

DIPHTHERIA CARRIERS WITH PARTICULAR REFERENCE
TO THE TREATMENT OF THE CONVALESCENT CARRIER
WITH DETOXICATED DIPHTHERIA VACCINE.

by

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One of the most difficult problems to deal with in Fever Hospital practice is that of the convalescent diphtheria carrier. The patient having survived the various dangers of the disease, and having reached an advanced stage of convalescence, the all important question of discharge now arises. This disease, unlike Scarlatina (in which the causative organism is as yet unknown) is not one in which the patient can be discharged after a stay of a certain definite number of weeks in hospital. Here the freedom of the original focus of infection from the Diphtheria bacillus is an essential. It is the attainment of this object that one finds so difficult in a certain percentage of cases. Even though this percentage may be small Ker¹ states: "It is large enough to make the management and treatment of carriers a most important question." Many varying methods of treatment may be tried, but the condition may still persist in spite of all efforts, and cause considerable inconvenience to the patient, and give rise to a certain amount of unpleasantness/

unpleasantness with importunate relatives.

The term 'diphtheria carrier' is used to denote an individual, who while not clinically suffering from diphtheria, harbours the Klebs-Loeffler bacillus, and through the elimination of which he is capable of infecting others.

The more commonly recognised sites in which the diphtheria bacillus persists are the tonsils, pharynx and the nasal fossae. These form by far the majority of cases. In addition to these however, virulent diphtheria bacilli have occasionally been isolated from ear discharges in cases of otitis media, and the conjunctiva, vaginal or even urethral mucous membrane may occasionally be infected. I have isolated typical morphological diphtheria bacilli from all the above situations, membrane being present in all cases. The skin may also become infected, particularly at the corners of the mouth or angles of the external nares in which situations cracks and fissures are liable to appear.

Ledingham and Arkwright² divide carriers into three classes. Those who have had clinical diphtheria, and in whom the bacillus persists after the disappearance of the membrane and apparent return to health, are termed convalescent carriers. The second class/

class consists of those who harbour the diphtheria bacillus, but who have not had a clinically recognisable attack of diphtheria. These are termed healthy carriers. The patient who harbours the bacillus for more than three months is termed the chronic carrier. Most of the latter class probably have some unhealthy condition of the mucous membrane of the fauces, naso pharynx or accessory sinuses of the nose. Cases of membranous or fibrinous rhinitis fall into this group.

The classification by Muir & Ritchie³ into convalescent, temporary and chronic carriers, and secondly, healthy carriers, is simpler and a rather better classification to my mind. A few of the healthy carriers may subsequently develop the disease and are thus termed precocious carriers.

Simon⁴ on the other hand divides carriers into active and passive. The active group correspond to the convalescent carrier, temporary or chronic. The passive are the so called healthy carriers already mentioned.

With the exception of twelve, all my series of 223 cases fall into the convalescent group of carriers. On admission, 211 showed a definite clinical diphtheria of either faucial, nasal or laryngeal type. The twelve healthy or passive carriers were patients who/

who on admission had no clinical symptoms suggestive of diphtheria. Most of the latter were contacts, and were found to harbour the diphtheria bacillus in throat or nose. A few were convalescent Scarlatina patients who were incidentally found to be diphtheria carriers.

Discovery of diphtheria bacillus.

Klebs in 1883 described the diphtheria bacillus as occurring in the false membrane from diphtheria.

In 1884 Loeffler published the account of his successful attempts to cultivate this bacillus and of his experiments which demonstrated its pathogenicity for animals. It is interesting to note that in his first researches he obtained typical virulent diphtheria bacilli from the throat of a healthy individual. It occurred to him that at a time when Diphtheria was prevalent the specific organism might possibly occur in the throat of a child without setting up any symptoms of disease. In this supposition we see the present day healthy or passive carrier idea hinted at.

Previous to the discovery of the specific organism it had been believed by Trousseau⁵ that severe cases of Diphtheria could give rise by infection to mild/

mild cases of sore throat, and these in turn to fresh cases of Membranous Diphtheria. This belief was greatly strengthened by the discovery of the apparent causal organism.

Loeffler's discoveries were verified a few years later by many observers, all of whom with one or two exceptions found the characteristic bacillus almost constantly occurring in typical clinical cases of diphtheria.

In 1890 Roux and Yersin⁶ found the bacilli in the throat two weeks after the disappearance of the membrane. The conclusion they came to was:-

"Dans la diphterie, le bacille specifique peut disparaitre de la bouche en meme temps que les fausses membranes, ou y persister quelques jours apres elles, ou meme y demeurer a l'etat virulent pendant un temps assez long, mais qu'il est impossible de precises."

Loeffler in 1890 reported a case in which virulent bacilli could be demonstrated three weeks after the temperature had become normal and Abel in one instance found the bacilli sixty-five days after the disappearance of the membrane.

In consequence of these facts Loeffler in 1894 proposed that "Convalescents from diphtheria should not/

not be permitted to resume normal social intercourse until the complete disappearance of the bacilli had been demonstrated by bacteriological examination."

The importance of the healthy carrier in the spread of diphtheria is now recognised.

Occurrence of Bacillus Diphtheriae in cases of disease.

The percentage of cases of clinical diphtheria in which the bacillus is found varies according to different observers.

Woodhead⁷ states that of the 12,172 cases admitted into the Metropolitan Asylums Board Hospitals during 1895-6 and certified as suffering from diphtheria, at least 20 per cent offered no bacteriological evidence of diphtheritic infection. Graham Smith⁸ from collected records of examinations of 30,000 certified cases found that diphtheria bacilli are found in about 71 per cent of the cases. One of my predecessors, Meikle⁹, found that out of 571 cases admitted to the Edinburgh City Hospital in 1905, 17.2 per cent could not be classified as diphtheria using Neisser's staining method. He makes the interesting statement that though none of the non bacteriological cases were fatal yet four showed signs of diphtheritic paralysis. In a few apparently typical clinical cases/

cases of faucial diphtheria I have been unable to detect the diphtheria bacillus on repeated culture. From these figures we see that there is evidently a marked difference between clinical and bacteriological diphtheria on admission to hospital. When however cases are finally classified in hospital as clinical diphtheria this discrepancy between bacteriological and clinical diagnosis dwindles down to somewhere about 5 per cent. The reasons for not being able to demonstrate the bacilli in these cases may be due to the scarcity of the organism, to its being overgrown, or perhaps to the particular strain in question not staining typically with Neisser.

The distribution of diphtheria bacilli in contacts.

It is obvious that the percentage of contacts who become carriers of the diphtheria bacillus will vary very greatly according to the closeness of their relationship to the diseased person.

It has been proved by many observers that infection may be conveyed to others in the saliva, through coughing and sneezing. The kissing of children is another fertile source of spread. In schools we have infection arising through the media of infected sweets, pencils, slates, etc. which are literally passed/

passed from mouth to mouth. The pleasure the average child appears to derive from sucking pencils, or in fact most things, is evident daily, and it is by this means one gets such rapid spread of infection in school classes particularly of younger children. Another equally dangerous source of infection is the nasal discharge laden with virulent diphtheria bacilli.

Interesting examples in support of the above statements are given by numerous observers among whom might be mentioned White¹⁰, Bugbee¹¹, Newsholme¹², and Ford¹³.

Family life, nursing the sick, and school life, are the best examples of close contact where the possibilities of infection are greatest. Besides these we have the less prolonged and close proximity of ordinary everyday life and business.

Very many records of the percentage of contact carriers have been gathered.

In the case of infected families, it has been found that the proportion of infected persons varies between 50 per cent and 100 per cent, but may fall as low as 10 per cent where the hygienic conditions are very good.

Cobbett¹⁴ during an epidemic examined 650 adults and children and found 3 per cent were carriers.

In an examination conducted for the State Board of/

of Health of Minnesota¹⁵ it was found, that in five schools containing 225 children the number of carriers in the individual schools varied from 3 per cent to 25 per cent.

Graham Smith¹⁶ in a school epidemic at Colchester found 10.4 per cent carriers out of 510 persons. The same observer¹⁷ gives the percentage of carriers found among healthy contacts as varying from a mean of 66 per cent in members of the family if the conditions for spread are favourable, to a mean of 8.7 per cent in infected schools. In hospital wards and institutions he gives from the collected observations of numerous observers the percentage of carriers as 14 per cent.^{18.}

Weaver¹⁹ found in an investigation extending over two years that 15.7 per cent of the nurses in a hospital for contagious diseases became carriers.

From these figures we see that the number of carriers in those who have been in immediate contact with a case of diphtheria may be very considerable. These carriers in turn pass the bacillus on to others and thus we get the bacillus in the throats of people who have never been in contact with a clinical case.

The distribution of diphtheria bacilli in non-contacts,

The percentage of carriers among people, who as far as it can be ascertained, have never been in the immediate proximity of a clinical case of diphtheria has been estimated by various observers.

Ritchie²⁰ in an examination of 1000 scarlatina cases admitted to this hospital, found morphological diphtheria bacilli in 11.8 per cent; as this result was based on the examination of only one swab, the real percentage was probably higher.

A recent exhaustive investigation by Guthrie, Gelien, and Moss²¹ of the diphtheria carrier problem has given some interesting facts. A single throat culture from each of 1217 children revealed diphtheria bacilli in 3.61 per cent. An examination of 1290 individuals in the general population of the city of Baltimore showed 3.48 per cent of carriers. The total of 2507 persons gives a carrier percentage of 3.55 per cent. The investigation was carried out in the spring months.

It was found on a re-examination of 46 of the school children who had previously given a positive culture that after an interval of six to twelve weeks ten again gave positive cultures. They investigated forty-nine school children carriers as to their previous/

previous relations with clinical diphtheria. It was found that only three had had clinical diphtheria, respectively three, four and five years previously. Another three gave a history of exposure to diphtheria three, five and seven years previously. An interesting point was that not one of these forty-nine carriers subsequently developed clinical diphtheria, nor did a case of clinical diphtheria arise among their associates.

The second investigation of eight hundred school children in Baltimore yielded 10.62 per cent with positive throat cultures. An examination of the same children three months later gave 8.62 per cent of positive cultures. Only ten of the children gave positive cultures at both examinations. The weak point is the fact that only one culture was examined from each child.

Fifty children with positive throats were examined at few weekly intervals and after three months only six were found positive.

Virulence tests performed on guinea pigs were carried out and gave 11.11 per cent and 12.76 per cent of virulent strains from two groups of ninety-nine and forty-seven child carriers respectively.

None of 160 carriers subsequently developed diphtheria/

diphtheria nor did any cases of the disease arise among the contacts of these carriers. None of these carriers gave a history of a recent exposure to diphtheria.

Graham Smith²² from a series of 2,132 cases investigated by different observers gives the percentage of carriers as 2.80 per cent. In a series of 2955 non contacts quoted in the "Report on diphtheria bacilli in well persons"²³ 1.3 per cent showed diphtheria bacilli in their throats.

Distribution of virulent and non virulent bacilli.

As the disease producing power of this organism depends upon its virulence, it is of the highest importance to ascertain what proportion of carriers harbour virulent bacilli. In the series of 2,132 cases gathered together by Graham Smith, and quoted a few lines back, the proportion of carriers of virulent bacilli was 0.18 per cent out of 2.80 per cent of carriers. The same observer²⁴ states, that in 1 to 2 per cent of healthy persons, whether contacts or non contacts, non virulent diphtheria bacilli are to be found. It is interesting to note that these avirulent bacilli may also be encountered in the throats/

throats of sufferers from clinical diphtheria.

On the other hand the virulence of diphtheria bacilli harboured by contacts is usually marked. The percentage of fully virulent bacilli in recently infected contacts is given as 80.1 per cent by Graham Smith²⁵.

Weaver²⁶ states that bacilli from people who have been in close contact with cases of diphtheria are practically always virulent.

In one of my cases - that of a Scarlatina patient who was also found to be a faucial carrier, though no history of contact with a clinical case of diphtheria could be traced - the strain was found to be highly virulent, killing the inoculated guinea pig in seventy-two hours with typical lesions.

This obviously is of the greatest interest, because it means that every patient who is discharged with diphtheria bacilli still in throat or nose is a potential source of infection of virulent bacilli to all with whom he comes in contact. Secondly, working on this fact, the removal of contacts to hospital seems a wise precaution, as they are in all probability harbouring a virulent strain.

The next interesting question that arises is whether the non virulent strain in a carrier can change to a virulent strain, either by the method of passage/

passage, or by the effect of unknown agencies. This knowledge assumes considerable importance in the case of hospital convalescent carriers in whom no treatment has been of any avail. If it is found on animal experiment that the particular strain harboured by the patient is non virulent, and if we can accept it as a fact that a non virulent cannot change into a virulent strain, then we can discharge the patient with an easy conscience.

I might mention here, that a strain of the diphtheria bacillus which causes little or no reaction in a guinea pig when the standard virulence test is applied, has always turned out to be non pathogenic to man.

Muir and Ritchie²⁸ state that from the accumulated experience of numerous observers quite avirulent bacilli do not give rise to infection, and may be disregarded. The same observers²⁹ also agree that attempts to render freshly isolated avirulent organisms virulent have generally failed.

Ledingham and Arkwright³⁰ state "There is no known method by which a virulent strain of Bacillus diphtheriae can be converted into a non-virulent strain or vice versa, and almost all attempts in this direction have completely failed." According to Graham Smith,³¹ totally non pathogenic diphtheria bacilli/

bacilli cannot be rendered virulent.

As to whether gradations of virulence can occur, there seems to be some difference of opinion. Arkwright³² records having found in the course of an investigation of a school epidemic, strains of varying virulence. He isolated twenty strains, and on testing for virulence, found that seven were non virulent strains, six partially virulent, and seven were fully virulent strains. He acknowledges however, that after trying many methods, he has always failed to transform true non-virulent diphtheria bacilli into virulent diphtheria bacilli.

Abbott³² found that under certain unknown circumstances the virulence of *Bacillus diphtheriae* is diminished or lost.

It was found by Meikle³³ that the diphtheria bacilli in several convalescent carriers had become avirulent, and he ascribes this loss of virulence to thorough and continued local antiseptic treatment.

Moss, Guthrie and Marshall³⁴ very recently experimentally inoculated the throats of five healthy persons, previously proved to be free from diphtheria bacilli, with a pure culture of non virulent bacilli. The object of this experiment, among other things, was to see if the organisms introduced were capable of (1) producing clinical diphtheria, (2) any subjective or/
or/

or objective symptoms, (3) whether organisms are changed morphologically or in their ability to produce toxin.

Inoculation of the throats was done by swabbing and spraying, the latter method being used in the second inoculation at an interval of two days after the first. Daily cultures were then taken from all five, and were examined for some weeks, 114 days in the case of four of them; then at an interval of a few days for three months, and subsequently at longer intervals for about fifteen months.

From these experiments it was found that the carrier state was easily produced by inoculation with avirulent diphtheria bacilli, and lasted in two cases for fifteen months at least. Avirulent bacilli did not produce either clinical diphtheria, subjective, or objective symptoms. In the associates of these healthy carriers no case of clinical diphtheria occurred. The bacilli showed no tendency to become virulent. The bacilli after long residence in the throat were not altered in morphology, staining, or cultural characteristics. This experiment confirms the guinea pig test for virulence.

