and disinfection are indispensable, if the doctor has touched infectious matter as after examination of women with carcinoma, handling of cadavera, etc., this is not enough and he must pause in his obstetric activity, as clinical experience has shown that one does not always succeed in rendering infectious stuff on the fingers harmless. The duration of this pause must be decided by numerous observations, it must probably vary according to the intensity and the nature of the infection. He recommends the use of each hand alternately. If both hands are infected then one must wait till the upper layer of the epidermis to which the infectious matters cling is rubbed off. How long this requires cannot be stated as conditions vary, but undoubtedly frequent washings (mechanical disinfection) will assist the result. The author does not however give even a suggestion how long actually abstinence should be observed.

Zweifel 31) speaking at a discussion in 1899 recalled the fact that already twenty years before he had expressed his belief that not rapid disinfection but only time, "an indefinable something", frees the hands from germs.

Löhlein 32) considers it impracticable to carry out abstinence for a long time, and it affords no guarantee against later infection. He thinks one may rely on disinfection.

Fritsch 33) supports Löhlein's views and remarks that, "as not time but the disinfectant kills the bacteria, so has abstinence little sense.

Wiener 34) approves of abstinence especially for those

incompetent to disinfect themselves properly, as midwives in particular, and looks upon the sense of smell as a good test of success of disinfection after handling septic matter.

v. Swiecicki 35) gives a review of the views of various authorities. Winckel 36), Zweifel 37), Schröder 38), Martin 39), and Spiegelberg 40) had written in favour of the necessity of observing an abstinence time, based chiefly on their clinical experience and the times given ranged from 1 to 14 days. On the other hand Küstner 41), Ahlfeld 42), and Volkmann 43) looked upon it as uncalled for. v. Swiecicki himself is in favour of it and puts it at 8 days at least.

Fürbringer and Freyhan 44) point out that even the most conscientious operators get bad results, but the practitioner cannot wait a number of days till the epidermis of his hands has renewed itself. He must therefore disinfect them. Unavoidable misfortunes should not lead to pessimism.

Henke 45) attempted to settle the question of the possibility of disinfecting the hands at one sitting by experimental methods. The experiments led to the conclusion that artificially infected hands were more easily disinfected than "normal" not specially infected hands, the same process being used for both. This paradoxical result induced him to decry the observance of abstinence time as useless.

His results were severely criticised by Sperling who pointed out the fallacies in the methods of experiment and the conclusions. 46)

Sarwey 47) in an admirable review of the whole question also criticises Henke's work very adversely. He deals with the subject from the point of view of the lying-in institutions attended by students and gives an interesting table showing the methods in vogue at twenty one such places in Germany. At all of them the students were required to avoid infection for from 1 to 4 days before examining

patients in child-bed; 1 or 2 days was required in the majority of cases. The reasons for this enforcement are also given and these are usually on disciplinary grounds or in order to give the students opportunity for repeated disinfection.

Of the writings of others dealing with the question from a more general point of view, to determine the length of time that organisms might remain on the skin there is not much to be referred to. Binaghi (48) found that, if pure cultures or such septic matter as faeces, purulent urine, or sputum was spread over the skin and allowed to dry on, after 6 days the germs had diminished in number and virulence. He ascribes to the skin a certain disinfectant power, but this is hardly a legitimate conclusion as he had previously disinfected the skin.

Haegler (loc. cit. p. 177) experimented with virulent staphylococci on the skin of his thigh but found no diminution in their virulence after 6 days. He also found that an artificially infected hand, if cleaned immediately, could be readily freed from the specific organisms employed; if however they were allowed to dry on and the hand was not washed for several hours, but especially if the skin was at all rough or cracked, then even three days later isolated colonies of the germs could be obtained in spite of frequent mechanical cleansing. Haegler however expresses the opinion that the question of the abstinence does not admit of solution by experimental methods.

Heile (49) in an article on operation gloves mentions incidentally that he had once found *B. Prodigiosus* to persist on the hands for 15 days although 15 times disinfected in the meantime.

While admitting the great possibilities of experimental fallacies, in spite of the frequently uttered opinion of the uselessness of experiment on such a subject, it seemed to me that the unsatisfactory results obtained by experimentors have been due to their taking a too restricted view of the objects of investigation. It seemed clear that in consideration of the not to be excluded personal element, it was desirable to aim at determining the principles at work and the relative importance of the factors involved. Only with a knowledge of these could one arrive at conclusions applicable to special points, as, for instance, the abstinence time. But not only has it proved necessary to weigh the relative significance of these factors, but experimental work has been necessary in some cases to determine what factors actually required consideration, as these are not all at first obvious. On the other hand some which one would have imagined important proved not to be so, and again it was found that some only come into prominence in the presence of certain other factors. All this has made the work a somewhat complicated matter and the results have in certain instances been surprising. It is not pretended that absolutely identical results would be obtained by any other experimenter on similar lines, but by aiming only at a settlement of the principles involved, it may be permissible to presume that any practical deductions made from them may justly be applied to special cases.

It is necessary to premise here that I agree with the opinion of Döderlein already quoted that the germs found to occur on the skin may be divided into two classes, the permanent and the temporary dwellers. When I speak of permanent organisms I refer to those micro-organisms belonging to a group, representatives of which are always to be found in inoculations made from the skin, namely staphylo-

cocci of various kinds. In hundreds of inoculations made I have I may say invariably found staphylococci giving colonies either white or of a great variety of shades of yellow. What determines the existence of these as regular epiphytes is considered in a subsequent section. As for the temporary germs it is obvious that they may consist of an innumerable variety depending entirely on what the skin has come in contact with and so picked up. As naturally the majority of these reach the skin from the air and dust they are for the most part non-pathogenic, a small minority however may be of considerable virulence depending on the environment, as for instance in the case of the medical man who comes in contact with pus or infected materials or air. Haegler (loc. cit. p. 90) mentions having found tetanus bacilli on the fingers of working people.

How long such germs sojourn on the skin and what brings about their disappearance is the question in point to be settled.

Experiments therefore necessitate an artificial infection of the skin. The choice of suitable organisms for this purpose is of great importance. After consideration it was decided to employ *Bacillus Prodigiosus* and *Bacillus Pyocyaneus*.* They are both very actively growing, and their appearance is characteristic under the microscope or in colony growth on agar or gelatine, even in presence of other organisms, though as a matter of fact they usually appear on cultures, at room temperature at any rate, before other germ growth takes place. They are of small size so that one may assume that they are capable of penetrating into the recesses of the skin as far as any other germs. While *B. Pyocyaneus* grows perhaps best at body temperature *B. Prodigiosus* grows better at a considerably lower one, and so the varying degrees of temperature of the exposed skin surfaces are fully allowed for by the use of these

*In one experiment *Bacillus Violaceus* was made use of. It was found however that it was not so satisfactory as the two others as its colonies did not form the characteristic colour with sufficient rapidity.

SERIES 1. Experiments on normal skin of the hands.

No.	Preparation of skin.	Mode of infection	Parts tested.	Bacillus used.	Inoculations made after	In the interval											
						1/2	1	1 1/2	2	3 1/2	6	8	12	24	36	48	72 hours.
1.	L. Hand, 1 hour after last washing, soaked with water to soften skin.	Bouillon culture rubbed firmly in for 5 minutes. Drying avoided; hand immediately rinsed with cold water.	Palm. Dorsum. Nail folds & grooves	Prodigiousus. Violaceus Pyocyaneus.							1	2	2	3	5	5 washings with soap	
2.	L. Hand, 10 minutes after last washing.	Bouillon culture stroked over skin till dry. First washing 2 hours later.	Palm. Dorsum.	Prodigiousus. Pyocyaneus.						1	2	2	3	2	3	ditto	
3.	R. hand. 1 hour after last washing.	Bouillon culture stroked over skin till dry. First washing 6-12 hrs. later. Hand rinsed with cold water between 1st. and 3rd. inoculations	Palm. Dorsum.	Pyocyaneus. Prodigiousus.				0 0		0		1	2			ditto	
4.	L. Hand.	Bouillon cult. stroked over skin at night. Allowed to dry on 1st washing between 1st. and 2nd. inoculations.	Palm. Dorsum.	Pyocyaneus. Prodigiousus.								0 1				ditto	
5.	R. Hand. A few mins. after last washing.	Bouillon cult. stroked on skin till dry.	Palm. Dorsum.	Pyocyaneus. Prodigiousus.		0	0	0						5		ditto	
6.	R. Hand.	Bouillon cult. rubbed firmly over skin till dry.	Palm. Dorsum.	Pyocyaneus. Prodigiousus.				0						5	6	ditto	

REMARKS:- All inoculations made with sterile knife on liquefied gelatine in Petri dishes. Incubated at room temp. -20-21°C. All control inoculations (not entered on table) made immediately after infection of the skin from a small area were positive.

† implies a positive, 0 implies a negative result, or regards growth of the specific organisms employed to produce the artificial infection.

