LABORATORY NOTES ON PNEUMONIA IN KENYA

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This paper deals with laboratory work on pneumonia carried out from the beginning of 1930 until the writer's enforced retirement from work in October 1935, and is divisible under the following headings:

A. The type constitution of pneumonia in the Highlands of Kenya.
B. Type constitution of the Kenya *Pneumococcus* group IV.
C. Experiments with prophylactic *Pneumococcus* group vaccine.
D. Technique of surface culture suspensions for serological and bile-solubility tests, and for preparing specific agglutinating sera.

A. Type constitution of pneumonia in the Highlands of Kenya

The work of typing was carried out continuously through each year, and was confined to cases of lobar pneumonia, together with a few cases of pneumococcal meningitis as they arose, the bulk of these cases being among natives admitted to the Native Hospital, Nairobi, from January 1930 to October 1935. As natives of all tribes were represented in the series of cases, and a very considerable amount of travelling is done by the natives, the figures obtained for *Pneumococcus* type incidence may probably be applicable to the whole of the populated areas of the Kenya Highlands, if not to the coastal area as well. The main feature which appeared from the work was a very striking preponderance of the "group IV" pneumococci over the standard American types I, II, and III as causatives of pneumonia among the natives. This preponderance of group IV pneumococci appeared to be the rule also in lobar pneumonia amongst Europeans, as far as could be judged on the small series of cases from which material for typing could be obtained. Below are the tabulated results of the typing work for the five years; the table is constructed from the Annual Reports of the Medical Research Laboratories. The total number of cases typed was 776; this figure includes a small proportion of cases of pneumococcal meningitis, in which type I greatly preponderated.

	Type			
	I	II	III	Group IV
Incidence % for 1930 to 1935	22·0	5·6	8·3	64·1

These figures show a fairly close correspondence for each year between the relative incidences of the types I, II, and III and the group IV. The highest figure obtained for type I was 28·4%, and the lowest for group IV was 54·2% in 1934.

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B. Type constitution of the Kenya *Pneumococcus* group IV

Serological analysis of the Kenya group IV was begun in 1931, and twenty-one distinct types of the group IV were separated out up to the date of final discontinuation of the work in 1935. While the bulk of the very severe and fatal cases were due to type I, many were attributable to group IV. The Kenya types of group IV designated by me as Kw. L, Kw. J, Kw. D, and Kw. V were found to be especially significant, and roughly in the order of clinical importance here given; and the types Kw. M and Kw. O also were important at times both from the point of view of virulence and frequency of occurrence. The Kenya type L was found to predominate not only in the more severe and fatal pneumonias that were due to group IV cocci, but to be responsible also for nearly all the cases of pneumococcal meningitis that were not caused by type I. In 1933, for example, out of 107 cases which were fatal or classed as severe, and which were caused by pneumococcal types up to then serologically defined, type I was responsible for 27, type II for 12, type III for 13, and the type Kw. L for 14; in the remaining 41 severe and fatal cases, for which group IV types were responsible, the type Kw. J was the most prominent.

Geographical distribution of Kenya group IV types. By the kind collaboration of Dr Griffith of the Ministry of Health Laboratories, of Sir Spencer Lister of the South African Institute for Medical Research, and of Miss Georgia Cooper of the Bureau of Laboratories, Health Department, New York City, my Kenya types D, J, and L were identified, by means of agglutination tests of formol saline suspensions of surface cultures, with the New York types VIII, VII, and V respectively (1931); while my types L and J were identified with Lister's important type A, and his type K, respectively, of his South African series. Several other of the Kenya types were identified by my successor Dr R. M. Dowdeswell in 1935 with American types by means of agglutination tests with formol saline suspensions kindly sent me in large numbers, together with their corresponding agglutinating sera, by Miss Georgia Cooper. Several formol saline suspensions of surface cultures derived from pneumonia cases were also sent me by the Chief Medical Officer of the Kilo Moto mines of the Belgian Congo, and one of these was identified with the Kenya type Kw. O. The very wide geographical distribution of these few penetrative and dangerous strains from America to Johannesburg and Nairobi in Africa indicates the great likelihood of their being of etiological importance in Great Britain and elsewhere in Europe; and it is likely enough that all of the comparatively few group IV serological species separated in Nairobi are identifiable with some of the very many American types which have been distinguished. It is noteworthy that each of the twenty-one Kenya types were primarily isolated from severe cases of pneumonia, and sometimes from post-mortem specimens of hepatized lung (kindly collected by Dr F. W. Vint, Morbid Anatomist to the Department), which lends support to the deeper respect that is nowadays

felt for the "group IV" pneumococci. In the series of 181 native cases of lobar pneumonia typed in 1933, the total case mortality was 21·4%, and the case mortality for group IV infections only was 16·4%. These figures, together with the fact that very severe pneumonia attacks due to group IV cocci were observed among Europeans, do not suggest that Kenya Africans are specially susceptible.

C. EXPERIMENTS WITH PROPHYLACTIC *PNEUMOCOCCUS* GROUP VACCINE

In 1934 it was decided to try to imitate Sir Spencer Lister's (1916) well known and very successful application of group *Pneumococcus* vaccine for prophylaxis of pneumonia in the South African gold mines. A similar field for such experiment was offered by the recently established gold-fields in the Kakamega district of Kenya, where the native labour was quite heavily afflicted by pneumonia. A case mortality of 30% was reported by Dr Charles Searle, medical officer to one of the gold-mining companies.