This research very strongly supports the statements that avirulent diphtheria bacilli are devoid of pathogenic importance for man, and hence the carrier of/
of/

of avirulent organisms is not dangerous to the health of the community.

In a very recent investigation, Eagleton, Glenny and Baxter¹⁰² in a series of 80 carriers of virulent diphtheria bacilli, found that three of the patients before becoming free of bacilli, yielded an avirulent strain. They explain this very reasonably, as an instance of an infection by an avirulent bacillus after the disappearance of the virulent bacilli. The other explanation is that a double infection may occasionally be missed, and they give a case in point.

In one carrier, both virulent and avirulent bacilli were obtained, the latter one month later. It was found after a prolonged investigation, in the course of which more than 200 colonies from subcultures of these strains were tested for virulence, that no mutation occurred, and the virulent and avirulent bacilli remained true to type.

Another investigation in progress they suggest shows that avirulent strains produce no immunity in animals against diphtheria toxin.

The conclusions they come to are, that, in vitro at least, the two strains of bacilli, virulent and avirulent, remain always true to type, the avirulent bacillus does not give rise to diphtheria, and that it does not change to the virulent variety in the throat.

It is on collected evidence such as this that I think we are perfectly justified in discharging carriers with bacilli still present in throat or nose, when it has been found by careful experiment on guinea pigs, that the bacilli in question are quite avirulent.

The persistence of diphtheria bacilli in convalescents.

The duration of persistence of the specific organism in the throat or nose of convalescents is a question of great importance. Before we can discharge the patient, we have to obtain one or more negative cultures from throat, or nose, or both, as the case may be. In the majority of cases luckily, the bacillus has disappeared from the focus of disease before the patient is clinically fit to be discharged. There are, however, cases in which culture after culture shows diphtheria bacilli, which may persist for weeks, months, or even years, and all efforts to dislodge them are apparently useless.

Many investigations have been made to find out the length of time during which convalescent diphtheria patients retain the organism in the throat or nose.

Park and Beebe³⁵ in an investigation of 752 cases under/

under the direction of the Health Department of New York City, 1893-1894, found that in 43.2 per cent the bacilli disappeared within three days after the complete disappearance of the membrane; 90.1 per cent within fifteen days and 97.6 per cent within three weeks. The mean duration was seven days from the date of disappearance of the membrane.

Prip³⁶ in 1900 investigated a series of 654 cases, and found that in 93.2 per cent the organism had disappeared from the throat within four weeks.

Woodhead³⁷ in a large number of cases examined at the Metropolitan Asylums Board Hospitals found the mean persistence to be fifty-one days. He found that the administration of antitoxin did not diminish the persistence.

Simon³⁸ quotes a table from an investigation of 1338 cases by Tjaden.

In 67.0% of cases the organisms were gone after 2 weeks

| | | | | | | | | |
|-------|---|---|---|---|---|---|----|---|
| 75.0% | " | " | " | " | " | " | 3 | " |
| 83.6 | " | " | " | " | " | " | 4 | " |
| 89.1 | " | " | " | " | " | " | 5 | " |
| 100.0 | " | " | " | " | " | " | 17 | " |

Weaver³⁹ gives a table showing the rate of disappearance of diphtheria bacilli in 500 consecutive cases of diphtheria at Durand Hospital. The following extracts show the numbers free of bacilli after three, four/

four, and five weeks respectively.

| | |
|----------|--------------------------------------|
| In 54.2% | the organisms had gone after 20 days |
| 79.8% | " " " " 30 " |
| 88.88 | " " " " 35 " |
| 100.00 | " " " " 75 " |

The same observer studied the rate of disappearance of diphtheria bacilli from the throat and nose in fifty-two carriers, and found the following results.

| | |
|-----------|--------------------------------------|
| In 57.69% | the organisms had gone after 20 days |
| 76.92% | " " " " 30 " |
| 84.00% | " " " " 35 " |

One case persisted for eleven months.

Graham Smith⁴⁰ gives a table showing the mean persistence of diphtheria bacilli in convalescent throats as found by various investigators:-

| | |
|--|-----------|
| Massachusetts Board of Health(1896-1905) | 28.0 days |
| Wesbrook (1900) | 29.0 " |
| Park | 6.6 " |
| Prip (1901) | 24.7 " |
| Minnesota Board of Health | 26.8 " |
| Gluchsmann | 24.8 " |
| Scheller (1905) | 20.9 " |
| Tobieson | 8.4 " |
| Woodhead | 51.2 " |

To/

41

To these might be added:-

| | | |
|---------------------|---------------|-----------|
| Graham Smith (1904) | | 36.0 days |
| Roux and Yersin . | Days from | 13.6 " |
| Meikle . . . | disappearance | 18.4 " |
| Walsh . . . | of exudate | 22.0 " |

Repeated examinations of 605 cases of diphtheria were made by Park and Williams⁴² to see how long the bacilli persisted. They found that:-

| | |
|--------|---|
| 70.99% | of cases were free of bacilli within 7 days of disappearance of membrane. |
| 89.40% | " " " " " 12 days " " |
| 94.30% | " " " " " 15 " " " |
| 98.10% | " " " " " 21 " " " |
| 99.2% | " " " " " 28 " " " |

The mean persistence was 6 to 12 days after the disappearance of the membrane.

Wesbrook⁴³ found in clinical cases of diphtheria that:-

| | |
|---------------|---------------------------------------|
| 51.0 per cent | had lost the bacillus by the 21st day |
| 83.0 | " " " " " 28th " |
| 93.0 | " " " " " 35th " |
| 100.0 | " " " " " 70th " |

Hartley and Martin⁴⁴ in a series of 3075 observations on 457 cases admitted to a military hospital for infectious diseases at Rouen, gives the/

the rate of disappearance of diphtheria bacilli as follows:-

| | | | | | | | |
|--------|---|---|---|---|----|---|---|
| 74.18% | | | | | | | |
| | | | | | | | |
| 79.87% | " | " | " | " | 35 | " | " |
| 84.68 | " | " | " | " | 40 | " | " |

The time the patient reported sick with a sore throat was taken as the day of onset, and he was considered free of bacilli on the day half way between the last positive finding, and the first of three successive negatives.

From the above collected statistics we may take it, that in at least 80 to 85 per cent of cases, the diphtheria bacillus has disappeared from the throat by the end of the fifth week, and ⁱⁿ 98 per cent by the end of the ninth week.

In my own series of fifty-four control cases, 81.48 per cent were free of bacilli by the end of the fifth week.

There are exceptional cases however, in which the bacilli may persist in throat, or nose, or both, for a very much longer time, and it is these cases which give rise to such difficulty in fever hospital administration.

Statistics collected by Graham Smith⁴⁵ and presented/

presented in the form of a table, show the longest periods during which various observers have found diphtheria bacilli persisting in the throat or nose. This period varies from 45 days in the case of Gladin to a case noted by Prip which carried for 669 days. It appears, however, that in the very long periods of persistence, the observers in question have only taken into consideration the morphological appearance of the bacilli.

Many cases of prolonged persistence can be found in the literature. Simon⁴⁶ quotes various observers who demonstrated the diphtheria bacillus at intervals of six, nine, and eighteen months, and Prip mentions a case in which they were found after four years. Neisser has described a case of chronic nasal diphtheria in which he could isolate the organism after eight years.

Macdonald⁴⁷ describes a case where bacilli remained in nose and throat of a diphtheria convalescent for more than fifteen weeks; in another case, virulent diphtheria bacilli were demonstrated in the pus from an ear, in a patient, eight months from onset of illness.

Wesbrook⁴⁸ found virulent diphtheria bacilli in a girl of ten for 109 days, and in the case of a school teacher, for 80 days, after her attack.

The Virulence of Bacilli in Convalescent Carriers.

When one is faced with these cases of prolonged persistence the question of virulence assumes considerable importance. Perhaps by some unknown agency the bacilli originally fully virulent may have become attenuated, and non virulent, during their prolonged stay in the throat or nose.

Unfortunately, on reviewing the results of various observers who have investigated this question, we find that in the great majority of cases the bacilli retain their full virulence up to the time of final disappearance.

Graham Smith⁴⁹ quotes various observers who found fully virulent diphtheria bacilli in the noses and throats of patients six weeks, seven weeks, five months, seven months, and seven and a half months after the attack.

Cobbett isolated, and examined for virulence, the bacilli from a number of persons until their disappearance. In one person, virulent bacilli were isolated on the 2nd, 23rd and 30th days, and in another on the 1st, 30th, 36th and 66th days. In two contacts he examined, virulent bacilli were isolated on the 15th, 36th and 55th days in one case, and on the 1st, 18th, 28th, 60th, 69th and 82nd days in/

in the other. At the same time he repeatedly examined three other contacts who were harbouring non-virulent bacilli, and he found they remained non-virulent in one case, on the 1st and 31st days, in another on the 24th and 34th days, and in the third from the 1st to the 93rd day, ten examinations being performed in all in the latter case.

Prip mentions a case where full virulence was found in one case up to 335 days.

Wesbrook, as already mentioned, found virulent bacilli up to 80 and 109 days respectively.

On the other hand, Meikle⁵⁰ tested for virulence cultures taken from five convalescent diphtheria patients. Four of them had been running persistently positive for four weeks, and one for two weeks. In no case did the inoculated rabbit die. He suggests that this apparent loss of virulence might be due to the active antiseptic treatment which these cases had received during their sojourn in hospital.

Wadsworth⁵¹ gives the results of 548 virulence tests of cultures obtained from convalescent and contact carriers. He finds that 90 per cent of cultures obtained from convalescent patients during the first 3 months after the onset of the disease, and from contact carriers, are virulent. In 147 strains isolated/

isolated from convalescents within three months of attack, it was found that 92.5 per cent were virulent. Examinations of convalescent carriers of more than three months standing showed 89.2 per cent virulent strains. He also examined 35 strains of *B. diphtheriae* from healthy contact carriers, and found 80 per cent virulent. The conclusion he comes to, is, that in convalescent and contact carriers the diphtheria bacilli retain their virulence for several months.

In a very recent paper by Eagleton & Baxter,⁵² virulent cultures were obtained from 90.9 per cent of convalescent carriers. In cultures from contact carriers they found 38.8 per cent were virulent.

Fraser and Duncan⁵³ describe three cases in which bacilli persisted for 18 months (throat), 3 years (throat and nose) and 23 months (nose). In the last two cases they state the bacilli were virulent.

As already mentioned, the bacilli found by Macdonald in an ear discharge for a period of eight months, were virulent all through.

Weaver⁵⁴ describes a nasal carrier, a baby of four months, in whom the cultures were virulent for guinea pigs for eleven months.

Guthrie, Marshall and Moss⁵⁵ inoculated eight people with virulent diphtheria bacilli obtained from the throat of a healthy carrier. In four cases clinical/

clinical diphtheria developed; three did not develop the disease, but became healthy carriers. The carrier state persisted from 33 days to more than 72 days. The really important point is, that they found that the bacilli, even after prolonged persistence in the throats of the carriers, still retained their virulence.

When one considers these findings I am afraid on the weight of the evidence, we must assume that the bacillus harboured by a convalescent carrier retains its virulence, and the carrier must be dealt with as a potential danger to the community. Mark you I use the word potential, advisedly, because in the case of faucial carriers I am inclined to think the danger in many cases is more potential than real.

Briefly summarising these facts about carriers, we see that the convalescent diphtheria patient harbours the diphtheria bacillus in throat, nose, or both, for a varying period of time after the disappearance of the membrane. We can postulate with some degree of confidence, that at least 80 to 85 per cent of cases have rid themselves of bacilli by the end of the fifth week of disease. The diphtheria bacillus has been found to persist in other situations such as the ear, eye, and wounds, but in routine fever hospital practice one so seldom comes across these cases that they/

they are of relatively little importance. Mark you these cases have apparently given rise to clinical diphtheria in contacts on several occasions, and only recently I read an account of a series of cases which were apparently infected from a chronic sore on the scalp of the mother, which contained diphtheria bacilli; so one must treat them with caution.

Luckily, in the majority of cases, the bacilli disappear from the focus of infection either before, or at such a time, as to enable the patient to be discharged at the termination of the period of convalescence.

There is, however, the very troublesome minority, who develop the so-called chronic carrier condition.

We limit the term chronic carrier in this hospital, to patients who have consistently given positive cultures for a period of three months, and the bacilli may persist in these cases for months or even years.

In the great majority of these chronic carriers the bacilli retain their virulence, and hence they are a potential danger to the community. Under such circumstances one is forced to insist on quarantine, thus causing great inconvenience and distress to the patients themselves, their relatives, and last but not least to the physician who really becomes their jailor. They are also the bane of the unfortunate bacteriologist, who examines countless cultures and invariably/

invariably returns a positive result, with great resultant loss of popularity all round! It is very curious how in some mysterious way or other he is usually held personally responsible for the undue persistence of the organism as if he were loth to part with it! If the strain harboured by the patient has been found to be non virulent to guinea pigs, then we are justified in assuming it is non-pathogenic to man, and the patient can be discharged with bacilli still present.

The question now arises, what can be done for these chronic carriers of virulent diphtheria bacilli? Can we by any method of treatment hasten the disappearance of the offending organism? In this field a large amount of work has been done, and many methods tried. Sufficient comment on the results of all this work is the mere fact that we still have the persistent carrier. However, even though one may be convinced that no treatment is of any avail, yet one is driven to trying something if only to satisfy the patient and his friends.

In the following pages the treatments I shall briefly review are.-

- (1) Effect of antiseptics.
- (2) Effect of antitoxin.
- (3) " " bactericidal serum.
- (4)/

- (4) Effect of ferment from *B. pyocyaneus*.
- (5) " " staphylococcal spray.
- (6) " " diphtheria endotoxin (Hewlett).
- (7) " " vaccines (ordinary and detoxicated).
- (8) " " tonsillectomy and adenoidectomy.

ANTISEPTICS. When one takes into consideration that the organism is not a hardy one, the thorough application of antiseptics should on the face of it dispose of the diphtheria bacilli. A very great variety of mouth washes, gargles, and nasal sprays, have been tried. Amongst the medicaments employed are carbolic, iodine, alcohol, chlorine, menthol, thymol, corrosive sublimate, izal, lysoform, peroxide of hydrogen, toluol, chinosol, boroglyceride, hydrochloric acid, zinc chloride, double chloride of sodium and gold, nitrate of silver, collargol, lactic acid, buttermilk, potassium chlorate, sulphurous acid, and gentian violet. In some cases good results seemed to follow.

Park⁵⁶ found that thorough irrigation of the throat and nose with 1 : 4000 perchloride of mercury at intervals of a few hours, will lead in one half to two thirds of the cases, to the disappearance of the diphtheria/

diphtheria bacilli within three or four days of the disappearance of the membrane. In the remainder the bacilli still persisted.