NOTE:- "Washing" means ordinary with soap and cold water, without use of nail brush. Skin wiped but not rubbed dry with towel

particular organisms. All cultures were made upon gelatine at room temperature (about 20°) at which *B.Pyocyaneus* also grows sufficiently rapidly and with the distinguishing colour formation, and gelatine was found to give the most readily identified cultures. Another advantage of this procedure was that the always numerous staphylococci colonies did not appear till comparatively late and so did not mask the growth of these two bacilli.

It may be objected that as they are not pathogenic, tests made with them are of little practical value, but it should be kept in mind that in their saprophytic existence on the skin surface this matters but slightly or not at all; of prime importance is the power of resistance and vitality. As these are features of the bacilli chosen what is demonstrated by experiments with them applies fully to less resistant even if pathologically very virulent parasitic organisms.

1st. Series.

In this first series of experiments the palm and the back of one or other hand and in one case the nail-folds and the spaces under the nails of all the fingers were infected with an active bouillon culture of *E.Prodigiosus* or *B.Pyocyaneus*. The skin of the hands was normal--not prepared in any way, but smooth and well preserved-- and it is important to note that they did not come in contact with any antiseptics during the course of the tests. The infection was produced usually a short time after an ordinary washing with soap and water. In one case (Exp.1) the skin was soaked with water to soften it immediately before inoculation. In the first case the cultures were firmly rubbed into the skin for 5 minutes, but not allowed to dry at any point, and the hand was at once thereafter well rinsed under the cold water tap. In other four cases (Exps. 2, 3, 4, 5) the cultures were spread over the skin not using

any great force but were allowed to dry on. The hands in the last experiment (6) were rubbed firmly with the culture till it had quite dried on to the skin. The hands were used in the ordinary daily occupations and no special attention was given to them. The only precaution taken was that gloves were not worn during an experiment to avoid a possible reinfection from this source. Inoculations were subsequently made from the skin at successive periods of time varying as a rule from 3 to 72 hours. This was performed by scraping the surface with a sterilised steel knife sufficiently to remove enormous numbers of epithelial squames, a different portion of skin being selected on each occasion. Liquefied gelatine in Petri dishes was inoculated and agitated so as to distribute the cells throughout the fluid. The gelatine was then allowed to cool and kept at room temperature (20--21°C.) under observation for about three weeks. This method was found by comparative tests to give the best results. The inoculations were made sometimes with, sometimes without, any soap and water washing of the hands in the intervals, sometimes after a simple rinsing with cold running water to remove any gross dirt that might have accumulated. The activity of the cultures used was proved by subsequent recultures and as a rule a small portion of skin was scraped immediately after infection and always resulted in a profuse growth of the corresponding bacillus.

Full details of each experiment are shown in the Table No. I. As may be seen all results excepting in Exps. 5 and 6 were negative, even three hours after infection had taken place. In exp. 4 the skin was infected at night and the inoculations made in the morning on rising so that the hand had been subjected to a minimum of friction compared with that which occurs during the daytime.

In exp. 5 four out of six inoculations made $\frac{1}{2}$, $1\frac{1}{2}$, and 3 hours after infection proved positive, several but not many colonies being present. As all were not positive it

SERIES II:- On "normal" skin of hands, right or left.

<u>Mode of infection.</u>	<u>CULTURE</u>	<u>Hands</u>	<u>Palm.</u>	<u>Dorsum.</u>	<u>No.</u>
stroked	{ not dried on	{ washed	†	†	7.
		{ rinsed	†	†	8.
	{ dried on	{ washed	†	†	9.
		{ rinsed	†	†	10.
rubbed	{ not dried on	{ washed	†	†	11.
		{ rinsed	†	†	12.
	{ dried on	{ washed			
		{ rinsed			
etc.					

REMARKS:-

In each experiment bouillon cultures of *E. Prodigiosus* and *E. Pyocyaneus* were employed for the infection of the skin, the one culture for the palm, the other for the dorsum of the hand, and these were alternated as also was the particular hand tested. All inoculations were made and incubated as in Series I.

"Stroked" implies that if the culture was not allowed to dry it remained 1 minute on the skin and was constantly gently painted over the surface to spread it and prevent drying. Subsequent washing or rinsing at the end of 1 minute. Then immediate inoculation. If culture dried on, inoculation made after about 20 mins. to ensure thorough drying.

"Rubbed" implies firm rubbing in of culture on the skin with the end of a test tube till dry (2-5 mins) or for 5 mins. till hand washed or rinsed.
 "Washed" implies 2 minutes washing with soap and cold water. Then rinsing off of soap under cold running water for 1/2 minute, then douching with sterilised water.
 "Rinsed" implies 2 mins. rinsing with cold running water. Then douching with sterilised water

would appear that the organisms had disappeared from some portions of the hand surface sooner than from others. In exp. 6 one single colony resulted in the plate inoculated from the palm 3 hours after infection.

The conclusions which may be drawn from these and the experiments of the other series are discussed later after all have been described.

2nd. Series.

As the results in the previous series had mostly proved negative this one was planned in order to demonstrate if possible what circumstances had contributed to effecting this early disappearance of the bacilli from the surface of the hands. Had the methods employed--the rinsing off of the culture immediately after infection, the subsequent rinsings or washings, removed the bacilli; did the mere stroking on of the culture not suffice to give the germs a hold on the skin or could forcible rubbing in of it make any difference; did it affect the result in any way if the culture was permitted to dry on the surface? These were questions which required solution. As each factor had to be considered in each case it was found necessary after a few experiments to draw up a scheme in which all the possible combinations would be tested so as to estimate their relative significance. This was arranged on the supposition that the bacilli were more likely to remain on the skin if they were rubbed in rather than being merely stroked over the surface, if they were dried on instead of being immediately rinsed off, and lastly if the skin was simply rinsed with cold water subsequently and not washed with soap.

The scheme is shown on Table II. If the culture was stroked on and not allowed to dry, it was kept a minute on the surface and constantly spread the while over the whole area with the pipette so that it did not dry at any point.

The hand was then immediately rinsed or washed as the case might be. The inoculation followed at once. If allowed to dry on 20 minutes was given for this. If the culture was rubbed in, the closed end of a test-tube was employed for the purpose using all force. Rinsing implied holding the hand under the running cold water tap for 2 minutes and subsequent douching with sterile water. By washing was meant the use of ordinary household soap and cold running water for 2 minutes, followed by $\frac{1}{2}$ minute under the cold tap to remove the soap and final douching with sterilised water; no brush was used nor was the skin rubbed with a towel. In all cases the gelatine was inoculated immediately after the procedure was at an end and the methods were in all respects the same as in the first series.

As the Table shows, the result in every case was a positive one. The number of the colonies is not given and it is obvious can have little relative value, but it may be said the growth was in every instance profuse and the plate was found sometimes to be completely liquefied within 24 hours with the characteristic colour formation from the active growth of countless colonies. Exp. 7 it is worthy of mention showed innumerable colonies throughout the whole medium inoculated from the palm, although in this case the culture of *E. Pyocyaneus* had only been in contact with the skin for 1 minute, had not dried anywhere, and had been immediately subjected to soap and water washing, that is to say the combination of circumstances which in this series may be supposed to have given the bacteria the least chance of adhering to the surface.

As the results however were all positive they do not admit of contrasting the relative importance of the six factors concerned as had been hoped. Some light was thrown on these points by later experiments to be presently described. It was not considered necessary to perform the last two experiments included in the scheme as all the preceding ones had proved positive.

SERIES III. Experiments on skin of hands, right or left, after application of
lanoline.

<u>Mode of infection</u>	<u>Culture</u>	<u>Hands</u>	<u>PPalm</u>	<u>Dorsum.</u>	<u>No.</u>
Stroked	{ not dried on	{ washed	†	†	13.
		{ rinsed	†	†	14.
	{ dried on	{ washed	†	†	15.
		{ rinsed	†	†	16.
Rubbed	dried on	{ rinsed	†	†	17.
		{ cleaned with aether and gauze	0	0	18.
Rubbed	dried on	{ cleaned with cold water and gauze	0	†	19.
Stroked	dried on	{ held in steam-bath and cleaned with gauze.	0		20.
Laid on (skin cleated)	not dried on	rinsed	†		21.
Laid on (skin decleated)	not dried on	rinsed	†		22.

REMARKS: -

Inoculations made and incubated as in preceding Series.
For detailed explanation of terms see Series II.

3rd. Series.

This set of experiments was carried out with the idea of elucidating the effect produced by the presence or absence of the fatty material which is always demonstrable on skin in its normal condition. The belief that the fatty covering of the skin has an important bearing on problems connected with disinfection is of long standing. It has been generally accepted that its removal in any process of disinfection is desirable as it collects and contains to a considerable extent the dirt and dust and therefore the germs of the skin. More especially in relation to cleansing and disinfecting methods with alcohol has stress been laid upon it, as the advantages resulting from the use of spirit have been attributed largely or entirely to its fat-removing properties (Fürbringer 44,50) Krönig 14). But the exact significance of the fatty material has been but vaguely understood as some writers rather appreciate the deoleation of the skin produced by alcohol because it permits other disinfectants to act upon the epidermis once the fat is cleaned away, than for any direct germ removing properties. This is in the main the view of the two authors mentioned as also of Haegler 11). Many observers attribute the unsatisfactory action of various disinfectants to their being unable to attack bacteria, surrounded as they are on the skin, by a fatty envelope, and again some have supposed that the fat in the ducts and follicles prevents disinfectants reaching germs lying in such situations. It is evident therefore that the relation of this fatty material to the skin germs has not been clearly defined.

In this series in the first place four experiments were made similar to those just described in the 2nd. Series, but in this case after the skin had been artificially greased with lanoline. One idea present in trying this procedure was as a control on the foregoing series, as it had been noticed that the hands were, although smooth, if

anything rather dry, probably as a result of frequent handling of bandages of plaster of Paris which has a marked effect in removing fat from the skin. The hands were washed clean and sterilised lanoline was then rubbed well into the surface, after which the surplus was wiped off with muslin till no obvious greasiness remained. This was sufficient however to leave an appreciable amount of fat on the skin as shown by the impossibility of getting water to lie evenly on it. The bouillon cultures were then applied as before, being either stroked or rubbed on the skin, and either rinsed off after a definite time or else allowed to dry on. On account of the greasiness of the skin the culture required constant stroking to get it to spread, but this may be considered as approximating to the condition of normal skin (unaffected by frequent mechanical disinfection) as most of the lanoline had been wiped away. The results however showed no difference from those of the preceding series, as in each case they turned out positive. But in the fifth of these experiments (exp. 17, 18) after the rinsing, the palm and back of the hand were cleaned by rubbing for about 2-3 minutes with gauze swabs soaked in aether with the intention of deoleating the skin, and the subsequent final rinsing with water proved that this had been accomplished as it now lay evenly on the surface. The inoculations made after this proved negative.

Exp. 19 was similarly performed, but after infection the two surfaces were each rubbed with frequent changes of plain gauze under cold running water. It was found that after about two minutes energetic rubbing the fat began to disappear and at the end of three the water lay evenly on the skin where treated though not elsewhere. In this instance the inoculation from the palm was negative but that from the back showed three colonies of *B. Prodigiosus*.

In Exp. 20 on the palm, the hand was held after infection in a steam bath for 5 minutes with the idea of promoting the flow of perspiration and the skin was rubbed frequently

the while with gauze. It is probable that the most of the moisture that collected on the palm was really condensed steam. This procedure sufficed apparently to remove the greater part of the fat and there resulted no growth of the bacillus on the Petri dish.

It was next desired to test comparatively the power possessed by the skin when fatty or otherwise of attaching to itself microbes which might come in contact with it. A quantity of a bouillon culture of *B. Prodigiosus* was laid on the palm of the hand and left there without any stroking or rubbing for 2 minutes. The palm was then rinsed in cold running water for 2 minutes and an inoculation made. In exp. 21 lanoline was rubbed in on "normal" skin and the surplus wiped off as in previous experiments of this series; in exp. 22 the palm was cleaned and deoleated with aether. The result was in both cases positive, the colonies in the latter being very much more numerous, but in the former the lanoline tended perhaps to produce some clumping of the epithelial scales brought away on scraping and thus diminished the resulting number of separate colonies, so that one can hardly deduce that the non-fatty skin necessarily collected more germs than the fatty.

4th. Series.

The experiments in this series were all made on the hand the skin of which by means of scrubbing and cleansing with aether had been freed as far as possible of its normal fat, a condition similar in fact to that of the surgeon's hands after disinfection preparatory to an operation.

In the first two (Exps. 23 and 23) the culture was rubbed firmly in with the end of a test tube for 5 minutes but not allowed to dry on, the hand being rinsed with cold water. While inoculations made immediately after the rinsing were positive showing that the germs had not been removed entirely (cf. exps. in Series II) subsequent inocu-

SERIES IV. Experiments on the deoleated skin of the hands, right or left.

No.	Preparation of Skin	Mode of infection	Parts tested	Bacillus used	Inoculations made after	3	6	12	24	36	48	72	96	hours.	
					In the interval		2	2	2	4	2	7		washings with soap.	
22.	L. Hand. Skin deoleated; scrubbed soap and hot water and brush; then rubbed with aether and gauze 5 mins. Then rinsed ster. water	Bouillon culture firmly rubbed in for 5 minutes. <u>Drying avoided;</u> hand rinsed at once with cold water, 2-3 mins.	Palm Dorsum	Pyocyaneus. Prodigiosus.			0 0	0 0	0 0	0 0	0 0	0 0			
23.	L. Hand. Skin deoleated as in 22. Subsequent rubbing with sterile water and gauze 3 mins.	As in 22	Palm Dorsum	Prodigiosus Pyocyaneus.	" "		0 0		4 0		5 0			ditto	
24.	L. Hand. As in 22	Bouillon cult. stroked over skin till dry.	Palm. Dorsum.	Pyocyaneus Prodigiosus.	" "		0 0		0 0		5 0	6 0		ditto	
25.	L. Hand. As in 22, but aether treatment for 3 minutes	Bouillon culture firmly rubbed in till dry.	Palm. Dorsum	Prodigiosus. Pyocyaneus.	" "		0 †	0 †	1 0	2 0		2 0		ditto	
26.	R. Hand. as in 25	As in 25.	Palm Dorsum	Prodigiosus Pyocyaneus	" "		0 †	0 0	0 †	0 0		0 †	0 0	ditto	
												65 cols.			
27.	L. Hand. As in 25.	As in 25.	Palm. Dorsum	Pyocyaneus Prodigiosus	" "				1 0	2 0	2 0	2 †	3 0	4 0	ditto
28.	L. Hand. Deoleated as in 25. Then sterilised lanoline rubbed into skin and the surplus finally wiped off.	As in 25.	Palm Dorsum	Pyocyaneus Prodigiosus	" "		0 †		5 0			10 0		ditto	

REMARKS: - See Series I.

SERIES V. Experiments on skin of arms and forearm under various conditions.

No.	Preparation of skin.	Mode of infection.	Part tested	Bacillus.	Inoculations made after	1	1 1/2	2	2 1/2	3	4	5	6	7	8	9	10	11	12	13	15	17	18	19	21	22	24	days.	
29.	Washed with soap. Rinsed with water.	Bouillon culture stroked over skin till dry. Infected area covered over with watch glass.	External as- pect of left and upper arm.	B. Prodig. Pyocy- aneus mixed	after 1/2	†Py.	†Pr.	0	†Py.	†Py.	†Py.	†Pr.																	
30.	As in 29.	Bouillon cultures rubbed on skin till dry. Covered with watch glass for 4 days, then removed.	Ditto right arm.	Ditto							†Py Pr	0	0				†Pr 2 cols.	†Pr 1 col.	0	0					†Pr 1 col.	0			
31.	Skin deoiled. Washed with soap and water; rinsed; rubbed with ether and gauze.	Bouillon culture stroked on skin till dry. Covered with watch-glass for 4 days, then removed	Ditto left arm.	Ditto		†Pr		†Pr			†Pr Py	†Pr Py		†Py		†Py Pr		†Py		†Py O*	0	0	0	0	0				
32.	Skin deoiled as in 31. Then pure steril- ised lanoline rubbed in and the excess wiped away till obvious greasiness removed.	Bouillon culture firmly rubbed on skin till dry	Ditto right arm.	Ditto		0	0				0	0	0	0	0	0				0									
33.	As in 32.	As in 32.	Radial as- pect right forearm.	Ditto				0	0		†	0	0			0													

* Between the two inoculations on the 13th. day in Exp. 31, the skin was cleaned for 3 minutes with gauze soaked in ether.

lations made from 3 to 72 hours after proved negative.

In the next experiment (24) the cultures were lightly painted over the surface but allowed to dry on. While the presence of the germs was of course demonstrable immediately afterwards, all later scrapings during similar periods gave negative results.

In three other experiments (25, 26, 27) the cultures were rubbed on the surfaces till dry which required from 3 to 4 minutes. Inoculations were made from 3 hours to 7 days thereafter. In each experiment no result was obtained from the plates inoculated from the palm, but from the dorsum of the hand positive results occurred up to 2 days (48 hrs.) after the infection.

In the last experiment of this series as a control the deoleated hand was treated with lanoline as in those of Series III. Positive evidence was obtained of the persistence of the bacilli both on the palm and the dorsum 3 hours after infection but not later than that. (Exp. 28).

5th. Series.

In this it was endeavoured to clear up several points of interest which had arisen in the course of the investigation; among others to determine, for instance, whether the negative results in other experiments were attributable to the natural death of the germs employed on a skin surface in the periods during which their presence was tested, further to estimate the effect of friction on such exposed areas as the hands present, and also to ascertain if it was possible for "accidental" organisms to become more or less permanent epiphytes of the skin. For these purposes the tests were made on the skin of the upper arm and forearm, and friction was excluded by covering the infected surface with a deep sterilised watch-glass affixed in position by means of sticking plaster. In this way the skin was protected for the desired time from rubbing, the effects of

any outward influences as air and from washing.