Meikle⁵⁷ gave a thorough trial of antiseptic local treatment in a series of 245 cases. He used various coal tar preparations, chlorine, corrosive, toluol and borax, or^a combination of these. The average persistence of diphtheria bacilli in the cases varies in the different antiseptic groups from 16 to 43 days, and even after this thorough antiseptic treatment, 27 cases were still harbouring diphtheria bacilli on discharge. An analysis of his results shows, that the various antiseptics had no effect on lessening the duration of stay of the bacilli in the local focus. Graham-Smith has come to the conclusion that the duration of persistence of the bacilli is not materially affected by antiseptic treatment.

Moss⁵⁸ on the other hand treated six cases by daily application of 50 per cent iodized phenol to throat and pharynx, and 20 per cent argyrol into the nostrils at three hour intervals. The cases all cleared up in from four to six days. These cases were apparently contact carriers. He gives no data to say how long they were carrying. The fallacy here is that diphtheria bacilli have a habit of disappearing spontaneously in a shorter or longer period/

period of time, hence they may have disappeared within the same time, even had no antiseptics been used.

The same observer gives $6\frac{1}{2}$ days as the average time required to clear up a carrier. He found, however, that seven cases in another series, persisted positive after one week of active local treatment; three both nose and throat, four nose only. The local treatment consisted of $33\frac{1}{3}$ per cent hydrogen peroxide gargle, and 15 per cent argyrol instilled into the nose.

These cases in which the diphtheria bacillus has persisted after one week of active local treatment, he feels fully justified in regarding as true carriers. I shall speak of these cases again.

Very recently Moss and Guthrie⁵⁹ tried the application of a spray of 1 : 10,000 gentian violet once daily to the throats of carriers, but found it had no effect in hastening the disappearance of the bacilli.

This lack of success in the use of antiseptics may seem curious, as we have the idea that the diphtheria bacillus exists on the mucous surfaces of the throat and nose. Why is it then, that we get persistence after antiseptic applications, which we know are strong enough, if not in all cases to kill, at least to inhibit the growth of the bacillus.

A negative result is nearly always obtained if the throat/

throat be swabbed within an hour or two of a thorough application of antiseptics, but a culture taken twenty-four hours later will again show diphtheria organisms to be present. The reason for this is probably that the bacilli situated superficially, and thus within the zone of antiseptic influence are attenuated or killed off, but it is in the tonsillar crypts, and recesses and folds of the mucous membranes, in the nasal sinuses, that the culprits lurk. The superficial application of antiseptics, no matter how powerful, will not reach these hiding places, which act as a constant source of replenishment. In one of my cases, where tonsillectomy had to be performed in an attempt to clear up a carrier, on sectioning and staining the tonsil, I saw a group of morphologically typical diphtheria bacilli intermingled with debris near the bottom of a tonsillar crypt. Ballantyne and Cornell⁵⁹ report finding, on sectioning and staining tonsils from four cases, the bacilli in the very bottom of the crypts. When one thus sees the position of the organisms, one realises the futility of gargles and mouth washes.

Albert,⁶⁰ who realised the difficulty, has attempted to overcome it in the case of tonsillar carriers, by treating each crypt by means of a suitable thin, dressed/

dressed probe, with a 5 to 10 per cent solution of silver nitrate. The principle of this method appears to me to be very sound. The technique I should imagine would be none too easy, and when one considers the minute size of a potentially suitable nidus for the diphtheria bacillus, it is evident that the application must be very thoroughly and conscientiously performed. He records success in fourteen carrier cases so treated, the organisms disappearing in all cases within three days.

Another investigator reports that he managed to free of bacilli some thirteen faucial carriers by squeezing the tonsils, and so forcing from the crypts the plugs of debris containing the bacilli.

A point to criticise in these cases, is whether the above were carrying for a sufficient length of time to be termed chronic carriers. In 85 per cent of cases the throat becomes free of bacilli within five weeks, without local treatment of any kind. An attempt has been made to excite an acute inflammatory process in the crypts by probes dressed with mustard oil. The desired inflammation was produced in the superficial part of the crypt, but the bacilli still persisted in the deeper parts.

Mark you I do not decry the use of mild anti-septic mouth washes, nasal douchés, etc. in clinical diphtheria./

diphtheria. These applications serve a very useful purpose in the mechanical cleansing they ensure, and I should think they will serve to keep the growth of the organism in check to some extent. By all means use some mild antiseptic preparation, a very large choice is open, but do so not with the hope of causing the early disappearance of the causative organism, but merely to keep the focus of disease sweet and clean. Meikle suggests that the thorough use of antiseptics might explain the loss of virulence found by him in cultures from five clinical cases of diphtheria taken about the second and fourth weeks of convalescence.

If the organism be more or less inaccessible in the tonsillar crypts, how much more so still in the various sinuses and recesses of the nose.

Personally I have only tried one special antiseptic line of treatment, and that is by nascent iodine. Nankivell⁶¹ tried the administration of potassium iodide until slight iodism resulted, with the idea that the free iodine might act antiseptically on the lining membrane of the accessory nasal air sinuses. His results were not successful. At Dr Ker's suggestion I tried the effect of administering potassium iodide in XXV gr. doses at 9 a.m. and following it up with one ounce of chlorine water at 2 p.m., 4 p.m., and 6 p.m. This was tried in three cases.

CASE I. Faucial convalescent carrier.

Diphtheria bacilli still in throat 79 days after admission to hospital. The above treatment given on three consecutive days. Diphtheria bacilli still present in throat three weeks after the treatment.

Result = failure.

CASE II. Faucial convalescent carrier.

Diphtheria bacilli still in throat 38 days after admission to hospital. Above treatment given on three consecutive days. Diphtheria bacilli were still present 12 days after the course. A second course of two days was given, and diphtheria bacilli disappeared on the following day, e.g. 53rd day after admission. This case not conclusive, as he could not be classed as a persistent carrier.

CASE III. Nasal carrier of 78 days duration after admission to hospital. Above treatment given on seven consecutive days. Diphtheria bacilli still persisted in the nose 12 days after the last day of treatment.

Result = failure.

These results show that this attempt at treatment by nascent Iodine was not successful in my hands.

Inhalation/

Inhalation of various antiseptics have been tried, but as far as I can ascertain without much success. Simon⁶² states that in the French army in 1910 the carrier was made to inhale the fumes of a mixture containing iodine, guaiacol, thymol, etc. vaporised by means of hot water. Five sittings of two to three minutes duration in the twenty-four hours was the routine. He assumes this treatment was not very successful as no report was issued concerning its efficacy. I have seen this inhalation method used against the Meningococcus in the Naval Barracks at Devonport but never for Diphtheria.

I do not see how inhalations are going to get at the bacilli any more efficiently than the ordinary methods of gargling or nasal douching, etc.

ANTITOXIN. Most observers agree that the administration of antitoxin has absolutely no effect on the persistence of the bacilli, and the carrier state. Simon⁶³ makes a very categorical statement to that effect, and also quotes Prip who regards antitoxin as useless in the treatment of the carrier from the point of view of hastening the disappearance of the bacilli. Woodhead⁶⁴ cites two cases, one which had antitoxin, the other not, in both of which the bacilli persisted for more than

200/

200 days. Meikle⁶⁵ quotes two cases in his series which had no antitoxin, and in whom the persistence was 20 days, as against 12 to 23 days in antitoxin treated cases.

I have had no cases untreated by antitoxin, but it seems to me reasonable, that if antitoxin should have any effect in hastening the disappearance of the bacilli, then the cases which receive the larger doses should become free of bacilli more quickly than the cases with lower dosage. In 59 control cases I find that 42 received doses of 6000 units of antitoxin and under; 17 received doses of 8000 units and over, one case having 32,000 units. In the first group the mean time of persistence of the bacilli was 20.00 days, in the second group 27.70 days.

In my series of 145 vaccine treated cases 112 received 6000 units of antitoxin or under, and the persistence of bacilli from the first symptom was 17.95 days, whilst in 33 cases which had more than 8000 units the persistence was 26.84 days.

It is evident from these figures, that the larger doses of antitoxin do not rid the convalescent throat of bacilli any more quickly than the small doses, and in fact that the antitoxin administration from this point of view is ineffective. Walsh found that in cases /

cases untreated with antitoxin the bacilli persisted for a mean of 24 days, whilst in cases which received antitoxin 25 days was the mean time of persistence.

BACTERICIDAL SERUM.

Attempts have also been made to clear up carriers by the local application of a bactericidal serum. The latter was obtained by injecting a horse with the dead bodies of diphtheria bacilli. By this means a serum with marked agglutinating properties was obtained. This serum when dried and used in the form of pastilles was claimed to cause a rapid decrease in the number of bacilli in the throat.

The Serum Department of the Lister Institute injected diphtheria bacilli into two horses intravenously, The sera obtained agglutinated the race of bacillus employed in dilutions of 1 in 200 to 1 in 300. Pastilles containing this serum were used in a series of clinical cases but no shortening of the period of persistence of the diphtheria bacilli in the throat was observed.

FERMENT /

FERMENT FROM B. PYOCYANEUS. Emmerich⁶⁶ attempted to clear up the carrier state by the use of a ferment obtained from the *B. pyocyaneus*. This ferment was found to bring about the digestion of *B. diphtheriae* in the test tube. The use of this however has apparently not been a success.

STAPHYLOCOCCUS SPRAY. On several occasions I have noticed that throats which have been harbouring diphtheria bacilli many weeks, suddenly become free of bacilli after an intercurrent staphylococcal infection. One case I find in my notes which is an excellent example. A female of 13 was admitted on 26th February 1921 suffering from Scarlatina. A culture from the throat taken on 6th March was found to contain numerous diphtheria bacilli. Cultures taken at varying intervals up to 23rd March, eleven in all, consistently showed numerous diphtheria bacilli. On 28th March she complained of slight sore throat. On the following day the tonsils, fauces and palate were congested, with a scummy soft broken exudate on both tonsils, and the throat was very sore. On examination of the throat culture of 30th March no diphtheria bacilli/

bacilli could be seen, but an almost pure culture of Staphylococci was obtained. Two more consecutive cultures on 1st and 2nd April were both negative for the diphtheria bacillus. The case strongly suggests that the invading and flourishing staphylococci, had stamped out the diphtheria organism.

In routine examination of over 12,000 diphtheria cultures I have noticed that when in a particular slide an almost pure culture of staphylococci appears, then diphtheria bacilli are usually absent or extremely scanty. The staphylococcus appears to overgrow and crowd out the diphtheria bacillus.

It was in 1909 that Schiøtz⁶⁷ of Copenhagen, noticed, that not only did intercurrent attacks of staphylococcal sore throat expel diphtheria bacilli from the throats of diphtheria convalescents, but also that patients with staphylococcus sore throats, admitted by mistake into diphtheria wards, did not take diphtheria. Working on this idea of the antagonistic action suggested by these observations, he inoculated six diphtheria carriers (3 adults and 3 children) with a staphylococcus culture isolated from the throat of a healthy patient in a surgical ward. In each case the desired negative result was rapidly obtained. Page⁶⁸ found that two hourly spraying/

spraying of the throat with a bouillon culture of *Staphylococcus pyogenes aureus* rendered the throat free of diphtheria bacilli in a few days. Other observers who report successful use of the staphylococcal spray are Catlin, Day and Scott⁶⁹ who cleared up eight cases of diphtheria carriers within 48 to 72 hours. They sprayed a 24 hour broth culture into the nose and throat two or three times daily, and gave no other treatment. The desired result has also been obtained by Leary in two cases, Wiener in one case, Lake in thirteen cases, Bell in two persistent carriers.

Lorenz and Ravenel⁷⁰ in a survey of 17 cases consisting of, three carriers pure and simple, six which had clinical symptoms, and eight which had treatment early in the disease, state that the staphylococcus spray treatment was most successful in the carrier cases, and less so in the others.

Rolleston⁷¹ tried this treatment on 10 cases, consisting of four adults and six children, all convalescent diphtheria cases. Eight were faucial cases only, and two nasal cases. In eight cases the organisms were virulent. In one case the use of the spray was started on the 25th day, but in the other nine the earliest date on which the spray was used was the 46th, and the latest the 70th. A pure culture of/
of/

of *Staphylococcus pyogenes aureus* incubated in broth for 18 to 24 hours, was sprayed on the palate and fauces, and the nostrils in the case of nasal carriers, three or four times daily. Swabs dipped in the culture were then applied to the tonsil and surrounding parts. It is interesting to note that in eight cases a mild sore throat, some constitutional disturbance, and a considerable degree of malaise was produced. In Fever hospital practice this is a point to consider, as the patients in rate supported institutions have to be treated very circumspectly. It was found that six of the faucial cases became negative within two to seven days after starting the treatment. This treatment of the nasal carriers was ineffective.

Alden⁷² states that they decided to use the Staphylococcal spray on all cases that remained positive after four weeks. He used three different strains of *Staphylococcus pyogenes aureus* isolated from throat cultures. These were grown on agar, and then in broth for 18 hours. With his broth culture the throat and nose were sprayed at least once daily. Test cultures were taken every 24 hours.

In his first 16 cases, which, with the exception of three, had carried for a month, 15 gave a negative culture within a week after the use of the spray.

In/

In one case the treatment failed. No untoward complications were noticed.

I am able to speak of the results of this treatment in two cases only.

CASE I. Male, age 37. Mild faucial diphtheria. 1750 units antitoxin. Cultures from the throat, seven in all, covering a period of 31 days from admission, showed diphtheria bacilli to be present in fair numbers. On the 31st day after admission his throat was on two occasions thoroughly sprayed with a 24 hour bouillon culture of *Staphylococcus pyogenes aureus*, and this was repeated twice daily for seven consecutive days. Cultures were taken on the 33rd, 35th, 37th and 39th days. Diphtheria bacilli persisted all through the treatment and remained in the throat for 16 days after the cessation of the treatment. He complained of some soreness of the throat, but no constitutional symptoms arose.

Result was a failure.

We may be open to the criticism that the application and dosage was not sufficient.

CASE II./

CASE II. Female, age 22. Mild faucial diphtheria. 4000 units antitoxin. Nine cultures extending over a period of 40 days after admission, with one exception, showed diphtheria bacilli to be present. On the 40th day after admission the twice daily thorough application of the staphylococcal spray was commenced. This treatment was carried on for seven consecutive days. Cultures taken on the 41st, 43rd, 45, 46th, 49th and 51st days all showed diphtheria bacilli in considerable numbers. This patient was still harbouring diphtheria bacilli in her throat 100 days after admission. Result was again a complete failure.

In this case also the patient complained of soreness and dryness of the throat, which was congested. There were no constitutional symptoms. The question here again is; was the dosage sufficiently large?

I must confess I found these results rather disappointing, as I think the idea underlying the treatment is good. It appears to me however, that it is essential if the treatment is to be effective, ^{that} the spray must be applied to such an extent as to cause a definite local reaction along with constitutional symptoms. It is only by the lodgment, and growth of the staphylococci in the fauces and nose, that we can hope for the extermination of the diphtheria bacilli./

bacilli. The action I am inclined to think is one of crowding out to a considerable extent. In the test tube De Witt ⁷³ has found that diphtheria bacilli and staphylococci will flourish together, and there is apparently no particular product of the staphylococcus growth (in artificial media at least) which destroys or inhibits the diphtheria bacillus.