In Exp. 29 the skin was washed and a mixed bouillon cultures of B.Prod. and B.Pyocy. were then painted on and permitted to dry. The infected portion was then covered over as described and inoculations made every 12 hours up to 4 days (96 hours), the glass being temporarily lifted for this purpose. The results were except in one instance uniformly positive. The exception is probably due to the fact that only a very minute portion of skin was scraped at each inoculation (about 1/2-1 q.cm.) and in this case a part to which no germs had adhered was tested.

Exp. 30 was similar, but after the skin had been covered for four days the glass was removed and inoculations were made from the now unprotected skin surface at intervals of two or three days till 24 days had elapsed. The skin was now of course exposed to the effects of friction and it was washed with soap and water once a day. The results were in this instance more erratic, for while both Prod. and Pyocy. were present, naturally, at the end of the first four days, only 3 of the subsequent 8 inoculations were positive and the number of colonies appearing was limited to two or three of Prodigiosus, and in the last positive result on the 21st. day one colony was detectible. It should be mentioned that probably some of the negative results, at least the earlier ones, are to be accounted for by the irregular adherence of organisms to the normal (fatty) skin, for the bouillon did not spread on the skin of the arm so readily as on that of the hand and even when firmly rubbed on as in this case tended always to collect in droplets unless constantly spread.

In Exp. 31 on the other hand the surface was first de-cleated by washing and then rubbing with aether soaked swabs. The cultures now lay evenly on the skin and it was rubbed in till dry. After the glass was removed on the fourth day inoculations were made every second day up to 22 days; but on the 14th. day after the first inoculation the skin was cleaned with aether and gauze for three min-

45.

utes. The inoculation made immediately thereafter and all succeeding ones were negative, whereas all previous ones proved positive. The growth in this experiment was as compared with the one just described extremely profuse, the plates generally becoming completely liquefied with the characteristic colour formation within a few days. It is interesting to note that at first the B. Prod. was the predominant organism, while later the B. Pyocy. became so and overgrew all others. On the 9th. day for instance there could be counted some 350 colonies of B. Pyocyanus and only one area of red liquefaction of B. Prodigiosus, and on the 18th. day in the last plate previous to the aether cleansing there occurred a diffuse liquefaction throughout the whole of the gelatine from countless thousands of B. Pyocy. colonies. These scrapings were all made from but a minute area of skin (so that the scraping itself should not produce a premature removal of the bacilli from the infected surface), but the last one or two of each experiment was made from the whole area of skin.

The last two experiments which may be included in this series (32, 33) were made upon deoleated skin on which however lanoline had been rubbed in previous to infection, thus returning to it, as it were, its normal fatty covering. One was tested on the upper arm, the other on the forearm. They showed a complete disappearance of the bacilli even by the time of the first inoculation on the first or second day, except in one case when two colonies appeared in an inoculation made on the fifth day. In these two experiments the skin was not covered as in the others during the first few days.

Factors of importance with regard to the incidence and duration of bacteria on the skin.

Friction.	}	Normal mechanical removal
Washing.		affected by position of skin.
Sweat stream.	}	Removal by natu-
Growth of epithelium.		ral processes.

Condition of the skin:-

smooth or rough-
dry or moist (from sweat)-
hairy or otherwise-
fatty or deprived of fat.

Artificial removal:-

aether with rubbing	}	removal of fat.
water with rubbing		
perspiration with rubbing		

Drying on if on deoleated skin

Tenacity of life and resistance of germs.

Factors of apparently no importance.

Disinfectant action of normal skin.

Drying on on normal skin.

Stroking painting or energetic rubbing.

Removal by rinsing or washing without simultaneous friction

The conclusions which I would feel inclined to draw from the experiments described, I have placed in a tabular form indicating those factors whose importance has been indicated by the results. In another list I have placed such as have in so far as the experiments go apparently no significance, although they are factors which one might a priori be inclined to put stress on.

As will be seen the factors which seem to influence the incidence of germs on the skin are very numerous and an analysis of them is a complicated matter. I shall endeavour to elucidate them as far as possible considering them in turn.

One feature at once obvious on glancing over the different series is the striking contrast between the results shown by the hands and by other portions of skin as the arm. Under otherwise similar conditions the hands free themselves of germs very much more rapidly. The most outstanding difference between the conditions affecting them is in regard to the amount of friction and rubbing to which they are exposed. Therefore I would be inclined to place friction, the rubbing of the skin surface to which it is so constantly subjected in the course of our daily occupations, as perhaps the most important factor in removing the foreign germs. One would be inclined at first to suppose that washing with soap and water might account for this difference between the two skin surfaces. The experiments in the 2nd, and 3rd, series show however how little effect this really has. Friction and washing may however be classed together as the two processes of work which effect the mechanical removal of germs.

Another striking fact with regard to the hands themselves is that except with in perhaps the first hour or two the organisms have completely disappeared from the palm while they may be still present on the dorsum. I think that this is best accounted for by the stream of sweat which itself may carry off any germs. But it also no doubt produces this result by preventing the adherence of them to the surface. The palmar aspect of the hand is rarely quite dry for any

great length of time. It is possible also that Nature effects a removal of germs by the growth and casting off of epithelial scales. This may have more effect on the less exposed portions of skin, but on the hands germs are probably removed by other means before this has had time to take place.

The condition of the skin surface is a matter of the utmost significance. That a rough skin surface holds the germs longer than a smooth is a well recognised fact, and is probably because the germs lying under raised up epithelial scales are less exposed to the factors at work which have just been mentioned. But ~~And~~ the comparative roughness of the dorsum of the hand as compared with the palm is perhaps less the reason for the longer persistence of germs there than is the absence of a profuse stream of perspiration. That the hairs are supposed to attach bacteria to them very readily is generally believed, but my experience has not proved it to be so. In inoculations made from the dorsum of the hand usually numerous hairs were brought away, but a growth of germs directly from them in the medium was the exception rather than the rule.

The comparative dryness or moistness of the skin is important in determining whether organisms become attached firmly to its surface. But while the sweat as I have stated acts in this way, the natural fatty matter of the hand and skin generally acts also efficaciously. This probably explains the persistence of germs on the dorsum of the deoiled hand, while the palm though similarly treated freed itself so much sooner from infection. This observation in regard to the deoiled skin is of great significance for that is the condition of the operator's hand after disinfection, and there is usually superadded to this a roughening of the skin from the energetic treatment. It is interesting to note that the length of time which the bacilli persisted on the hand under these circumstances exactly corresponds to the average time required for abstinence to be observed by such authorities as I have above quoted, viz., 48 hours. But the abstinence time necessary on the basis of these experiments for the normal hand is nothing

like so much. In the case of the skin other than that of the hand the matter is different, but it must not be lost sight of. The experiments of Series V show how very much longer bacilli persist upon, especially, the deoleated skin in this situation. Perhaps this is to be accounted for by the more readily produced scabiness as a result of vigorous mechanical disinfection of the thinner skin. In exp. 81 the use of aether completely removed germs which had persisted for about a fortnight in spite of repeated soap and water washings. The action of the aether was probably to remove the desquamated scales which were giving the germs shelter. In this connection one may note that there was no indication that the germs had any real tendency to become permanent skin epiphytes. Yet according to Schimmelsbusch the *E. Pyocy.* is a normal tenant of the skin in certain situations, such as the axilla and the flexures. This observation would seem to show therefore that only specially favoured organisms can obtain a hold on the skin, although others as with the *E. Pyocyaneus* in this situation maintain a mechanical existence for a considerable period if not removed.

As concerns the drying on of organisms an interesting point is brought out, that while it makes little difference on normal skin it makes a considerable one on deoleated skin, and the natural instinctive tendency of the surgeon whose hands become infected during an operation to prevent drying has support from the experimental side. That it makes no difference on the normal hands, is probably because a layer of fatty matter intervenes between the germs and the epithelium, or else as in the case of the palm an early outpouring of sweat has the same effect. Again between the effects of rubbing in of infectious material and simple contact there would seem to be no difference as to the final results, which implies I suppose that it is difficult or impossible to produce a greater or less penetration of the germs.

With regard to the various methods of removal of the or-

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ganisms Series II and III show how ineffective soap and water washing, unaided by friction with some rough material, and still less so presumably simple rinsing, are in effecting the removal of germs. This suggests that the common practice of rinsing the hands at intervals during an operation is worse than useless, for it has the disadvantage of keeping the skin macerated which as already pointed out allows of a very much more ready separation of germs of the skin not already removed in the disinfection.

The explanation of why this apparently useless rinsing seems to affect the ultimate duration of germs on the hands (vide expts. 22 and 23) is not at once obvious. Perhaps it acts by maintaining the surroundings of the germs moist and preventing their immediate adhesion to the epidermis.

Exps. 18 to 20 bring out the value of a mechanical cleansing in removing germs from the skin which is protected by a fatty covering, and secures results which would be impossible by this simple means were the hands not so protected.