The reason the same observer gives for the apparently favorable action of the spray is that it reinforces the friendly throat flora in the cases in which they are not able to regain their natural, normal ascendancy.

On the other hand, the local and constitutional reaction which is merely an indication of the hyperactivity of the protective mechanism of the body, might initiate the production of antibodies, or a leucocytosis, which either singly, or in conjunction, dispose of the diphtheria bacilli.

This same local reaction is not an unmixed blessing because one is rather chary of using a curative agent, particularly if the attainment of the desired object is very doubtful, which entails considerable discomfort to the patient. The nasal spray I would be very loth to use, as staphylococcal infections of the nose and its accessory sinuses, are already/

already unfortunately too common, and it appears to me that subjecting the nose to a heavy artificial inoculation of this sort is playing with fire.

Hewlett⁷⁴ attempted to overgrow the diphtheria bacillus in the nose or throat by applications of living cultures of *B. Hofmanni*, cocci and yeasts, i.e. the normal flora, but met with no success in two chronic cases so treated.

DIPHTHERIA ENDOTOXIN. Hewlett⁷⁵ prepared diphtheria endotoxin by growing a virulent *B. diphtheriae* on serum or blood agar. The growth was collected and washed in saline. The bacterial mass was then ground by the Macfadyen method in the presence of intense cold, and filtered through a Berkefeld filter. The filtrate forms the endotoxin, and sterile saline solution was added so that 1 cc. contained 5 mgms. This endotoxin solution when tested on guinea pigs was found to be harmless, and it gave rise on injection to a considerable protective power against living *B. Diphtheriae*. This preparation is apparently a vaccine consisting of the endotoxin.

He gave one injection of 2.0 mgms. of endotoxin to five clinical cases of diphtheria while the membrane/

membrane was still on the throat; all the cases at the time gave practically pure cultures of *B. diphtheriae*. Four cases were free of bacilli between 10 to 14 days from the date of injection, and the fifth case was clear in a month. He implies that the credit of this rapid disappearance should be ascribed to the use of endotoxin. He makes the statement that ordinary cases of faucial diphtheria are not as a rule free from infection for a month or five weeks. Looking back for a moment at the figures for persistence we find that diphtheria bacilli have disappeared from the throats of convalescents without any treatment, within four weeks, in 75 per cent, 83 per cent, and 79 per cent of cases respectively. In my own series of cases 74.07 per cent were free of bacilli by the 28th day of disease.

Meikle found that most cases of diphtheria were free from bacilli after three weeks from the beginning of the illness. Ker⁷⁶ states that only a comparatively small minority usually harbour the bacillus after the fifth week is over. From these figures, we see that the four cases treated with endotoxin with such happy results, might in all probability have cleared up equally rapidly without any treatment.

The results of endotoxin treatment of chronic carriers is/

is interesting. Three faucial carriers which had been consistently positive for 76, 188, and 108 days respectively, cleared up within a fortnight or less. In the first case 2 mgms. of endotoxin was given, in the second 4 mgms. and 2 mgms. in the third. The results in the case of 10 nasal carriers of from 35 to 126 days duration were even more encouraging. The amount of endotoxin given to these cases varied from 2 mgms. to 8 mgms. in all, and in the most persistent case the bacilli had disappeared within 10 days of the last dose of endotoxin. He also records six failures. They include both faucial and nasal carriers, and the dosage of endotoxin given varied from 0.5 mgm. to 12 mgms. In the latter case a necrotic turbinate bone was the cause of persistence of the infection. He ascribes these failures to the dosage of endotoxin being too small. He records another faucial carrier treated with 7 mgms. endotoxin in all, who cleared up within five days of the second dose. The bacilli had been demonstrated in this case in throat cultures for 48 days.

In another paper he gives successful results in four more cases, and one failure. In this paper he recommends the dosage of endotoxin which I subsequently used, namely 0.5 cc., 1.0 ccs. and 1.5 cc. containing 2.5 mgms., 5 mgms. and $7\frac{1}{2}$ mgms. endotoxin respectively, /

respectively, at intervals of seven days.

Results such as these certainly warranted a trial of endotoxin in persistent cases. On application by Dr Ker, Dr Hewlett very kindly forwarded a few cubic centimetres of endotoxin. The first batch contained 5 mgms. in one cubic centimetre, the second 10 mgms. in the cubic centimetre.

I have had the opportunity of observing its effect on five carrier cases.

CASE I. Female, age 22. Mild faucial diphtheria.

4000 units antitoxin. She persisted positive for a period of 95 days after admission into hospital, 31 cultures in all having been examined. On the 95th day $2\frac{1}{2}$ mgms. (0.5 cc) were injected subcutaneously, and subsequently 5 mgms. (1.0 cc.) and $7\frac{1}{2}$ mgms. (1.5 cc.) at seven day intervals. Three consecutive negative cultures were obtained on the 106th, 109th and 110th days after admission. The bacilli apparently disappeared four days after the second injection of endotoxin.

The endotoxin in this case was apparently successful.

CASE II. Male, age $2\frac{1}{2}$. Nasal carrier. 1000 units antitoxin. Seventeen nasal cultures taken over a period of 98 days consistently showed numerous diphtheria bacilli. Four doses of endotoxin/

endotoxin consisting of $2\frac{1}{2}$ mgms. (0.5 cc.) 5 mgms. (1.0 cc.) $7\frac{1}{2}$ mgms. (1.50 cc.) and 5 mgms. (1.0 cc) were given on the 99th, 106th, 113th, and 120th days after admission respectively. Three consecutive negative cultures were obtained on the 121st, 122nd and 123rd days after admission. In this particular case a boot button was extracted from one nostril on the 119th day, and in all probability its removal was the real factor in clearing up this persistent carrier.

The result of endotoxin in this case was doubtful.

CASE III. Adult female. Severe faucial diphtheria. 14000 units antitoxin. Sixteen nasal cultures taken during a period of 85 days after admission had shown diphtheria bacilli to be consistently present. On the 86th, 93rd and 100th days after admission doses of $2\frac{1}{2}$ mgms. (0.5 cc.) 5 mgms. (1.0 cc.) and $7\frac{1}{2}$ mgms. (1.50 cc.) respectively of endotoxin were injected subcutaneously into the forearms. Three consecutive negative cultures were obtained on the 99th, 100th and 101st days after admission respectively. The endotoxin was apparently successful in this case.

CASE IV. Infant of one year. Nasal diphtheria. 12000 units antitoxin. Cultures from nose taken on 58th, 60th, 63rd and 64th days after/

* Supply of endotoxin run out.

after admission showed numerous diphtheria bacilli still present. Twelve consecutive nasal cultures taken up to the 102nd day after admission, all showed numerous diphtheria bacilli. On the 102nd day the first dose of $2\frac{1}{2}$ mgms. (0.25 cc.) was injected; 5 mgms. (0.50 cc.), $7\frac{1}{2}$ mgms. (0.75 cc.) and 10 mgms. (1.00 cc.) were injected on the 109th, 116th and 123rd days after admission respectively.

The diphtheria bacilli diminished in number very considerably after this course of endotoxin, but they were still present in cultures for more than 6 weeks after the last dose of endotoxin.

Result of endotoxin in this case was a partial success.

CASE V. Female, age $2\frac{1}{2}$. Mild faucial diphtheria. 3000 units antitoxin. Diphtheria bacilli disappeared from the throat in 45 days, but 13 nasal cultures, with one exception, showed numerous diphtheria bacilli, the last being taken on the 78th day after admission. On the 79th, 85th, 92nd and 99th days after admission $2\frac{1}{2}$ mgms. (0.25 cc.), 5 mgms. (0.5 cc.), $7\frac{1}{2}$ mgms. (0.75 cc.) and 10 mgms. (1.00 cc.) of endotoxin were injected. Cultures still showed diphtheria bacilli to be present four days after the last injection, but they had disappeared 10 days later, i.e. on the 113th day after admission. As tonsils/



tonsils and adenoids were removed on the 107th day in this case it rather complicates the result.

I am inclined to think the endotoxin was not the causal factor in clearing up this case.

I might add here that the diphtheria bacilli in these cases were cultured pure, and then tested for acid production in glucose and saccharose peptone water. The bacillus in question produced acid in glucose, but none in the saccharose peptone water.

Summarising these results we find.-

| <u>Total Carrier cases</u> | <u>Successful</u> | <u>Partially Successful</u> | <u>Failure</u> |
|--------------------------------|--------------------------|---------------------------------|--------------------|
| 5 | 2 (1 Faucial 1 Nasal) | 1 (Nasal) | 2 (Nasal Nasal) |

Distinct
diminution
in number of
diphtheria
bacilli.

The dosage used may not have been large enough, but it is the dosage recommended, and as our supply of endotoxin was limited could not be exceeded. The local reaction consists of slight redness and tenderness at site of injection which quickly passes off. I had no general reactions.

VACCINES./

VACCINES. According to Ledingham and Arkwright⁷⁷ the first to use vaccines of dead virulent diphtheria bacilli in the attempt to cure carriers was Petruschky in 1908. He apparently had success in five cases, which ceased to carry while vaccines were being administered. In one of his cases, the bacilli persisted for 14 months, even though a long course of vaccine was given.

Hall and Williamson⁷⁸ treated six cases with vaccines. In one case diphtheria bacilli had been present for six months, and in another for several years; neither of these cases were cleared up by the use of vaccines. Three cases in which bacilli had been present three months, cleared up after four weekly injections of vaccine, and another case of four months duration also cleared up.

The dose they used varied from 75 to 1000 millions emulsified autogenous diphtheria bacilli given every seven days.

In some observations they made upon nasal and bronchial secretions, they found that the exudate did not show lytic, agglutinative or opsonic factors after the subcutaneous injections of sterilised cultures of Klebs-Löffler bacilli, until it became purulent.

Forbes, Duncan and Newsholme gave autogenous diphtheria/

diphtheria vaccine in doses of 5 to 400 million bacilli, to three cases of membranous rhinitis. The membrane rapidly disappeared, but the organisms, although much diminished in number, still persisted.

Love⁸⁰ reports the effect of vaccine treatment on seven "persistent" or "true" carriers. I have already drawn attention to these cases, which owing to a persistence through one week of local treatment, were classed as "true" carriers. The local treatment consisted of a gargle of $33\frac{1}{3}$ per cent peroxide of hydrogen and instilling 15 per cent argyrol into the nose. Of these seven cases, three harboured diphtheria bacilli in both nose and throat, whilst the other four were purely nasal infections. The vaccine was prepared from several virulent strains of diphtheria bacilli obtained during his investigation of carriers. The dosage used was 100, 250, 500, 500, 1000 millions on the first, second, third, fourth, fifth and subsequent days respectively. One case cleared up on the fourth day of vaccine treatment, four on the fifth day; one on the seventh, and the last on the 24th day. The injections of vaccine caused no local or general reactions.

These results are interesting, but as the cases had only been observed to be carrying for one week previous to vaccine treatment, hardly convincing.

Brownlie⁸¹/

Brownlie⁸¹ used vaccine in the treatment of 50 consecutive convalescent carriers at varying periods of convalescence, from the third to the 20th week. The vaccine used apparently consisted of several strains of *B. diphtheriae*, not autogenous, and prepared in the ordinary way. The dosage varied from 10 million organisms in 11 cases, to 200 million in two cases. In 44 cases, three doses of vaccine were effective in causing the disappearance of diphtheria bacilli. Three cases required four, seven, and eight doses of vaccine respectively, and three other cases five doses, to clear them up. Vaccine was injected every fourth day. Forty-four cases got rid of diphtheria bacilli within a fortnight. Twenty-five per cent of the cases, who could be observed, remained negative for from 8 to 27 days. As for reactions, a moderate increase of the pulse rate was all that was noticed. No pyrexia or local pain and stiffness occurred.

He finds that diphtheria vaccine produces well defined degeneracy in morphological appearances of the cultured organism, followed by its complete disappearance, and is effective in the treatment of the positive throat of diphtheria convalescents.

Wood⁸² in 1913, treated three diphtheria cases in/

in children by a simultaneous, but separate injection of a vaccine of 300 to 500 million bacilli, and 1000 units of antitoxin. This was followed up by two more doses of vaccine of 500 million bacilli, each at three day intervals. Cultures taken on the 10th day were found negative. This observer has used the above method for the past six years with great success. He advises the vaccine dosage to be regulated according to the severity of the case. Where there is great toxæmia, decrease the vaccine dose to 100 millions, and increase the antitoxin. The second dose in these severe cases may be 300 millions, and the third 500 millions.

Wood claims for this method that the production of toxin is stopped from the time the vaccine is administered, the patient now producing his own antitoxin. The antitoxin neutralises the toxin already formed. In carriers, the bacilli are destroyed. He also suggests its use to immunise contacts. He states that in cases of diphtheria, the throat is free within 10 days, thus shortening the quarantine period.

It is evident, that if these much to be desired results can be obtained by the relatively simple procedure of injecting three doses of diphtheria vaccine at appropriate intervals, the method is worth a trial.

I think this idea of administering a diphtheria vaccine therapeutically, at the same time as antitoxin, had occurred to me previous to seeing Wood's article, and in any case received considerable encouragement when I came across his results, which were brought to my notice by Dr Ker.

In October 1920 I made a tentative effort to clear up^a nasal carrier of 65 days duration, by administering a vaccine consisting of some eight strains of virulent diphtheria bacilli obtained from clinical cases. Three doses of 100, 150 and 200 million dead organisms were injected subcutaneously at four day intervals. The result on the bacilli in the throat was absolutely nil, and owing to the local reactions being marked, and also some slight constitutional reaction occurring, the treatment was not carried farther.

It was then that the idea of trying a detoxicated vaccine occurred to me. It was claimed for these preparations that very large doses could be given without any of the unpleasant local or general reactions one is so liable to get with even moderate doses of a vaccine prepared in the ordinary way.

Fraser and Duncan⁸³ using a detoxicated Klebs-Loeffler vaccine prepared according to the method of Thomson,⁸⁴ met with success in three persistent carrier cases. The first case^{which} had apparently been carrying for/

for 18 months, and certainly for three months at least, cleared up after nine injections of vaccine totalling 9,700 million organisms. The first six doses were given subcutaneously, and the latter intravenously, at four day intervals.

The second case was a nasal carrier of three years duration, who had given rise to three definite outbreaks of diphtheria with three deaths. She had a very unhealthy condition of the nose and nasopharynx. Efforts were made to improve the local condition and a course of '606' was given, but without result, diphtheria bacilli still persisted. After eight subcutaneous doses of vaccine extending over a period of 41 days, cultures did not contain diphtheria bacilli. In all, 21,700 million organisms were given in the eight doses. Two months later a non-virulent bacillus was again found in the nose, which disappeared in nine days, during which three doses of vaccine had been given. The maximum dose in one injection given in the second course, was 250,000 million organisms.

The third case was a soldier whose nose had been injured during the War. He had harboured virulent diphtheria bacilli in his nasal cavity for a period of 23 months. Twelve injections of vaccine extending over 53 days resulted in a non-virulent organism being/

being obtained from the nose in place of the previous persistently virulent strain. The dosage employed rose from 4000 million to the huge single dose of 350,000 million organisms. No reaction either general, or local, ensued in any of these cases, even with the enormous doses administered.

These results appeared distinctly encouraging.

Before proceeding to describe the vaccine treatment I carried out, it would perhaps be profitable to discuss the theory bearing on the disappearance of diphtheria bacilli from the local focus.

The relationship in which the individual stands to the organism he harbours in the carrier condition is an interesting problem. Why is it that in some convalescents the diphtheria bacilli should disappear so quickly, whilst in others they persist for a longer time, and in still others, luckily the minority, for even years?

Diphtheria is an example of a disease in which the causative organism remains localised at or near the point of invasion, usually in the throat or nose. The bacilli in this focus elaborate toxins, which are absorbed along the lymphatics, blood vessels, and nerves, and find their way to, and attack that part of the body for which they have a selective affinity. The body, no matter what the disease, strives to get rid/

rid of the offending parasite. In the case of diphtheria, the chief reaction is directed against the toxic substances or so called exotoxins, which are being passed into the body, and thus antitoxin is formed. In the case of bacterial diseases in which the micro-organism does not remain localised to the focus of infection, but invades the blood and tissues, and multiplies there, the chief forces of resistance are directed against the microbes themselves.

Let us consider for a moment what are the possible agencies by which the diphtheria bacilli can be destroyed and disappear. The three methods which appear to me to be rational are.- (1) by the bacteriolytic activity of the serum, (2) by phagocytosis, and (3) by such alterations in the environment or flora of the local focus as to render the pabulum inimical to the continued growth of the diphtheria organism.

Is the disappearance of the diphtheria bacilli in convalescents due to the bacteriolytic activity of the patients' serum?

The persistence of bacilli in the throat or nose in some cases, may be due to either a lack of sufficient antibactericidal substances in the tissues and plasma of the individual, or perhaps the bacilli may be in such a favourable situation that these substances, even though/

though in ample amount, cannot reach and act upon them. As an example of the latter, the bacilli situated in the debris at the bottom of a tonsillar crypt might be cited. If we could in some way stimulate the production of antibacterial substances it seems reasonable that the disappearance of the bacilli would be hastened. I have already stated that the reaction of the body is against the diphtheria toxin, and antitoxin is thus formed. It seems, however, that antitoxic sera have not only antitoxic properties, but also antimicrobial properties to a certain extent. Apparently however this antimicrobial property is not powerful enough to cause the disappearance of the bacilli in the local focus.

Certainly from clinical observation, antitoxin appears to have distinct antimicrobial powers. One has noticed time and again, how the spread of a diphtheritic membrane has stopped when an adequate dose of antitoxin has been administered, and enough time allowed to elapse for it to act. A sufficient dose of antitoxin given on the first day of clinical diphtheria will always prevent the development of the extensive patching so often seen in cases which do not come under medical care until the sixth or seventh day of illness. This clinical evidence rather points to the fact that the antitoxic serum contains some substance which prevents the multiplication of the bacilli in the local focus. Following this to a logical conclusion one/

one should find that by overdosing the cases the bacilli should disappear more quickly but it is not so. It seems that the antitoxin has more an inhibitory action than a real bactericidal action. Another explanation which appears to me reasonable is that the antitoxin by neutralising the toxin formed may prevent the extensive necrosis of tissue at the local focus, and hence the formation of a suitable pabulum for the diphtheria bacillus. In this case the action is purely antitoxic.

When an animal is injected with first dead, and then living virulent cultures of an organism, certain antibacterial properties are found to develop in the serum. The three main actions of such a serum are bactericidal, opsonic, and agglutinative.

If the disappearance of the diphtheria bacilli is due to bactericidal activity of the serum then vaccine administration on the face of it should be useful.

Lipstein immunised rabbits and guinea pigs with mixtures of diphtheria antitoxin and emulsion of virulent diphtheria bacilli. By this mixture he neutralised all traces of diphtheria toxin which adheres to, and are contained in the bacillary bodies. He concluded from a very thorough series of experiments that there was no amboceptor production and therefore no/

no bactericidal action.

Lambotte⁸⁵ on the other hand, found on injecting guinea pigs with an emulsion of diphtheria bacilli that the serum contained a specific bacteriolysin for diphtheria bacilli.

Several observers have obtained a serum with powerful agglutinative properties for the diphtheria bacillus by immunising horses first with dead and then living cultures of the bacilli.

Very recently Bell⁸⁶ has found it possible to classify a large number of strains of diphtheria bacilli by their agglutinative reactions.

He finds that whereas the bacterial antigen is specific, the toxin antigen is non specific.

The experimental evidence as to the production of bactericidal substances seems to be rather conflicting, and does not help one very much.

Several recent observers who have administered vaccine to convalescent carriers, have recorded previous to the disappearance of the diphtheria bacilli the presence of degenerated and involution forms. This appears suggestive of bactericidal activity.

According to Kolmer,⁸⁷ in bacteriolysis, loss of motility is followed by many of the bacilli becoming swollen and distorted, and later irregular or broken fragments or granules become apparent. In the numerous/

numerous routine examinations I have made on the persistence of bacilli in convalescent carriers, I have rather gained the impression that the bacillus retains its typical morphological appearance right up to the time of disappearance in most cases. In fact in a few cases in which the type has had some little peculiarity, I have been able to associate the organism under the microscope with a particular patient, and the organism has retained this more or less characteristic appearance right up to the time of disappearance. Were bacteriolysins the real factor in causing the disappearance of the bacilli in the throat, one would expect to find indications of degeneracy such as poor staining, variations in granularity, alteration in shape, etc. to be a constant feature previous to the ultimate disappearance of the bacilli. In some cases I have noticed these alterations in morphology suggestive of degeneration, which might be ascribed to the bactericidal action of the serum, but also equally well to many other local factors which may alter the suitability of the pabulum on which the bacillus has been persisting.

Is phagocytosis the important factor in the disposal of the diphtheria bacilli in the convalescent?

According to Wright the opsonic qualities of the serum constitute the means by which the body frees itself/

itself of the invading bacilli. In cases where the opsonic index is high, and phagocytosis active, the bacilli are rapidly destroyed.

In every attack of diphtheria there is a general leucocytosis, and on examining the structure of the membrane and contiguous tissues, numerous polymorpho-nuclears and endothelial cells are seen in the exudate. It is these cells, which in the presence of sufficient opsonin probably engulf and digest the diphtheria bacilli. In the case of a local infection like diphtheria, the opsonin in the focus of infection may be used up, and the general mechanism may not have been stimulated to produce a conquering amount of opsonin. The object of administering a vaccine is to stimulate the general production of opsonins so that the focus is flooded with a lymph rich in opsonins, and the bacilli are consequently phagocyted and destroyed. Dealing with this subject Weaver says that carriers usually have an abundant supply of opsonins, and their blood leucocytes are active, hence the uncertain value of vaccines.

It appears to me that phagocytosis may play a very important part in the disappearance of bacilli from the throat and nose of the diphtheria convalescent.

Does/

Does environment affect the life of the diphtheria bacillus in the diphtheria convalescent?

It is quite reasonable to suppose that in the persistent carrier the growth conditions in the local focus may be peculiarly suitable to the diphtheria bacillus. We know what slight differences in artificial media are sufficient to inhibit the growth of certain organisms; how some organisms grow best in symbiosis, and so on. The same factors may be at work in the local focus, throat or nose, in the convalescent. The method of treatment by the Staphylococcal spray aims at upsetting the favourable conditions for growth of the diphtheria bacillus.

Another very important factor is the actual situation of the bacilli. If situated in the debris in the tonsillar crypts then they are in^a peculiarly favourable position away from all destructive agencies.

Other important lines of defence in healthy individuals against bacterial invasion, are the epithelial layers of the mucous membranes the constant shedding of which is of great service. The mucous and saliva in nose and throat also have an important mechanical cleansing action, as also have the voluntary clearing of nose and throat and such actions as coughing and sneezing. These no doubt play a part in ridding the convalescent of diphtheria organisms.

THE ADMINISTRATION OF VACCINE. When one considers the best time to start administering the vaccine, one has the option of two methods. Should one wait for four weeks, by which time roughly 75 per cent of patients have rid themselves of diphtheria bacilli, and then administer vaccine to those who still harbour bacilli? On the other hand should one endeavour to counteract both toxin and bacilli at the earliest possible moment by the simultaneous, but separate injections of suitable doses of antitoxin and vaccine. This latter method rather appealed to me.

By this method of early administration, I hoped to shorten the life of the bacillus in the throat, and secondly to prevent the occurrence of the chronic carrier. Wood reported excellent results. Other considerations were to note any effect on the occurrence of paralysis, and lastly if it caused any difference in the percentage mortality.

I think it can be stated that in vaccine therapy, the amount of immunity produced is largely proportional to the quantity of vaccine injected. When using an ordinary autogenous diphtheria vaccine the dosage is seriously limited by the reactions produced. The usual dosage administered varies from 100 to 500 million/

million killed diphtheria bacilli. Detoxicated vaccines however, as prepared by Thomson, apparently can be used in very large doses without unpleasant reactions. Single doses up to 350,000 million have been given. In these vaccines the endotoxin is removed by chemical action without destroying the immunising properties of the vaccine.

Lees⁸⁸ speaks highly of the use of Thomson's detoxicated gonococcal vaccine in the treatment of gonorrhoea. He finds it useful both in early cases, and in chronic lesions. The vaccine stimulates the rapid production of antibodies in the early stages, and causes an increased production in the later stages. His initial dose is 2500 millions which is gradually increased to 10,000 millions, the injection being given subcutaneously every fourth day. He states that antibody can be detected in the blood from four to five months after treatment.

On application to Genatosan Ltd. a supply of detoxicated Klebs-Loeffler vaccine was kindly placed at my disposal. The first bottle of 25 ccs. contained 32,000 million bacilli to the cubic centimetre, the second lot I obtained more concentrated with 64,000 million bacilli in one cubic centimetre.

The next point to decide was the dosage to be used.

Brownlie, as already mentioned, using an ordinary diphtheria vaccine, successfully cleared up some 50 convalescent cases by injecting from 10 to 200 million bacilli in all. Wood administered three doses of 300 to 500 million bacilli at three day intervals. In studying the three carrier cases successfully treated with detoxicated diphtheria vaccine by Fraser and Duncan, I found that a negative was obtained in the first case after a dosage of 8900 millions, and in the second case after 20,700 million bacilli. The third case required 304,000 million bacilli in all. As I intended to give vaccine on the principle of the vaccine antitoxin treatment of Wood, the administration of a dosage approaching 300,000 million to each case would have been absolutely prohibitive in cost in a series of 150 or more cases.

On considering the successful results obtained by 10 to 500 millions of ordinary vaccine, and 9000 to 21,000 million of detoxicated vaccine, it appeared to me that an initial dose of 2000 millions, followed at three day intervals by 4000 millions, and 8000 million bacilli, should be fairly effective.

This dosage I adopted. The first dose of 2000 millions was given irrespective of age, or severity of disease, either on the day of admission, or on the following morning. The first injection was given subcutaneously in/

in the right forearm; the second in the left forearm; and the third in the right. All doses were given subcutaneously: none intravenously. The skin was prepared by a thorough cleansing with ether. The dose was accurately measured by means of an all glass three piece tuberculin syringe graduated in 1/100ths of a cubic centimetre. Previous to use, the syringe and needle were boiled for 10 minutes. Between each injection the needle was cleansed with ether. In no case did sepsis arise.

Reactions. The highest single dose of vaccine I injected was 20,000 million bacilli contained in 0.31 cubic centimetre approximately. The local reaction varied from a pin's head area of pinkness round the site of the needle prick, to a reddish pink, very slightly raised oval area some $1\frac{1}{2}$ " by $\frac{3}{4}$ ". Beyond slight tenderness to touch, in most cases there was no local discomfort. In all I administered 466 doses of vaccine ranging from 2000 millions to 20,000 millions, and with the exception of one case, I did not get any constitutional reaction. In one case there was a rise of temperature of 2° Fahrenheit, for which the only obvious explanation that could be found, was the injection of vaccine given some eight hours previously. I have not the least doubt that much/

much larger doses than 20,000 million may be given with impunity.

I endeavoured as far as possible to avoid any selection of cases. It was obvious from the start that I could not tackle every case of diphtheria that came into hospital. In as far as I confined myself to definite clinical cases of diphtheria, which had either been found positive previous to admission, or in which I found the diphtheria bacillus after admission, I plead guilty of selection. A certain percentage of cases sent in as diphtheria are found to be septic throats, Vincent's angina, and even Scarlatina, hence I had to exclude such cases from my series.

During the months of January and February I alternated cases which received vaccine, with non vaccine or control cases. My original idea was to continue this vaccine case, control case, alternately all through the series, but when I lost some four weeks owing to an attack of diphtheria I had to drop this idea, and then treated every case with vaccine. Towards the end of my series, when my vaccine stock was nearly finished, I again reverted to alternating control cases with vaccine cases.

Cultures from throat, and nose if necessary, were taken at weekly intervals in both series of cases until a negative was obtained. Cultures were then taken/

taken on alternate days, until three consecutive negatives were obtained. If a positive culture followed a negative culture, as often happened, the culture time reverted back to weekly intervals until another negative was obtained, when two day intervals were again started. If three negatives were obtained early in convalescence, some considerable time before the patient was discharged, then cultures were again taken in the week previous to discharge to confirm the previous negatives, and to see if reinfection arose during the stay in hospital. This means that many cases had as many as five and ^{some} even seven consecutive negative cultures previous to discharge.

The day half way between the date of the last positive culture, and the first of three consecutive negative cultures, was taken as the day on which the diphtheria bacilli were last present.

The results thus obtained gave the number of days diphtheria bacilli persisted in the convalescent throat after admission to hospital. In working out the total length of persistence of bacilli in the diphtheria patient I have calculated the number of days from the date of the first symptom to the final disappearance of bacilli, as judged by at least three consecutive negative cultures. It is evident that diphtheria bacilli must be present in the throat on the/

the first complaint of sore throat if we see the patient two or more days later with a definite clinical diphtheria. The error of inaccurate histories, particularly in children, creeps into this, but taken over a series of 200 odd cases I think the error is practically negligible.

In working out days of persistence some observers count from the date of disappearance of the membrane, others from the date on which bacilli were first found, and still others from the date of admission to hospital. None of these methods per se, will give total time of persistence, which can only be arrived at by considering the history. I admit the consideration of the history is not even accurate owing to the factor of the precocious carrier, still, some definite point has to be fixed, and I have chosen the day of commencement of the illness as evidenced by subjective symptoms.

Let me recapitulate the purpose of the investigation once again.-

- (1) Does the diphtheria bacillus disappear earlier from the throat and nose of the vaccine treated case, than the non-vaccine treated case?
- (2) Does the administration of detoxicated vaccine in the dosage indicated prevent the convalescent carrier from passing into the chronic carrier?