The question of the significance of the power of resistance of various germs is shown to be slight as far as the hands are concerned, for their early disappearance from that situation was proved by the experiments on the arms, not to be due to any death of the specific bacilli nor to any capacity of the skin or its secretions to unfavourably influence the life of these germs; and as for their eventual disappearance from the skin of the arm I found by control experiment that these bacilli were able to remain in existence apart from the skin (bouillon culture dried on sterile filter paper) for at least a very much longer period than their duration on that portion of skin.

The experimental investigation described above was carried out in the bacteriological laboratory of the Pathologisch-Anatomisches Institut under the charge of Professor Weichselbaum in the General Hospital, Vienna. To him and to Professor Ghon I am much indebted for the permission to work in the laboratory and for their kind interest in the progress of the work.

SECTION III.

The nature of the permanent skin epiphytes from the surgical aspect.

In the previous section I have dealt with the conditions affecting what I have called the temporary or foreign organisms of the skin. It has been shown that in all probability the majority of germs find but a comparatively short-lived existence on the skin surface and only under exceptional circumstances obtain a prolonged ^{hold} upon it. That any organisms at all, other than those of a very limited variety, become permanent epiphytes is extremely doubtful. The explanation of this is not so easy and the question does not lend itself so readily to experimental investigation as that concerning the temporary skin inhabitants. It has been indicated in the foregoing section how great an influence friction has in removing organisms from the skin and how important the skin secretions are in preventing or limiting an invasion by them. One may almost imagine that the flow of these secretions forms a constantly streaming tide, which drives away before it any would-be invaders, while, behind this tide as it were, there exists undisturbed an army of bacteria of a specially favoured kind, which for reasons to be presently considered are able to maintain a permanent colonisation of the skin. It is conceivable that this stream may also in some degree hinder the egress as it undoubtedly checks the ingress of germ life.

Inoculations made from skin have given me growths of an enormous variety of organisms, including moulds of many kinds, white and pink torulae, sarcinae of various kinds, bacilli of putrefaction, bacillus coli, bacillus subtilis, streptococcus pyogenes, staphylococcus pyogenes albus, citreus, and aureus, cereus albus, cereus flavus, and many others which were not without more extended investigation identifiable. It is of course patent that an illimitable choice of germs may be found, depending of course on what



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the environment may happen to be, but out of the many kinds I have only found representatives of one kind constantly and that is staphylococci. Except in cases of cultures containing some very rapidly growing organism, as for instance in my own experiments on infection with B.Prod. or B.Pyocy., I may say that I have invariably found staphylococci present and usually in large numbers. It is a fact that soon becomes apparent in making experiments on the skin that the more it has been cleaned (mechanically disinfected) the more limited is the variety of organisms to be obtained from it; and if the surface has been cleansed with the greatest mechanical thoroughness possible only staphylococci are to be found, if any growth at all results. This demonstrates quite clearly that these have a deeper situation in the skin and a more constant existence there than any other forms of germ life. We may therefore take it that they are alone to be regarded as the natural or permanent epiphytes of the skin.

Why this should be so is far from clear, that is to say it is not at once evident why they should not be affected by the conditions which are found to affect other types of organism. Certainly it is not to be expected that any germ would find the circumstances surrounding it in the skin suitable for its own existence, not to mention its multiplication: the skin surface is acid, unless perhaps this acidity is neutralised by an excessive flow of sweat (Heuss 52), and the secretion from the sebaceous glands, while not perhaps inimical, is at least not favourable to germ-growth. Gottstein 53) found that germs died out in lanoline. Haegler (loc.cit, p.21) found that while no increase of various germs tested took place in lanoline, in some cases they remained alive for months, and he also found that staphylococci failed to develop on the surface of comedones. It is obvious then that organisms must have a considerable power of resistance and of adaptability in

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order to maintain an existence on the skin. This tenacity of life staphylococci of course possess in the first degree; still there are other organisms of great resistance which perhaps may be even better suited to the environment, which however are not known to take up their abode there.

Two explanations present themselves to account for this predominance of the staphylococci. One, that these bacteria are constantly being brought to the skin from the atmosphere and the general surroundings and that they in virtue of their powers of resistance perpetuate their existence longer in an environment which is sooner or from the first fatal to other kinds. The other, that they on account of their greater adaptability are able to reproduce themselves and from the time of their first arrival ~~at~~ constantly form new generations, thus appearing as permanent inhabitants of the skin. The former theory naturally implies that any one coccus though remaining some time is eventually cast off as the epithelium grows and the scale to which it has become attached is loosened and removed. The latter postulates an existence independent largely of the casting off of the epithelium. Both propositions present difficulties. In the former case it is difficult to understand the indisputably deep situation in which the cocci are to be found. The commonest coccus or other organism on the skin would be that most commonly present in the surroundings. That these skin cocci are not of a single kind as is so generally accepted I shall have occasion to point out later. In the second explanation the chief difficulty is that of the multiplication of the organism and evidence is wanting to prove that this takes place.

Many authorities deny that under normal circumstances any increase occurs. Haegler (loc.cit.pp.20-26) denies that germs lying in the orifices of the hair follicles can multiply. He examined microscopically skin into which cultures had been rubbed immediately before and at intervals of 24, 48, or 72, hours before its removal from the body. Contrary-

ting the results in each, he found that in the latter cases the number of contained germs was very considerably diminished or negative. He concluded that they were therefore to some extent removed by the stream of secretion. That germs are as little able to grow down into these openings of the skin he deduced from the fact that virulent yellow staphylococci kept applied to the skin on moist wool compresses covered with guttapercha tissue failed to produce impetigo or furuncle. This of course assumes that such must necessarily follow from the mere presence of these in the hair follicles, which is not perhaps altogether certain. But microscopic sections made from the skin after 24 hours application of such cocci failed to show any downward growth into the follicles. With regard to the germs which may be present on the surface of the epithelium or between the partly loosened horny lamellae, he assumes that if suitable nutrient material is there, such as pus or blood or other organic substances, and is kept in a moist condition on the hands for a considerable time, there is nothing to prevent a multiplication: a constant profuse perspiration, principally by producing a maceration of the epidermis, may also permit of germ-growth. He tested this by inoculations made from the well cleansed hands covered for 48 hours with sterile rubber gloves. While in the first 4 hours the increase of the germs was insignificant and principally due to deeper ones reaching the surface, after 10 hours the number had increased many thousand times (45,000), and after this period the growth was so rapid that within 2-3 hours 30-40,000 germs were obtainable by the inoculation, even though the hands had been thoroughly cleaned in the interval. It was then evident in his opinion that like those of the atmospheric dust the germs required a period to elapse before they were able to multiply, and therefore in all probability the bulk of the germs on the hands come directly from the dust and dirt of our surroundings. He

considers that on the hands of a cleanly person and especially on those of a surgeon a multiplication of germs is not probable, as the frequent washing deprives them of nutrient material and the surface is usually dry, conditions in fact which are unfavourable to the growth of germs.

If these views are accepted then the second of the theories put forward is not tenable, and we must accept the idea that all so-called permanent skin germs are in a constant state of renewal from without, as they are also constantly being detached from the skin.

If this is so then it is hardly permissible to believe as so many do that there are organisms on the skin which are of so constant a character as to warrant a special designation, as for instance the Staphylococcus Epidermidis albus described by Welch, or the Staphylococcus Gilvus described by Bossowski, as well as certain bacteria associated with some diseases of the skin; for such constancy of character as is attributed to these organisms is not to be accounted for if they are one and all derived from the environment. It is unfortunate that, in the description of the experiments on the hands just referred to, Haegler did not state the nature of the germs which appeared in such numbers under the rubber glove, but it is important to note that these observations of the similarity of the mode of growth of these organisms with that of the dust germs, concerns especially the hands, and it is conceivable that the external circumstances affecting them are not quite the same as those affecting other portions of the body surface: the more covered skin surfaces are not so directly exposed to the drying action of the air, nor are they subjected to such frequent attacks with soap and brush as to remove to the same degree the materials required for germ growth, and lastly as a result of these artificial and of normal conditions the epithelial covering of the skin is considerably thicker on the hands. It is however desirable to consider the general skin surface as well as the hands in dealing with the question from a practical point of view, as the skin of the patient at the site of operation and also the

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skin of the arms of the operator enter into the problems requiring solution.

With the hope of obtaining further light on these matters I carried out a series of experiments based on the idea that if no multiplication of the germs in depth takes place on normal skin, and such germs as are present can only reach it from outside, then if one protects the skin surface from renewed infection it must eventually free itself from them in the process of the growth of epithelium carrying the attached microbes to the surface. If now the shed epithelial scales were removed as they were cast off, then the skin must in process of time become sterile by this sort of auto-disinfection. If on the other hand it were found that the skin did not sooner or later become germ free, one would be entitled to come to the conclusion that a multiplication of the germs must be taking place in its depths.