(3)/

- (3) Does the administration of detoxicated vaccine diminish the tendency to paralysis?
- (4) Does the administration of detoxicated vaccine alter the percentage mortality in any way?

In a series of 207 cases, 146 received three injections of detoxicated Klebs-Loeffler vaccine of 2000, 4000, and 8000 million bacilli respectively, whilst 61 were control or non vaccine cases.

| Type of Case. | Jan ^r . | Feb ^r . | Mar. | April-May | July-Aug. | Sept. | Totals |
|---------------|--------------------|--------------------|------|-----------|-----------|-------|--------|
| Non Vaccine | 25 | 15 | 0 | 0 | 8 | 13 | 61 |
| Vaccine | 25 | 14 | 23 | 27 | 34 | 23 | 146 |
| Total | 50 | 29 | 23 | 27 | 42 | 36 | 207 |

Day of Disease on Admission to Hospital.

| Day of Disease. | 1st | 2nd | 3rd | 4th | 5th | 6th | 7th | 8th | 9th | 10th | 11th | 12th | 13th |
|-----------------|-----|-----|-----|-----|-----|-----|-----|-----|-----|------|------|------|------|
| Non Vaccine | 3 | 10 | 16 | 16 | 7 | 3 | 1 | 3 | 0 | 1 | 0 | 0 | 1 |
| Vaccine | 3 | 32 | 34 | 40 | 18 | 7 | 4 | 4 | 1 | 1 | 0 | 1 | 0 |
| Total. | 6 | 42 | 50 | 56 | 25 | 10 | 5 | 7 | 1 | 2 | 0 | 1 | 1 |

Mean number of days of illness previous to admission to hospital in:-

Non Vaccine Cases = 3.96 days.

Vaccine Cases = 3.77 days.

Dividing the cases up into those admitted up to, and on the fourth day of illness, and secondly, those admitted later we find:-

| Type of Case | 1st to 4th day of illness inclusive. | 5th day of illness and later. |
|-------------------|--------------------------------------|-------------------------------|
| Non Vaccine Cases | 73.77% | 26.23% |
| Vaccine Cases | 75.35% | 24.65% |

These figures show that treatment was started on approximately the same day of illness in both series of cases. If anything, the non-vaccine series received their first dose of antitoxin a shade later than the vaccine cases.

(1) Do diphtheria bacilli disappear more quickly from the throat and nose of the vaccine treated case, than the non-vaccine treated case?

55 Non Vaccine Cases.

Mean persistence of diphtheria bacilli in throat or nose after disappearance of membrane. = 16.51 days.

144 Vaccine Cases.

Mean persistence of diphtheria bacilli in throat or nose after disappearance of membrane. = 14.08 days.

Mean Persistence of diphtheria bacilli in throat or nose in whole series of 199 cases after disappearance of membrane. = 14.75 days.

Is the duration of Membrane affected by the administration of vaccine?

Mean Persistence of membrane in:-

| | | |
|----------------------|---|------------|
| 55 Non Vaccine Cases | = | 6.47 days. |
| 144 Vaccine Cases | = | 6.53 " |

The duration of membrane is calculated from the day of first symptom to its final disappearance.

Apparently the administration of vaccine did not hasten the disappearance of the membrane. When one considers the fact that the cases of the vaccine series had already been ill on an average 3.77 days previous to the administration of the vaccine it is evident that the vaccine has not much of an opportunity to curtail the duration of the membrane.

The total number of days which the bacilli persist in convalescents, I have computed by adding together the above two figures, namely duration of membrane, plus persistence of bacilli after membrane disappears.

Total persistence of diphtheria bacilli in throat and nose from first symptom to final disappearance.

| | | |
|------------------------------------|---|-------------|
| 55 Non Vaccine Cases | = | 22.98 days |
| 144 Vaccine Cases | = | 20.61 days |
| 199 Non Vaccine plus Vaccine Cases | = | 21.27 days. |

We thus see that the administration of vaccine has shortened the stay of diphtheria bacilli in the convalescent by 2.37 days. I am afraid this result can be explained by accidental circumstances, and not to any beneficial effect of the vaccine.

Is duration of persistence affected by season?

These figures have no particular bearing on the question of the efficacy of vaccine. I wish to see however if my findings agree with Walsh who found the period was four days shorter in Summer than in Winter, 20 and 24 days respectively.

| Month. | Total Cases. | Total Days of persistence. |
|-------------|--------------|----------------------------|
| Jan. | 25 | 22.64 |
| Feb. | 14 | 23.35 |
| March | 23 | 19.04 |
| April - May | 27 | 17.61 |
| July - Aug. | 34 | 23.24 |
| Sept. | 23 | 17.96 |

The figure for July - August, namely 23.24 days is unduly high, owing to the fact that there were several very persistent nasal carriers; in fact one case was inadvertently discharged after 78 days with diphtheria/

diphtheria bacilli still present in the nose.

Discounting the July - August figures we see that the persistence of bacilli in convalescents in the warmer months April, May and September is some three to five days less than in January or February which are the coldest months in Edinburgh. These figures support Walsh's findings.

It is interesting to compare the persistence in nasal carriers with that in faucial carriers.

| | Non Vaccine Cases. | Vaccine Cases. |
|------------------|--------------------|----------------|
| Nasal diphtheria | 4 | 13 |
| Mean persistence | 36 days | * 39 days |

* One case still positive after 120 days.

We thus see that diphtheria bacilli persist longer in the nose than in the throat. The administration of vaccine has had no effect.

The following table shewing the percentage rate of disappearance is interesting:-

TABLE./

| Days from onset of disease. | NON VACCINE CASES. | | | VACCINE CASES. | | |
|-----------------------------|--|-------------|--|----------------|--|--|
| | Number of Cases which became negative. | Percentage. | Number of Cases which became negative. | Percentage. | | |
| 0 - 7 | 6 | 11.11% | 10 | 7.09% | | |
| 8 - 14 | 14 | 37.03% | 51 | 43.26% | | |
| 15 - 21 | 7 | 50.00% | 23 | 59.57% | | |
| 22 - 28 | 13 | 74.07% | 25 | 77.30% | | |
| 29 - 35 | 4 | 81.48% | 16 | 88.65% | | |
| 36 - 42 | 6 | 92.59% | 6 | 92.90% | | |
| 43 - 49 | 1 | 94.44% | 6 | 97.16% | | |
| 50 - 56 | 3 | 100.00% | 0 | 97.16% | | |
| 57 - 63 | | | 3 | 99.29% | | |
| 64 - 70 | | | | | | |
| 71 - 77 | | | | | | |

One case with diphtheria bacilli still in nose 125 days after first symptom.

This table shows that in the non vaccine cases 81.48 per cent were free from bacilli at the end of 35 days; in the vaccine series a higher percentage was free of bacilli within 35 days, namely 88.65 per cent. It is interesting to compare these results with other observers already quoted in this paper. After 35 days 79.87, 88.88, 89.1, 93.0 per cent of diphtheria convalescents have rid themselves of diphtheria bacilli according to Hartley and Martin, Weaver, Tjaden and Wesbrook respectively.

This table also shows that the administration of vaccine has had no effect in hastening the disappearance of the diphtheria bacilli.

(2) Does the administration of vaccine prevent the chronic carrier?

This is a question in which I was very interested, and confess somewhat hopeful.

From the foregoing table we see that three cases harboured bacilli up to the eighth week, namely for 57, 58 and 61 days. Of these, two were faucial, and one a nasal carrier. As these cases still had bacilli present some 50 days after the administration of the vaccine, it was apparent the vaccine did not prevent/

prevent the prolonged persistence of bacilli. One case, treated with three doses of vaccine, a faucial and nasal diphtheria on admission, still harboured diphtheria bacilli in the nose at the time of writing, namely 125 days after the commencement of the illness.

(3) Does the administration of vaccine diminish the incidence of paralysis?

The first method of comparing the two series of cases in regard to the incidence of paresis is to work out the percentage incidence in either group only in cases that have received more than 6000 units of antitoxin. As a rule we expect paresis to occur only in the more severe cases.

| | Vaccine | Non Vaccine |
|--|---------|-------------|
| Cases receiving 6000 units of antitoxin and under | 112 | 42 |
| Cases receiving more than 6000 units of antitoxin | 34 | 18 |
| Percentage of cases receiving more than 6000 units of antitoxin. | 23.28% | 30% |

From these figures we see that there was a relatively higher proportion of severe cases in the non-vaccine group than in the vaccine group.

Cases receiving over 6000 units of antitoxin.

| | <u>Vaccine Cases.</u> | <u>Non Vaccine</u> |
|--|---------------------------|------------------------|
| Number of days ill previous to administration of antitoxin | 3.88 | 5.00 |

Incidence of Paralysis.

| | Vaccine Cases. | Non Vaccine. |
|--|-------------------|-----------------|
| Number of cases developing paralysis in cases receiving 6000 units and under. | 1 | 3 |
| Number of cases developing paralysis in cases receiving over 6000 units antitoxin. | 7 | 3 |
| | 8 | 6 |

In vaccine cases incidence of paralysis was 23.52% of cases receiving above 6000 units antitoxin.

In non vaccine cases incidence of paralysis was 33.33% of cases receiving above 6000 units antitoxin.

The localisation of paralysis in the two groups was as follows:-

TABLE/

VACCINE CASES.

| Day of Disease on admission. | Palate | Eye Muscles | Face | Neck | Pharynx | Legs | Dosage of Antitoxin | Type. |
|------------------------------|--------------|-------------|------|------|---------|------|---------------------|-------|
| 3rd | - | 50th day | - | - | 53rd | - | 20,000 | F + M |
| 12th | On admission | - | 55th | 55th | - | 51st | 12,000 | F |
| 4th | 24th day | 39th | 42nd | 45th | - | 34th | 32,000 | F + L |
| 3rd | - | - | - | - | - | 49th | 14,000 | F |
| 3rd | - | - | - | - | - | 42nd | 5,000 | F |
| 3rd | 9th | 45th | - | - | - | - | 8,000 | F |
| 5th | 43rd | - | - | - | - | - | 16,000 | F |
| 5th | 18th & 46th | 41st | 46th | 54th | - | 55th | 24,000 | F |

NON VACCINE CASES.

| Day of Disease on admission. | Palate. | Eye Muscles | Face | Neck | Pharynx | Legs. | Dosage of Antitoxin. | Type. |
|------------------------------|----------|-------------|------|------|---------|-------|----------------------|-------|
| 5th | - | - | - | - | - | 31st | 4,000 | F |
| 5th | 10th day | - | - | - | - | - | 20,000 (Died) | F.N. |
| 4th | - | 43rd | - | - | - | - | 6,000 | F |
| 9th | 36th | - | - | - | - | - | 6,000 | F |
| 4th | 16th | - | - | - | - | - | 9,000 | F |
| 6th | 9th | - | - | - | - | - | 32,000 (Died) | F.N. |

The date of onset of paralysis is calculated from day of admission to hospital.

F = faucial type.

N = nasal type.

L = laryngeal type.

Looking superficially at the percentage given above, it appears that the administration of vaccine has been instrumental in reducing the incidence of paralysis by some 10 per cent. Before we accept this figure as it stands, we must take into account the fact that the vaccine cases which received over 6000 units of antitoxin got their first injection after being ill on an average 3.88 days whereas the non vaccine cases of over 6000 units did not get their antitoxin until after five days of illness. Now I think it is an accepted fact that other things being equal, the longer the administration of antitoxin is delayed, the more the toxin is likely to get a hold, and the higher the incidence of paralysis.

When we compare the two tables showing the parts of the body involved, the most striking feature is the extensive involvement in several of the vaccine cases. Whereas in the non vaccine cases only the palate, eye or legs were affected, in the vaccine series one finds all these affected in the same individual in several cases. We must note however, that the cases in the vaccine series, as judged by the amount of antitoxin they received, were relatively more severe than in the non vaccine series, and secondly that the two cases in the non vaccine group which received 20,000 and 32,000 units/

units of antitoxin both died on the twelfth day after admission, hence did not get an opportunity to develop the various later paralysis which would in all probability have occurred.

Does the administration of vaccine alter the mortality rate in any way?

As all hospital treatment is performed with the aim of preventing the death of the patient if possible, the importance of the above question is paramount.

| | Vaccine Cases. | Non Vaccine Cases. |
|---|----------------|--------------------|
| Number of Deaths. | 1 | 6 |
| Number of cases receiving over 6000 units antitoxin | 34 | 18 |
| Percentage deaths of cases receiving over 6000 units. | 2.94% | 33.33% |

As the total deaths are only seven I shall give the cases in detail.

TABLE/

DEATH IN VACCINE CASES.

| Age | Sex | Day of Disease on admission. | Type | Antitoxin | Days membrane persisted | Vaccine Dosage | Albumin in urine. | Glands | Paralysis | Day of death. |
|-----|-----|------------------------------|-------|-----------|-------------------------|----------------------|-------------------|--------------------|-----------|------------------------|
| 2 | F | 4th | F + N | 10,000 | 10 | 2000 mill. 4000 " | + | marked enlargement | nil | 7th day Heart failure. |

DEATHS IN NON VACCINE CASES.

| | | | | | | | | | | |
|-----------------|---|------|-------|--------|----|-----|---|-------------------------|-----------------|-----------------|
| $\frac{8}{212}$ | F | 5th | F + N | 20,000 | 7 | nil | + | very marked enlargement | Palate 10th day | 12th heart |
| $\frac{10}{12}$ | M | 13th | F + L | 12,000 | - | - | + | nil | - | 10th Bronchopn. |
| $\frac{5}{112}$ | F | 3rd | L.N. | 6,000 | - | - | + | nil | - | 21st Bronchopn. |
| 4 | M | 2nd | F + N | 12,000 | 7 | nil | + | very marked enlargement | - | 16th Heart. |
| $\frac{2}{112}$ | F | 8th | F.M+L | 20,000 | 9 | nil | + | nil | - | 2nd Heart. |
| 12 | M | 6th | F + N | 32,000 | 17 | nil | + | very marked enlargement | Palate 9th | 12th Heart. |

Even excluding the two cases with Bronchopneumonia on admission, and the case that died on the day after admission, we still have three deaths out of 18 severe non vaccine cases, which gives a percentage of 16.66% as compared with 2.94% in the vaccine series.

From a table of diphtheria death rates classified according to the day of illness on which serum was first injected we find that

| Day of illness on which serum first injected. | * Percentage Mortality | Cases in Series. | |
|---|------------------------|-------------------------------|-------------|
| | | Cases of 6000 units and over. | |
| | | Vaccine | Non Vaccine |
| Third | 6.85% | 2.94% | - |
| Fourth | 10.91% | | - |
| Fifth | 14.92% | - | 16.66% |

* Ker.

The average mortality for 8,591 cases was 8.33%.

It is interesting to note that six out of the seven deaths occurred in cases with a nasal diphtheria as well as a faucial involvement.

In order to compare the mortality rate between the vaccine and non vaccine series, perhaps the fairest way would be to take the cases in each series which had both faucial and nasal diphtheria on admission, (the latter indicated by a profuse mucopurulent or sanguinous discharge containing numerous diphtheria bacilli).