A whole series of experiments were accordingly performed on this principle. The skin usually of the arm or leg was cleansed well by purely mechanical means, washing it with soap and water and finally cleaning it with aether on sterile gauze followed by a douching with sterile water. The procedure was carried out on the strictest possible aseptic lines to avoid reinfection of the skin surface. After the cleansing process the area experimented on was covered over to exclude air infection with either a sterilised watch glass or if a larger portion of skin was tested with sterilised protective-silk, in one case lanoline was tried as a covering. Inoculations were made at intervals of usually two or three days, one immediately on removing the covering and the other after the skin had been once more cleansed so as to remove all desquamated epithelial squames, and at the same time such bacteria as might have been brought to the surface or might have multiplied there in the preceding interval. This procedure was repeated again and again till the experiment had lasted over a period of up to

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in some cases 17 days, within which time it was thought it might reasonably be expected the epithelium had grown up to the surface from the deepest level at which bacterial epiphytes might be present. In no case was it found that even after the lapse of this time sterility had been attained. The results in each succeeding inoculation were indeed wonderfully constant, and allowance must be made, in comparing numerically the colonies present, for the impossibility of getting the same effect at each scraping of the skin with the knife, which, as in former experiments, was employed for making the inoculations. Of course uniformity in this respect was aimed at as far as possible. From an area of skin covered with a watch glass as a rule from about 90 to 200 colonies resulted from the inoculation preliminary to the scraping. The second inoculation in some cases gave but a very small and almost disappearing number of colonies. That is to say that within an interval of perhaps two days one or two hundred staphylococci had appeared on the surface of this small area. I specify staphylococci as the germs present as after the first cleansing of the skin no other germ forms appeared. The small number of colonies in the second inoculations cannot be taken as an indication that the skin was becoming sterile below the layer of shed epithelium on the surface, as it was found inadvisable to scrape the part too firmly lest excoriation be produced, and the number of epithelial cells brought away with the knife was comparatively very small once the surface had been thoroughly cleansed with gauze and aether. So far from the colonies diminishing as time went on an increase was rather observable, but as already explained I should not care to lay too much stress on this numerical difference.

These results were rather surprising and seemed to point fairly conclusively to the possibility of the epiphytes reproducing themselves at depth, and as the skin very rap-

idly became dry and somewhat slightly scaly on the surface as a result of the cleansing with aether, etc., and consequent removal of fat, there was no reason to suppose that the conditions at this level were favourable to germ growth. In case however the scraping might have resulted in microscopic abrasion and effusion of serous fluid which would provide nutrient material for the organisms, further experiment was made, but in this instance the intervals between the inoculations were prolonged to about a week each, so that no irritation of the skin could occur from too frequent processes of cleansing. The result was however similar; no evidence was forthcoming that the skin could free itself of organisms in the absence of infection from outside.

I felt myself therefore forced to the conclusion that the regular epiphytes of the skin were permanent in virtue of their capability of reproducing themselves in its depths and so persisting in spite of the fact that many were carried to the surface and there cast off along with the epidermic squames. This being so, one can understand how there is a certain uniformity in the character of the constant skin organisms even on the hands of the surgeon, frequently cleansed and scrubbed though they be.

These observations throw an interesting side-light on the preparation of the skin of the patient for operation. Even repeated mechanical cleansing of the skin fails to rid it of germs, excepting those which may happen to lie on the surface, and on the tenderer portions of skin which are those most commonly operated on, e.g. the abdomen, a too thorough mechanical treatment some time before operation may indeed lead to an actual increase of the cocci, instead of a diminution, especially if, as is almost inevitable in such a case, microscopic abrasions of the surface have been produced. Nor can one look for much assistance from the application of antiseptic coverings in view of our present extended knowledge of the lamentable inability of antiseptic agents to act on the deeper parts of the skin. Still worse

results are only to be expected if the skin is kept moist and macerated by the preliminary application of wet "soaks" of any kind.

With regard to the nature of the cocci which form the permanent epiphytes of the skin, I have already mentioned above that I have met in various inoculations with all the varieties of Staphylococcus usually described. While I referred to them under the several names under which they are commonly classed, I must here state that they very frequently presented differences in their culture growth which led to great difficulty if one attempted to give them a definite description, as the classification being based upon colour distinctions I have met with every conceivable shade of colour in staphylococcus colonies, varying from the purest white to the richest orange, those in the interval showing all varieties of buff, primrose or lemon yellow, yellowish and reddish orange, so that in many cases it would have been impossible to say whether a colony should be described as albus, citreus or aureus. This was also the fact with the non-liquefying colonies on gelatine which fall under the heading of Staphylococcus cereus, all shades between white and yellow being found.

I have in certain series of experiments come across staphylococci growing in colonies differing essentially from any of whose description I have read, and which failing more extended investigation of their characters I am unable at present to describe in detail. Yet these cocci appeared with regularity throughout a whole series of inoculations and in more than one experiment, suggesting that these might also be considered as permanent epiphytes. Did these variations appear at different times in different inoculations one might justly attribute them to some difference in the nutrient media, but in one and the same culture plate one might often find colonies presenting widely divergent characters. It was found that gelatine gave the

most characteristic growths of these various staphylococci; on agar it was less easy to note variations and it was indeed found that there was really less readiness of development of colour on agar media. Gelatine is of course more valuable in that the liquefying powers of various colonies may be compared. In this respect too it should be stated that great differences were noted among colonies not otherwise distinguishable growing on the same plate.

That there should be such great difficulty in differentiating staphylococci from their mere cultural appearance, makes one wonder if any hard and fast line can be drawn as a distinction between different kinds. Haegler has also called attention to this question (loc.cit.pp.183-4). He remarks that anyone who has had much to do with examining pus and suchlike must have noticed that between the pure white and the intensely golden yellow coloured colonies of staphylococci there exists an unbroken series of so fine nuances of colour that a classification according to this is often difficult, and if one is to go by this one must recognise not 3-4 but 10-20 varieties. Lubinski 54) has produced a lasting growth of white colonies from a race of yellow staphylococci after he had grown several generations anaerobically, and Haegler had obtained similar results from the effects of chemical antiseptic agents. To return to the white cocci their original colour proved much harder as was found by Bertoye 55), Courmont 56), Rodet 57), and Netter 58). Haegler however succeeded in doing this, recognising that as the golden staphylococcus is essentially an organism associated with the human body. He rubbed into the skin of his thigh or forearm white staphylococci partly obtained from normal skin, and from the resulting inflammations was able to cultivate colonies with all shades up to an intense golden colour. This would seem to demonstrate fairly conclusively that no sharp line can be drawn between the different varieties. It may of course be said that the

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growth colour is indicative of the virulence of the organism as the aureus is more virulent than the albus, but while the former is certainly more frequently present in pus than the latter, it fails to explain the difference in virulence between the latter and the Staphylococcus Epidermidis Albus of Welch.

I have already mentioned the generally accepted belief in the prevalence of the white staphylococci as the commonest form to be found on the skin. The conclusions of Welch (59) who worked up this question I will here quote. "A coccus which may appropriately be called the staphylococcus epidermidis albus, is a nearly if not quite, constant inhabitant of the epidermis, lying superficially and also deeper than can be reached by present methods of disinfection of the skin. This coccus is found frequently in aseptic wounds. It may be the cause of disturbances, usually of a relatively slight degree, in the healing of the wound, especially when drainage tubes are inserted. It is the most common cause of stitch abscesses in wounds treated antiseptically or aseptically." He considered that this coccus was to be distinguished from the staph. pyogenes albus of Rosenbach of which it was perhaps an attenuated form. It was characterised by much greater slowness of liquefaction of gelatine and of coagulation of milk, and by far less virulence than the aureus when inoculated into the circulation of rabbits. He stated that it may be present in graver suppurative inflammations, but that then it was nearly always associated with some other pyogenic organism, or has assumed the form of the typical staphylococcus pyogenes albus.

In the writings of many workers on hand disinfection such a white coccus is frequently referred to, eg. Schumacher 4), Lauenstein 13), and it is seldom credited with much virulence.

It is quite likely that in many observations the absence of colour in colonies may have been referable to antiseptic substances employed in the investigations. For my own part

I cannot substantiate the belief in the uniformity of the character of all the apparently permanent staphylococci of the skin. The numerous inoculations I have had occasion to observe from many varying portions of skin show a wonderful variety in the appearance of the colonies presented with regularity in a whole series of cultures. These have all been made from skin unaffected by antiseptics so that the results could not be prejudiced by such means. In many cases I have found yellow (not golden) staphylococci in very large numbers with perhaps complete absence of any white cocci, and the gelatine was rapidly liquefied by them. In plate cultures showing numerous white cocci great differences have been observed; some colonies have grown rapidly to a fairly large size and have produced early liquefaction, while others of identical colour have remained minute and only slowly softened the surrounding gelatine. These larger more vigorous colonies have in such cases usually been in the minority, but while no microscopic difference was detectible there were apparently two quite distinct varieties of white coccus growing simultaneously. Of other sorts there have been found the most diffuse shades as I have already described, but while in the absence of contact with pus aureus colonies were not so uncommon, there was apparently no constancy in their presence in any series, so that one was not justified in looking on them as representatives of permanent epiphytes. With regard to variations according to site from which the plates were inoculated, there is little to be said except that from the hands, more especially the flexor aspects, tinted colonies were relatively uncommon and from the palm the white coccus was found to predominate, and it seemed that the number to be obtained from it was distinctly smaller than from the dorsal surfaces or other parts of the body.