We/

We thus find:-

| | Non Vaccine Series. | Vaccine Series. |
|---|---------------------------|---------------------|
| Cases with both faucial and nasal diphtheria on admission | 7 | 14 |
| Mean age of patients | 4 $\frac{1}{2}$ years | 4 $\frac{1}{2}$ yrs |
| Mean days of illness before admission to hospital | 3.85 days | 3.92 days |
| Average dose of antitoxin administered per case | 16,500 units | 11,600 units |
| Cases developing paralysis | 3 | 2 |
| Deaths | 3 | 1 |
| Death rate per cent. | 42.85% | 7.14% |

Even though we take into consideration that the non vaccine cases received on an average 5000 units antitoxin more per case and hence were evidently of a more severe nature than the vaccine series, yet the discrepancy in the percentage mortality is remarkable. The fallacy lies in the small number of cases in the series. To overcome this difficulty to some extent I have worked out the percentage mortality in a series of 65 faucial and nasal diphtheria cases admitted to the hospital. The mortality in these cases works out at 26.15%, still much in excess of the 7.14% in vaccine cases.

These two series of cases are exactly comparable in/

in type of disease, age, duration of illness previous to administration of antitoxin, and have been treated in exactly the same way except for the administration of vaccine, yet show the above great variation in percentage mortality. I would not like to state that the vaccine was responsible for the improvement, yet the result is interesting. A death rate of three out of seven faucial and nasal diphtheria cases suggests that a run of rather unusually severe cases were encountered, and happened to fall into the non-vaccine series. The average mortality for faucial and nasal diphtheria cases in this hospital works out at 25 per cent to 30 per cent.

CONCLUSIONS.

- I. From the figures I have worked out, it appears that the administration of detoxicated vaccine in the particular manner I have adopted, does not have any appreciable effect in hastening the disappearance of the diphtheria bacillus from the throat or nose of the diphtheria convalescent.
- II. The vaccine does not prevent those cases of prolonged persistence to which, after an arbitrary/

arbitrary period of three months, we apply the term chronic carrier.

III. I do not think the administration of the vaccine has diminished the onset of paralysis to any extent.

IV. If we discount the small number of cases on which the non vaccine results are calculated, the administration of vaccine has diminished the percentage mortality very considerably. This may be a pure co-incidence, but is nevertheless interesting enough to warrant a farther trial in some two hundred or so severe diphtheria cases with both faucial and nasal involvement. I would suggest intravenous injections be used to some extent.

Treatment of Chronic Carriers
with detoxicated Klebs-Loeffler Vaccine.

Entirely apart from the above method of giving three doses of vaccine to clinical diphtheria cases on admission, I also treated several persistent carriers in the hope of ridding them of bacilli. I shall give these cases in some detail.

CASE I./

CASE I. Female. Nasal carrier. 1500 units anti-toxin.

Twenty-nine nasal cultures taken between 15.11.20 and 18.1.21 all showed numerous diphtheria bacilli. Detoxicated Klebs-Loeffler vaccine was then administered as follows:-

19.1.21. 2000 million bacilli.
 Culture 20.1.21. + +
 " 23.1.21. 4000 million bacilli. Culture + +
 Small bone button removed from left nostril.
 " 25.1.21. neg.
 " 26.1.21. +
 27.1.21. 8000 million bacilli.
 " 28.1.21. neg. (T & N)
 " 29.1.21. neg. (T & N)
 " 30.1.21. neg. (T & N)

In this case the button in all probability was the cause of the persistent carrier condition, and its removal, in my opinion, was the decisive factor in causing the disappearance of diphtheria bacilli, though the vaccine may have had an accessory action.

Carried for 140 days in nose.

CASE II. Adult female. Faucial carrier.

500 units antitoxin.

Tonsils very large with marked crypts.

Fourteen consecutive faucial cultures taken in the first 32 days after admission, with one exception, showed numerous diphtheria bacilli.

| | | <u>Dose of Vaccine.</u> | <u>Culture.</u> |
|---------------------------|-----|--------------------------------|-----------------|
| 33rd day after admission. | | 2000 million bacilli injected. | Not examined. |
| 34th | " " | | + + |
| 36th | " " | | + |
| 37th | " " | 4000 " " | Not examined. |
| 39th | " " | | + + |
| 41st | " " | 8000 " " | + (scanty) |
| 43rd | " " | | + |
| 45th | " " | | + + |
| 46th | " " | 10,000 " " | Not examined. |
| 47th | " " | | + + |
| 49th | " " | | + + |

Tonsillitis developed with broken exudate on both tonsils. Temperature 102° and pulse 106. Submaxillary glands enlarged.

| | | |
|---------------------------|-----|-----------|
| 56th day after admission. | | neg. |
| 68 | " " | neg. |
| 69th & 75th | | neg. neg. |

This case carried for 53 days.

Note/

Note marked enlargement of tonsils with large crypts, which I rather tend to associate with the chronic carrier condition. Was the vaccine responsible for the development of the apparent clinical diphtheria after some six weeks in hospital? If so the vaccine must have lowered, instead of increased the resistive power of the body to diphtheria. The appearance of the throat together with the enlargement of the submaxillary glands rather suggested a superadded septic infection might have been the cause of the mischief, and incidentally the causal factor in the disappearance of the diphtheria bacilli. I might mention that 53 days is not long enough to consider a case a persistent carrier.

CASE III. Adult female. Faucial carrier.

500 units antitoxin.

Fifteen consecutive faucial cultures over a period of 26 days after admission, showed diphtheria bacilli. As we were particularly desirous of ridding her of bacilli at the earliest date, vaccine administration was commenced.

29th/

| | <u>Vaccine.</u> | <u>Faucial Culture.</u> |
|--------------------------|-----------------------|-------------------------|
| 29th day after admission | | + |
| 31st " " | 2000 million bacilli. | neg. |
| 32nd " " | | neg. |
| 33rd " " | | neg. |

Carried for 30 days.

Even the most sanguine vaccine supporter can hardly in all fairness ascribe the rapid disappearance of bacilli to the administration of one dose of vaccine. This illustrates how the bacilli disappear spontaneously, and how this is apt to give an erroneous impression as to the efficacy of the particular method of destruction being employed.

CASE IV. Male, age 6. Nasal carrier.

1500 units antitoxin.

Twenty-two nasal cultures, with one exception, taken over a period of 49 days showed numerous diphtheria bacilli.

| | <u>Vaccine.</u> | <u>Nasal Culture.</u> |
|--------------------------|-----------------------|-----------------------|
| 67th day after admission | | + + |
| 68th " " | 2000 million bacilli. | |
| 69th/ | | |

| | | <u>Vaccine.</u> | <u>Nasal Culture.</u> | | |
|------|---------------------|-----------------------|-----------------------|---|------|
| 69th | day after admission | | + | + | + |
| 72nd | " " | 4000 million bacilli. | | | |
| 74th | " " | | | + | + |
| 77th | " " | 8000 " " | | | |
| 81st | " " | | | | + |
| 83rd | " " | 10,000 " " | | | |
| 84th | " " | | | | neg. |
| 85th | " " | | | | + |
| 88th | " " | | | | neg. |
| 89th | " " | | | | neg. |
| 90th | " " | | | | neg. |
| 91st | " " | | | | neg. |

Carried for 86 days.

The vaccine in this case has apparently been successful.

CASE V. Female, age 57. Mild faucial diphtheria.

4000 units antitoxin.

Tonsils very large with big crypts.

Fifteen faucial cultures over a period of 45 days after admission showed numerous diphtheria bacilli.

| | <u>Dose of Vaccine.</u> | <u>Faucial Culture.</u> | |
|------------------|-------------------------|-------------------------|---|
| 46th day | 2000 million bac. | | |
| 47th & 49th days | | + | + |
| 50th day | 4000 " " | | |
| 51st/ | | | |

| | <u>Dose of Vaccine.</u> | <u>Faucial Culture.</u> |
|-------------|--------------------------|-------------------------|
| 51st day | | + |
| 54th " | 8000 million bacilli. | + |
| 56th & 57th | | neg. & + + |
| 58th day | 10,000 " | |
| 59th & 61st | | + |
| 62nd day | 16,000 " | |
| 63rd & 65th | | + |
| 67th day | 20,000 " | |
| 69th " | | neg. |
| 70th " | | + + |
| 71st " | Tonsils removed. | |
| 73rd " | | + (sc) |
| 75th " | | neg. |
| 76th " | | + |
| 78th " | | neg. |
| 79th " | | + (sc) |
| 80th " | | neg. |
| 81st " | | neg. |
| 82nd " | | neg. |

Carried for 79 days.

The vaccine in this case apparently was a failure. The tonsillectomy however was eminently successful, the patient being free of bacilli ten days after the operation.

CASE VI. Female, age 5. Scarlatina and faucial carrier. Eight faucial cultures taken between 6.2.21 and 14.3.21 always showed diphtheria bacilli.

Dose of Vaccine. Faucial Culture.

| | | | | | |
|---------------------------------------|-----------------------|--|--|---------------|---|
| 37th day from first positive culture. | | | | + | + |
| 38th " " " | 2000 million bacilli. | | | | |
| 39th, 40th, 41st days | | | | + | |
| 42nd " | 4000 " " | | | | |
| 44th " | | | | + | |
| 46th " | 8000 " " | | | + | |
| 52nd " | 12,000 " " | | | | |
| 53rd, 55th & 56th | | | | All negative. | |

Carrying for 52 days from day on which first culture was taken and found positive.

Vaccine here was apparently successful. She can hardly be regarded as a persistent carrier.

CASE VII. Female, age 13. Scarlatina and faucial carrier. Carrying diphtheria bacilli in throat for 37 days previous to administration of vaccine.

| | <u>Dose of Vaccine.</u> | <u>Faucial Culture.</u> |
|----------|-------------------------|-------------------------|
| 38th day | 2000 million bacilli | |
| 42nd " | 4000 " " | + |
| 46th " | 8000 " " | + + |
| 50th " | 8000 " " | + |

On 51st day a sharp staphylococcal tonsillitis developed with some constitutional disturbance.

Three consecutive negatives obtained on 52nd, 54th and 55th days after first culture was taken.

In this case the superadded staphylococcal infection apparently played a large part in causing the disappearance of the diphtheria bacilli.

CASE VIII. Female age 7. Scarlatina and nasal carrier. Numerous diphtheria bacilli in nasal cultures for 24 days previous to administration of vaccine.

| | | | | |
|----------------|----------|----------------------|---|---|
| Vaccine dosage | 25th day | 2000 million bacilli | | |
| | 30th " | 4000 | " | " |
| | 34th " | 8000 | " | " |
| | 37th " | 8000 | " | " |

Diphtheria bacilli were still present in large numbers in cultures from the nose up to the 40th day; they then diminished in number, and disappeared on the 47th day; three consecutive negative nasal cultures being obtained on the 48th, 49th and 50th days from first examination.

Carried for 47 days.

The carrying period is again too short to lay much weight on the result. Note that the bacilli did not disappear from the nose for ten days after the fourth dose of vaccine. The beneficial effect of vaccine in this case is extremely doubtful.

CASE IX. Male, age 9. Scarlatina. Nasal carrier.
Diphtheria bacilli in nose for 37 days.
previous to vaccine administration.

38th day 2000 million bacilli.

39th, 40th & 41st days three negative results.

This is another case exemplifying the spontaneous disappearance of the diphtheria organisms. It is hardly likely that the dose of vaccine given on the 38th day would be so rapidly effective.

Results of vaccine administration in above nine carrier cases - five faucial and four nasal.-

| | | |
|-------------------|-----|------------------------------------|
| Result Successful | = 2 | One nasal and one faucial carrier. |
| " Doubtful | = 4 | Two nasal and two faucial. |
| " Unsuccessful | = 1 | Faucial carrier. |
| No conclusion | = 2 | One faucial and one nasal. |

REMOVAL OF TONSILS AND ADENOIDS.

I have noticed in many of the cases which turn into persistent carriers, that the tonsils are unusually large, with very irregular surface, and large crypts; in fact the type of tonsil which would present a suitable nidus for the diphtheria bacillus.

Stitt⁸⁹ draws attention to the fact that the crypt of the tonsils may harbour the bacilli, and thus protect them from the ordinary application of antiseptics.

Going on this assumption that the tonsillar crypts are the seat of residence of the bacilli, it seems reasonable to expect, that when the tonsils are removed, the throat should become free of bacilli.

According to Ker⁹⁰ however, tonsillectomy is sometimes, but by no means always effective in obstinate cases.

Ledingham and Arkwright⁹¹ state that extirpation of the tonsils in faucial carriers is likely to prove very useful as a means of getting rid of the bacilli.

Apparently Pegler⁹² suggested this method as far back as 1905. He had several times found that careful extirpation by means of morcellement of unhealthy/

unhealthy tonsil tissue in which crypts could be discovered led to the carrier condition coming to an end. In other cases he found that adenoid or naso-pharyngeal, tonsillar tissue was the source of trouble.

Simon⁹³ relates a case of a faucial carrier who had been quarantined and treated by various applications, even injections of silver nitrate into the substance of the tonsils, for a period of 136 days without result. Tonsillectomy was then performed, and repeated cultures taken 30 days later were negative, showing the diphtheria bacilli had disappeared. He also reports successful results obtained by tonsillectomy in Camp Sherman by McCord, Friedlander and Walker.

Tonsillectomy was performed in 294 cases at Camp Doniphan. All the cases, with one exception, became negative by the end of eight weeks. The remaining case still persisted positive.

Lynch⁹⁴ states that tonsillectomy finally removes the source of supply of diphtheria bacilli, and he has not seen or read of a failure, nor has he had any bad results.

Hartley and Martin⁹⁵ encountered four persistent carriers with abnormally large tonsils with deep crypts. They agree with Pegler and Sears that the best/

best treatment of such cases is radical enucleation of the tonsils. In all four cases this was done, and they all became negative. On section of the tonsils they observed the diphtheria bacilli deep down in the crypts, which in one case were over one centimetre in length. They state that the bacilli did not invade the tissue of the tonsil.

Arkwright⁹⁶ regards radical enucleation as a hopeful and justifiable procedure in those faucial carriers where the tonsils appear to be the seat of the bacilli. He recommends a prophylactic dose of antitoxin should be given before the operation.

Weaver⁹⁷ recommends tonsillectomy and adenoidectomy when other measures fail. Early disappearance of the bacilli has followed the operation in every case. In a period of five years 40 patients had been operated on. In all cases the tonsils were removed, and in five adenoids were also removed. All the cases were convalescing from clinical diphtheria, and were operated on from 21 to 73 days after the onset. Many became negative at once, and all except four, were negative within a week. One took 18 days to become free from diphtheria bacilli. He advises removal of the tonsils and enlarged adenoids at the end of a month if the bacilli persisted, and the patient's general condition be suitable. In small children/

children he prefers to wait for the natural disappearance of the bacilli. In the case of persistent nasal carriers he always looks for local lesions and foreign bodies.