It is possible that the yellow tinted coccus so commonly met with, corresponds to the *Staphylococcus Gilvus* of Bossowski (60), which to quote Welch, seems to bear

about the same relation to the pyogenic yellow staphylococcus which our epidermal white coccus does to the typical pyogenic white staphylococcus."

I am inclined to conclude on the basis of what I have stated above that there is not one but several varieties of staphylococcus settled on the skin as permanent epiphytes, which probably are indeed but modifications of a general staphylococcal type, but whose constancy of character is to be explained by the constancy of the nature of their environment in which they are capable of multiplication and in succeeding generations perpetuate their own special peculiarities.

But of whatever nature these various races may be, there would seem to be but little reason to doubt that one feature common to them all is their slight virulence, during such time, at any rate, as they may exist on the skin.

SECTION IV.

On the value of the use of lanoline as a protection
to the skin of the operator.

In the course of the foregoing investigations one thing has been made specially prominent and that is the wonderful power the skin seems to possess of protecting itself against any permanent bacterial invasion, in virtue of the secretions with which it covers its surface. This has been brought out both in the course of the experiments on "normal" skin and on skin which had its natural fatty covering replaced by the nearest practically available substitute for it, viz., lanoline, even when the surface had been subjected to a process of deoleation. Indeed the perhaps greater amount of fatty matter present on the skin in the latter circumstances seemed to result in an earlier removal of organisms than it was naturally able to attain when unaided by the artificial oleation.

On the other hand it has been made evident that germs may persist on the skin for a great length of time if reaching it when deprived of its natural protection, especially those parts less exposed to the effects of friction.

As this condition is produced on every occasion on which the operator thoroughly disinfects himself, the obvious practical conclusion which one must draw is that he should should if possible protect his skin by some artificial means from intimate contact with organisms. This is not only of importance in regard to the handling of such visible infectious materials as pus or the secretions from the septic canals of the body, but also in regard to the not only possible but probable though less observable infection from the atmosphere or already deposited dust of hospital wards or theatres. The work of Haegler and others have shown shown this to be equally important.

The protection of the hands from infection is a comparatively new development in the practice of aseptic surgery, and it is interesting to note that the use of such protec-

tive coverings of the hands during operations, as rubber gloves, has really developed out of the surgeon's desire to protect the operation wound from infection coming from his "disinfected" hands.

The surgeon who in this respect does not consider prevention better than cure, of necessity puts an implicit trust in his capacity for disinfecting his hands. As so much experimental research has shown, this is not to be relied on with certainty, though perhaps the removal of accidental infection is more readily attained than is that of the permanent skin epiphytes. But in any case the fact remains that the more the hands are disinfected, the more probably infective they may become, as a result of the more aggravated roughening of the skin surface.

The prophylactic protection of the hands can however be completely secured for the time by the use of rubber gloves. But the protection only exists for the period during which they are worn, and this is most commonly limited to the carrying out of septic operations. As soon as they are discarded the hands are again exposed to infection from any of an infinite number of sources. What is also significant is that while the wrists, forearms, and elbows are disinfected and deoleated, they are afforded no protection by the gloves, and not ^{only} are they thus specially liable to infection but they are less easily disinfected as they cannot, even if it is always kept in view, be submitted to such a thorough mechanical treatment as the tougher skin of the hands.

The disadvantages of rubber gloves from an operating point of view are familiar to those who use them and several substitutes have been suggested to more conveniently replace them, all of them as with the gloves themselves at first, proposed in order to prevent the separation of organisms from the hands and consequent wound infection during operation.

Thus Schleich 61) recommended a wax paste which he claimed, used in conjunction with his marble soap, would occlude the orifices of the skin glands and so prevent the escape of germs from these sources

Unna 62) suggested leaving a layer of soap on the hands after washing to prevent the separation of epithelial cells and attached bacteria.

Menge 63) proposed paraffin dissolved in xylol.

Haegler (loc.cit.cap.VI) tested these devices experimentally and also tried a solution of gutta percha, but came to the conclusion that all were unsatisfactory. The wax became emulsified and removed by the alkaline fluids in the wound, the paraffin and the guttapercha cracked too readily or were rubbed off. The paraffin was if anything the most reliable. But none of them was capable of preventing entirely the separation of germs from the skin.

If they were as compared with rubber gloves unsatisfactory for this purpose, one may presume they would be equally unsuited to protect the surgeon's hands from the patient, and which in view of the slight virulence of the deep skin germs may really be considered of more importance than the protection of the patient's wound from the surgeon's hands.

Arguing from my own experiments it struck me that the best procedure in this direction would be to imitate nature as far as possible, and that therefore the use of lanoline as the nearest approach to our own skin fat must be the best possible means of protecting the skin. My experiments demonstrated that unaided natural causes freed the skin from infection very rapidly if after mechanical disinfection it was covered with but a thin coating of this agent, whereas if not so protected the germs persisted for an enormously longer period. No doubt this result could be hastened by active cleansing and disinfection, still the advantage of avoiding the necessity of this if possible is obvious.

If the operator is to make use of this safeguard it ought to be applied to the skin as soon as the process of disinfection preparatory to an operation is at an end. If now the hands may become infected all that is necessary is a removal of the lanoline which will bring away with it the adventitious bacteria. The lanoline has also this advantage that not only does it prevent the adhesion of germs to the cells of the skin but infectious material, which is usually of a fluid nature, tends to run off from a surface covered with it. It forms a uniform coating which is only with difficulty rubbed off, and which does not crack in places as do most other substances used in this way, and it fills up all the crevices and grooves in the skin. At the same time it is the ideal application to the skin deprived of its fat and saves it from becoming dry and fissured. It does not make the hands slippery, in fact they are rather less so than otherwise and this is a distinct advantage during an operation, and of course in contrast to gloves the tactile sensation is in no way interfered with.

It must of course itself be sterilised, and this rather improves it for the purpose, as it renders it anhydrous and it is then firmer in consistence and not so greasy as in the ordinary state.

If it is to be used as a protection to the hands etc. against infection it is naturally desirable that it should be capable of easy removal if for instance it has been brought in contact with pus. With this object in view I carried out a series of experiments in order to ascertain by what means it could be most quickly removed.

The hands were thoroughly scrubbed as for an operation and treated with aether or bran to remove all fat from the skin. Sterilised lanoline was then rubbed well in and the surplus wiped off till hardly any was visible. The whole contents of a bouillon tube containing actively growing culture of B. Prodigiosus and then another of B. Pyocyaneus were rubbed all over the hands till they had dried on.

The hands were then treated in various ways in the different experiments and finally inoculations were made from the palmar surface of the hand and fingers, and from the dorsum of the hand and fingers and the inter-digital aspects, of first one hand and then the other. A sterilised knife was used for the purpose and the whole surface of the hand and fingers was tested on each occasion.

To effect the removal of the lanoline reliance was placed on the use of bran which has a striking power of extracting the fat from the skin. The bran was employed for washing as with soap, and aided in some cases with a nail brush or gauze swabs, and during the process the hands were held under a tap of running warm water.

Three minutes washing failed to completely remove the germs from the hands when the bran alone was used, when prolonged to five minutes, of the four inoculations one, from the palmar surface of the left hand was sterile, the other three plates showed 3, 2, and 2 colonies. Better results were obtained in three minutes if the washing with bran was aided by the use of gauze or a nail brush. The best results were obtained by the use of a soap composed of bran mixed with soap in an excess of water and made somewhat alkaline. With this alkaline bran soap the deoileation was rapid and satisfactory and, after treatment with it and gauze for five minutes, sterile results were obtained.

As will be seen this constitutes a very severe test for but rarely in practice are the hands infected to such an extreme extent as is represented by the rubbing in of a pure culture over the whole of both hands and fingers till dry. The results must therefore be considered satisfactory for by no other such simple procedure could hands be disinfected in the ordinary way and that without any injury to the skin. The process of course simply consists in a removal of the lanoline which brings along with it the contained germs. It does not probably act on any more deeply located germs but as there is every reason to suppose that

the lanoline prevents the deeper penetration of bacteria, the desired object, is attained.

After this treatment the skin is of course as liable as before to the risks of infection and the sterile lanoline should be reapplied.

Further experiments are being made with a view to improving on the method of deoleation and disinfection though it is hardly to be hoped that it can be shortened with safety.

It is another possible benefit obtained by the use of lanoline that it may hinder deeper organisms being given off from the hands during an operation, but this has not been tested by experiment so far.

SECTION V.

The microscopical evidence as to the localisation of bacteria on the skin under certain circumstances.

Section V. Table of microscopic preparations made.

- A. Skin from middle line of abdomen, elderly male corpse, moderately hairy.
 - 1. Normal skin.
 - 2. Normal skin rubbed 5 minutes with the end of a test tube, skin moistened with water.
 - 3. Normal skin rubbed 5 minutes in similar way with mixed bouillon cultures of B.Pyocyaneus and Prodig.
 - 4. Skin deoleated by rubbing 5 minutes with aether and gauze; then treated as in 3.
 - 5. Skin deoleated as in 4; then treated 5 minutes with alcohol and gauze; then cultures rubbed in as in 3.

- B. Skin as in A, but from just above pubis, very hairy.
 - 1. Normal skin rubbed 5 minutes with glass rod and surface of agar culture B.Pyocy. mixed with water.
 - 2. Skin deoleated as in A 4; then treated as in B1.

- C. Skin from middle line of abdomen, female corpse, slightly hairy.
 - 1. Normal skin rubbed 5 minutes with surface of agar culture mixed with lanoline.
 - 2. Skin deoleated with aether as in A4, then treated as in C1.
 - 3. Skin deoleated with aether and alcohol; then treated as in C1.

- D. Skin from middle line of abdomen, male corpse, rather hairy.
 - 1. Skin washed clean, then lanoline well rubbed over surface and then surplus wiped off. Then agar growth of B.Prodig. and B.Pyocy. rubbed in with glass rod.

- E. Skin from axilla. Hairs extracted.
 - 1. Agar surface growth of B.Pyocy. and B.Prod. rubbed in fairly firmly.

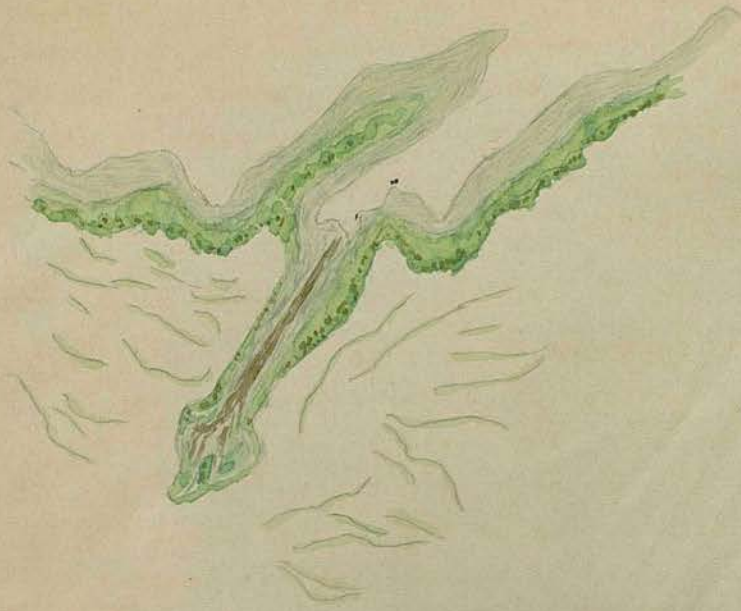
- F. Skin as in E.
 - 1. Surface scarified by scratching with a sharp pin. Then agar surface growth of Staph. Aur. B. Prod. & Pyocy. rubbed in.

With the object of correcting or confirming the results obtained by the experimental methods described in the previous sections, a large number sections of skin treated in various ways was examined microscopically. The skin was obtained from recent corpses and was then dealt with in the manner detailed on the Table. In most instances from about 30 to 100 sections were made and as far as possible serially. After numerous tests it was found that for investigation of the bacterial incidence in or on the skin the best and simplest results were obtained by staining the paraffin sections with carbol-thionin blue, and after thorough washing in water staining with a 2% solution in water of methyl green. Any bacteria retained the dark blue staining of the thionin and the nuclear elements of the tissues took up the bluish green colour of the methyl green. By this means the stained organisms stood out quite distinctly from their surroundings, and as the horny layer of the skin was not deeply coloured but only very slightly and delicately tinted, there was no obscuring of any germs present in this region.

A1.

The sections made from this normal skin, untreated in any way, showed but a sparse number of organisms in any one section. The predominant forms present were cocci occurring usually isolated but not infrequently in diplococcal form. They seemed to lie for the most part on the free surface of the epithelium and were often enclosed in such debris as was present on the skin, and which was often found to be collected in the grooves and furrows.

In no instance were any organisms detected in the sweat gland ducts. In the hair follicles they were but occasionally to be detected near their orifices. The majority of sections implicating the follicles were apparently free from germs.



Sketch of a section in Series A2 showing position of germs in a hair follicle.



Sketch of a section in Series B1 showing position of germs lying on wall of a hair follicle.

A2.

The conditions here were much as in A1. No deeper penetration of organisms was detectible, but, as in the previous case, the total number of them present was but small. A sketch made with the camera lucida eyepiece of the microscope will give an idea of the position of germs in the hair follicles and their scarcity in this situation.

A3.

As would be expected in this case the number of bacteria present was considerably increased as the result of the artificial infection. There was no evidence of increased penetration of the bacteria into the depths of the skin. Possibly the fatty matter of the skin hindered the entrance of the germs contained in a watery medium (bouillon).

A4.

As a result presumably of the treatment of the skin with aether and gauze with the object of removing fatty matter, it was found that the stratum corneum was in many places separated off at the level of the stratum lucidum. In such cases as is to be expected the organisms were to be found lying in direct contact with the deeper exposed layers, i. e. the stratum granulosum. It is probable that this separation occurred rather more readily in the skin of a corpse than it would in living skin, still as I have had occasion to point out before the skin of such portions as here on the abdomen have but a comparatively thin and delicate surface epithelium is easily abraded. Organisms then reaching the deeper strata would probably find here a suitable nidus for development, and may perhaps remain protected by the separated but still overhanging epithelium.

In the mouths of the hair-follicles the surface sometimes presented a broken up appearance as a result of the friction but this action did not seem to reach very deeply, at any

rate the contents of the follicles were not removed and, as the fatty matter in the deeper portions was probably also undisturbed, the organisms were not found at a deeper level here than in the previous cases. None were detectible in the sweat ducts.

A5.

I have previously referred to the action of alcohol as observed on the skin and the orifices of its ducts by Haegler, and in this case the skin examined had been treated with alcohol as well as aether. That little difference was to be seen between these and other sections is probably to be accounted for by the subsequent immersion of the portion of skin in alcohol to fix it having produced a similar result in all the sections examined.

B1 and B2.

The purpose of this examination was to ascertain if the nature and medium of the infecting agent made any difference in the penetration of germs. In this series the infection was produced by means ^{of} an agar growth in a watery mixture. As might be expected the organisms were more richly present than when bouillon was employed. The penetration of germs in hair follicles seemed to be deeper than in series A, but it is possible that this fact is to be attributed to the follicles of the skin being considerably larger as the hairs in this situation, just above the pubis, were much larger and more numerous. The skin showed in the sections a considerable accumulation of debris, which commonly though not always filled up the folds. The bacteria were contained in large numbers in this apparently fatty detritus and by its intervention prevented from reaching the epithelial surface. It was noted that the germs were often caught against the upper margin of the skin folds without reaching to the bottom of it. Here also I was unable to convince myself of the presence of germs in the sweat ducts.

C 1, 2, and 3.

In this series the bacteria were rubbed in contained in a fatty medium, the agar surface growth being mixed with lanoline. The germs in sections were found to be commonly aggregated together and very many lying at a distinct distance from the epithelial surface. In the folds of the skin, which were very numerous, was always to be found fatty material mixed with debris and containing numbers of organisms. It was difficult to say if the fatty medium allowed the bacteria further penetration as the hair follicles were in this skin comparatively small.

D1.

The skin had here been treated first with lanoline preliminary to the infection. The germs seemed to be usually contained in fatty matter and lying many of them at some distance from the surface, as if the lanoline had probably held them off from it. On the more prominent or superficial aspects of the skin margin, the bacteria appeared to be lying in contact with the epithelium. As far as one could judge from such sections, the lanoline seemed to protect the surface from the access of the germs, but as of course the bulk of the lanoline was wiped away it was impossible to say from the microscopical appearance if a layer of it, perhaps extremely thin, had at all points intervened between the germs and the stratum corneum. In any case it does not invalidate the arguments in favour of the methods proposed in the preceding section (IV), as any bacterium even if in seemingly close contact with the skin would inevitably be enveloped in fatty matter and thus prevented from becoming firmly adherent to the surface, and should therefore be readily removed in the cleansing. u

E1.

From this portion of skin taken from the hairy axilla the hairs were extracted before infection. In sections the

organisms seemed to have reached a distinctly deeper situation in the hair follicle but were not found in any very large numbers

F1.

These sections afforded a confirmation of the observation of Haegler of the enormous collections of germs which were to be found in accidental scratches or abrasions of the skin. This state of affairs was probably largely due to a growth of organisms in these clefts; my own sections showed, apart from any growth of the germs, very large accumulations of them as far as the innermost recesses, following rubbing in of pure cultures. It was noted that the cleft produced by the scratching was almost always bifid at the bottom like an inverted Y.



The investigations described in Sections III, IV, and V were carried out in the Surgical Laboratory of the University of Edinburgh and I am gratefully indebted to Professor Chiene for kind permission to make use of it for this object.

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