It was pointed out by Friedberg that the tonsil harbouring the diphtheria bacilli need not be enlarged. This statement I quite agree with. I have seen cases of prolonged persistence with quite inconspicuous tonsils.

Ballantyne and Cornell⁹⁸ report six cases which were treated by complete enucleation of the tonsils. These cases had been carrying diphtheria bacilli in the fauces for 10 to 42 days. The organisms disappeared in all cases with the healing of the wound. It is interesting to note that in four of the six cases the pathological findings showed the diphtheria bacilli in the very bottom of the crypts.

According to Scholes⁹⁹ the most rational treatment is the removal of tonsils and adenoids, as the former are almost invariably diseased in persistent carrier cases.

It is evident from even such a brief survey of the literature that these surgical procedures for the treatment of carriers have met with a very considerable measure of success.

I have had the opportunity of observing the result of tonsillectomy and adenoidectomy in probably a dozen cases or more, but I shall only describe six which happened to come into my series of cases. In these cases various methods of treatment had been tried, such as endotoxin, and detoxicated vaccine administration, but without success, and surgical interference was considered justifiable.

The following are the cases in detail:-

CASE I. Female, age 23. Mild faucial diphtheria. 3000 units antitoxin. Tonsils very large. Diphtheria bacilli still present in throat 50 days after admission. Tonsillectomy was performed on 51st day after admission. Three consecutive negative cultures were obtained on 10th, 11th and 13th days after tonsillectomy or 61st, 62nd and 64th days after admission.

Tonsillectomy in this case succeeded in ridding the throat of bacilli within 10 days after the operation.

CASE II. Female, age 57. Mild faucial diphtheria. 4000 units antitoxin. Tonsils very large with deep crypts.

This case received six doses of detoxicated Klebs-Loeffler vaccine ranging from 2000 to 20,000 million bacilli/

bacilli but without success.

Diphtheria bacilli were still present in the throat 71 days after admission to hospital.

Tonsillectomy was performed on the 72nd day.

Diphtheria bacilli disappeared from the throat on the 81st day, i.e. nine days after the operation.

On examining sections of the tonsil from this case, diphtheria bacilli were seen in the bottom of a crypt.

CASE III. Female, age 18. Mild faucial diphtheria.

1500 units antitoxin. Diphtheria bacilli were still present in the throat 67 days after admission. The tonsils were removed on the 68th day. Three consecutive negative cultures from the throat were obtained on the 76th, 77th and 79th days.

Tonsillectomy had been successful in freeing the throat of diphtheria bacilli within a week of the operation.

CASE IV. Male, age $1\frac{1}{2}$. Nasal diphtheria. 12,000 units.

Previous to tonsillectomy and adenoidectomy, this case had been treated with three doses of 50, 100, and 200 million autogenous diphtheria vaccine, and also had received four doses of Hewlett's endotoxin/

endotoxin but without success. Diphtheria bacilli persisted in the nose for 166 days prior to surgical interference. On the 167th day, tonsils and adenoids were removed. Two consecutive negatives were obtained on the 187th and 188th days after admission, i.e. the 19th day after the operation.

The fallacy in this case is that only two negative cultures were obtained previous to discharge.

CASE V. Female, age 9. Mild faucial diphtheria and nasal carrier. 3000 units antitoxin.

This case received, previous to operative procedure, four doses of Hewlett's endotoxin but without success. Diphtheria bacilli having persisted in the nose for 106 days after admission, tonsillectomy and adenoidectomy was performed on the 107th day.

Three consecutive negative cultures were obtained on the 113th, 114th and 115th days after admission or within a week after the operation.

CASE VI. Male, age 2. Faucial and nasal diphtheria. 6000 units antitoxin.

This case was treated on admission with three injections of 2000, 4000 and 8000 million detoxicated diphtheria vaccine. Diphtheria bacilli having persisted in the nose for 63 days after admission the/

the tonsils and adenoids were removed on the 64th day.

Diphtheria bacilli were still present in the nose 60 days after the operation.

Summarising these results we see that tonsillectomy cleared up three faucial carriers within ten days of the operation.

The removal of tonsils and adenoids in three nasal carriers was successful in two, but a complete failure in the third.

These results are very gratifying.

That the third nasal carrier, in which operative procedures proved useless, was harbouring a virulent diphtheria organism, was proved by the fact that he was inadvertently discharged on his 78th day, and in all probability was the source of infection of his sister, who was admitted some ten days later suffering from typical clinical and bacteriological diphtheria. In this carrier, we could not find any evidence of foreign bodies, or nasal abnormalities, which should always be looked for in chronic nasal carriers.

BACTERIOLOGY OF THE CASES EXAMINED.

In all, 2003 cultures were examined in the course of the above investigations.

The swabs were taken daily, usually between 2 p.m. and 4 p.m. Sterilised swabs were always used. In swabbing, particular efforts were made to thoroughly go over both tonsils, pillars of fauces, posterior pharyngeal wall, and any membrane that happened to be present. The blood serum culture tubes were immediately inoculated at the bedside by rubbing the swab firmly over the surface of the media, turning it round in the process. The culture tubes were then placed in the incubator at 37° C. until 9.30 a.m. the following day, thus giving 18 to 19 hours incubation. In making films, the sterilised platinum needle was drawn freely over the whole surface of media where growth was apparent. The film was then dried, and fixed by heat, stained by means of Neisser's modified cresoidin method, and examined by a 1/12 inch oil immersion objective. In staining, I found that 8 - 10 seconds with the methylene blue crystal violet solution, and 12 - 15 seconds with the cresoidin counterstain gave good results. Previous to deciding on this particular stain, /

stain, I gave a trial to Neisser's original method using Bismarck brown as counterstain, and also tried a counterstain of picro-erythrosin. After examining several hundreds of films stained by each method, I came to the conclusion that the modified cresoidin method showed up the diphtheria bacilli more clearly, and definitely than the others. When in doubt, I stained films with Gram's stain, Pugh's or Cobbett's stain.

The blood serum media were made according to Loeffler's formula, three parts of ox serum mixed with one part of 1 per cent glucose bouillon; about six cubic centimetres of this mixture was then poured into sterilised test tubes; these were sloped, and inspissated at 65° C. After the serum had coagulated, the tubes were sterilised by steam at 85° C. on three successive days for an hour each time.

In speaking about Neisser's stain, Graham-Smith states that nearly all organisms considered on morphological grounds alone to be diphtheria bacilli stained well with Neisser's stain, or Cobbett's modification of it. He found polar bodies in nearly all diphtheria bacilli, whereas Hopmann's bacillus did not show them. Many other observers find Neisser's stain to be one of the most important means of distinguishing between the diphtheria bacillus and the/

the pseudo-bacillus.

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I agree with Simon, who finds that Neisser's modified cresoidin method brings out in a perfect manner, not only the polar bodies, but the morphology of the bacilli as well. He states that the pseudo-organisms are rarely met with, and show no granules with this stain.

In considering the appearance of the bacilli to be looked for, the classification of Wesbrook, Wilson and McDaniel was closely studied. They divide their types into three main groups:-

(1) Granular, (2) barred rods, (3) solid or even stained rods. These again are subdivided into types according to their size or shape. The types that have been met with in clinical cases come into the following group:-

Group A. Involution forms. A granular, A₁ barred, A₂ solid staining.

Group C. The long diphtheria bacillus. C granular, C₁ barred, C₂ solid staining.

Group D. The short diphtheria bacillus. D granular, D₁ barred, D₂ solid staining,
The D₂ type is practically indistinguishable from Hofmann's bacillus.

The bacillus I have always encountered has been of
the/

the C, C₁ or D types. The solid staining type I have frequently seen, but always associated with some polar stained rods.

Hofmann's bacillus did not give rise to much trouble in diagnosis. The short straight, oval bacillus with rounded ends, and one median lightly stained transverse septum, absence of granules, and so often in parallel arrangement is fairly easily distinguishable. The colonies on serum media after 18 hours growth are much larger, and whiter, than these of the diphtheria bacillus. In several faucial cultures from clinical cases I came across a *Streptothrix* with granules, which owing to the fragmentation of the chains in a certain way, gave rise to a morphological appearance rather suggestive of diphtheria rods.

I find it much more difficult to recognise the diphtheria bacillus in nasal cultures. *B. coryzae segmentosus* is apt to lead to mistakes. I have frequently come across cocci which take on a dark staining of the same nature as the polar staining in diphtheria, and one has to look very carefully to satisfy oneself of the presence or absence of diphtheria bacilli.

In all cases in which I was doubtful from the morphology, if the bacillus in question was a true diphtheria/

diphtheria bacillus, I endeavoured to obtain a pure culture of the organism. The morphological appearance in pure culture having been studied, I then inoculated two test tubes containing the serum water of Hiss, tinted with litmus, to which had been added 1 per cent glucose, and 1 per cent saccharose respectively.

These were then incubated at 37°C. for several days, the production of acid being noted from day to day. If the litmus in the glucose tube had markedly changed colour after 48 to 72 hours incubation, and that in the saccharose remained unaffected, the bacillus was accepted as a true diphtheria bacillus. In some cases the saccharose tube also turned very slightly pink, but this was probably due to a contamination.

In a very recent paper by Eagleton and Baxter¹⁰⁰ the conclusions come to with reference to the sugar tests are as follows:-

"If glucose is not fermented, the culture is not virulent *B. diphtheriae*.

If glucose is fermented but not saccharose, the organism may or may not be virulent.

If both glucose and saccharose are fermented the organism may be *B. xerosis* or may be contaminated."

With reference to the very slight production of acid in the saccharose tubes which I noted in several/

several cases, it is interesting to note that these observers state it was due in their experiments to a very slight degree of contamination.

As we are not permitted to carry out animal experiments in the hospital, I was not able to test the virulence of the bacilli isolated, which is the really clinching test. In one case however, a nasal carrier of many weeks duration, the virulence was demonstrated in a very practical manner by the infection of his sister, as I have already described. Virulence was tested on a strain of morphologically typical diphtheria bacilli from a contact faucial carrier obtained in a Scarlatina ward, and the bacillus in question killed the inoculated guinea-pig in 72 hours with all the typical lesions, thus showing full virulence.

How many consecutive negative cultures should be obtained before a patient is discharged?

Graham-Smith makes three consecutive negative results the rule in his investigations.

Ker thinks that isolation should be maintained until at least two consecutive negative cultures have been obtained from the throat.

Scholes places no reliance on one negative test. He says that even with two or three negative cultures errors/

errors will be made, but the probability is very much lessened.

I am perfectly convinced that for accurate investigation, and to be really satisfied that the patient is no longer a carrier, three consecutive negative cultures are essential, and even more should be obtained, if time and circumstances permit. On several occasions I have found a positive result following two negative cultures in the same individual. In preference to three consecutive negative cultures taken on successive days, I would suggest three taken with a day intervening between each. Needless to say, no antiseptic application must be made to the throat or nose for several hours previous to the swabbing. It is very difficult to attain the three negative standard in hospital practice, particularly when cases are numerous, and space limited. Under such circumstances, we are content to accept two consecutive negatives as a standard of freedom from infection. In those cases however, where the patient is going back to a hospital or institution, where risk of infection is likely to be unduly great, I would always insist on three negative cultures on alternate days previous to discharge.

In my series of cases I always endeavoured to obtain/

obtain three consecutive negative cultures as a criterion of freedom from bacilli, and even more if possible. The following table gives the number of negative cultures obtained for different cases previous to discharge.

| | |
|---|------------------|
| Two consecutive negative cultures on successive days | 13 cases |
| " " " with day intervening | 8 " |
| Three " " on successive days | 32 " |
| " " " with day intervening between each | 100 " |
| Four " " not on successive days | 27 " |
| Five " " " " " | 24 " |
| Six " " " " " | 3 " |
| Seven " " " " " | 1 " |
| | <hr/> 208 cases. |

We thus see that in 89.90 per cent of the cases, three or more consecutive negative cultures were obtained as a criterion of freedom from diphtheria bacilli.

Of the 21 cases discharged on two consecutive negatives, five were found to harbour diphtheria bacilli when the third culture was examined immediately after the discharge of the patient, so these five were still carrying on discharge. As far as I could ascertain four/

four of them did not give rise to any cases of clinical diphtheria, but the fifth, a nasal carrier, was the almost certain source of infection of his sister who was admitted shortly after with a typical clinical diphtheria.

From the point of view of infectivity, it is the nasal carrier who is the real danger. The faucial carrier I think is very much less to be feared from the point of view of infectivity. I would be very unwilling to allow a nasal carrier, particularly a child, out of hospital with diphtheria bacilli still present. Any slight catarrh causes nasal discharge, and this discharge laden with diphtheria bacilli is a very potent source of infection.

CONCLUSIONS.

Neisser's modified cresoidin method is one of the most satisfactory means of staining for the morphological diagnosis of the diphtheria bacillus.

The polar staining or beaded rod is the type of bacillus practically always found after eighteen hours growth on blood serum at 37°C.

The diphtheria bacillus does not tend to lose its virulence after a long residence in the throat
or/

or nose of convalescent carriers.

The treatment of carriers with Potassium iodide and chlorine water is useless, as is the case also with the local application of antiseptics.

The staphylococcal spray is well worth a trial in faucial carriers, and may probably prove successful in a certain percentage of cases but certainly not in all.

Hewlett's endotoxin is worth a trial in persistent carriers, but too much must not be expected of it.

Detoxicated Klebs-Loeffler vaccine administered to cases on admission in the manner I have described (1) does not shorten the life of the bacillus in the throat or nose of the convalescent:

(2) does not prevent the development of the chronic carrier:

(3) does not have any appreciable influence on the development of paralysis:

(4) has apparently had a favourable effect on the mortality percentage.

I do not think there is any useful purpose to be served in routine administration of detoxicated diphtheria vaccine to all clinical cases on admission, but I think its administration in the severe faucial and nasal cases might lead to interesting results.

In/

In chronic carriers when for any reason surgical measures are not possible, detoxicated vaccine in large doses may lead to a successful issue. In such cases the three measures I would advocate are the staphylococcal spray, Hewlett's endotoxin, or detoxicated vaccine.

To really attack a chronic faucial carrier with definite hope of success, the method I would suggest is tonsillectomy.

In the case of nasal carriers, the greatest hope of success lies in adenoidectomy and tonsillectomy. Examine carefully for, and treat any unhealthy condition of the nasal cavities or accessory sinuses, and always look for, and remove foreign bodies.

Nasal carriers are a very real danger to the community, and should not be allowed out of quarantine until the bacilli have disappeared or have been proved non-virulent by guinea pig tests.

Faucial carriers are much less dangerous, and in special circumstances should be allowed out of quarantine, even though bacilli are still present. I am inclined to hold the view that cases of faucial diphtheria should be discharged at the termination of convalescence without regard to the presence of bacilli or otherwise.

Three/

Three consecutive negative results should be the absolute minimum, before one can say with any confidence that a convalescent carrier is free of bacilli.

I beg to acknowledge my indebtedness, and convey my thanks, to Dr Ker for many helpful ideas, and numerous references to the literature. I also wish to express my appreciation of the ever willing assistance rendered me by the sisters and nursing staffs of the wards in which I worked.

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