Preparation of group-valent prophylactic Pneumococcus *vaccine.* For this, the method of broth culture was employed, the medium being a trypsinated bullocks' heart broth, containing 0·3% added glucose, standardized to pH = 7·6, the salt content being sodium phosphate instead of sodium chloride, for the sake of the "buffer" effect. This broth was cheaply made from condemned bullock's hearts supplied *gratis* by the municipal abattoir, the trypsin-amylopsin liquor needed being also made in the laboratory from pig's pancreas obtained *gratis* from the same source. This broth yielded growths of *Pneumococcus* of between 2000 and 3000 millions per c.c., and cultures of not more than 18 hr. incubation were used for making the vaccine by the process of concentration to 8000 millions per c.c. described below. It would have been preferred to use suspensions of surface cultures on blood agar for vaccine, since pneumococci in broth culture rapidly became devitalized so that in an 18 hr. culture only a small proportion of the cocci are capable of retaining Gram's stain, and agglutination tests with type sera of the resuspended deposit are wanting in specific sharpness; whereas with surface cultures of the same age, all the pneumococci are strongly Gram-positive, of exuberant appearance, and invariably give beautifully definite agglutination with homologous type serum—even in the strongest concentrations of the serum, provided this has been prepared by the use of young and therefore strongly specific surface culture suspension as antigen, instead of broth culture deposit which is always, by reason of partial autolysis, more or less defective in specific qualities.

Lister's prophylactic vaccine originally contained eight *Pneumococcus* types—five group IV strains (including notably Lister's type A which, as above mentioned, is the Kenya type L and the American type V) in addition to those serological species separated by him which were identified with the classical American types I, II, and III. In later years, certain other kinds of bacteria, such as staphylococci, were found to have become of etiological importance in pneumonia, and were therefore included in his vaccine, while

certain of the *Pneumococcus* strains originally included appeared to have been immunized out of the native labour population of the Rand mines. In Kenya it happened similarly that five of the group IV types were markedly more prominent than the others, these being Kw. D, Kw. J, Kw. L, Kw. M, and Kw. O, in 1934 when the prophylactic vaccine experiment was instituted. But owing to loss of the stock cultures of Kw. M and Kw. O and a temporary dearth in pneumonia cases which prevented their recovery again for some little time, the first experimental batches of vaccine issued in September 1934 to the Kakamega gold-fields contained only the most prominent group IV types D, J, and L, in addition to types I, II, and III. Later in the year the type Kw. V was isolated, found to be important, and included in the vaccine together with the type M.

Cultural and concentration procedures, and dosage. The units of culture were 2 l. flasks of the broth described; these were inoculated each with a few cubic centimetres of very dense saline suspension of surface culture made by washing off the whole of the growth of a confluent culture made on blood agar in a tube of 1 in. diameter incubated for 18 hr. Such heavy inoculations were found necessary to get the broth cultures to "take" with reasonable certainty. The flasks were inoculated in groups of eight, each with a pure culture of one of the *Pneumococcus* types. The broth cultures after not more than 18 hr. incubation at 37° C. were killed and fixed by the addition of 0·3% of formalin, after Gram-stained smears of the centrifugalized deposit of a small volume of each culture had been examined to verify purity of growth.

Concentration of the vaccine cultures to the required strength of 8000 millions of cocci per c.c., as recommended by Lister, was effected by a process of filtration, the laboratory assistant Mr E. C. Young producing an ingenious device for the purpose, consisting of an arrangement of sterile brushes in the filter container to remove from time to time the deposit adhering to the surface of the filter candle. A fine-grade candle was found to be preferable to a coarse one, the pores of which became blocked by the cocci. The culture deposits so obtained were then suitably resuspended in 0·5% phenol saline to the required density, and these carbol saline suspensions of 8000 millions per c.c. of each of the *Pneumococcus* types were then mixed together in an 8 l. tubulated bottle, from which rubber-capped vaccine bottles were filled.

The dosage of this vaccine recommended by Lister was three injections of 1·0 c.c. at weekly intervals. Experimental trials of this dosage on the writer, other Europeans, and also natives, revealed the remarkably low toxicity of vaccine of such heavy strength; local symptoms following injection were slight, and subsequent general malaise was generally negligible and sometimes absent. It is noteworthy that this dose of 8000 millions is recommended in the Handbook of the South African Institute for Medical Research for *treatment* of pneumonia patients. Experiments in treatment of native pneumonia patients in Nairobi made with the same dosage of the Kenya vaccine showed slight or indefinite toxic reactions; but, by reason of the fact that native pneumonia

cases generally come for admission to hospital in a too advanced stage of their illness for effective vaccine treatment, the therapeutic results were inconclusive.

In September 1934, and the subsequent months, many thousands of doses of this vaccine were issued to the Kakamega gold-fields, and a considerable quantity to certain prisons, in which pneumonia figured prominently in the sickness rate. Very favourable reports, sometimes worded in glowing terms, were received in each case from the medical officers who undertook inoculations with the vaccine, with regard to its effects in reducing the incidence and severity of pneumonia among native labour. I have to thank Dr Charles Searle, previously mentioned, for constructing an elaborate chart, which it is unnecessary to reproduce, exhibiting the effects of anti-pneumonia inoculation. The reports in general do not, unfortunately, lend themselves to the construction of a numerical table of results; but they were to the effect that a very marked decrease in incidence of pneumonia occurred during the periods immediately following extensive inoculations, and that pneumonia attacks which occurred among vaccinated subjects were generally of a mild nature. Independent statements to such effect from capable medical officers cannot be sceptically ignored, although due allowance must be made for the various sources of error in estimating results of such human experiments with prophylactic vaccine. The pneumococcal types which were included in the vaccine were found to be responsible for 67% of the total number of pneumonia cases, and for 74% of the cases classed as severe or which were fatal, admitted to the Native Hospital of Nairobi in 1934. For convenience in the sometimes difficult matter of re-inoculating natives, the scheme of dosage was subsequently altered from three injections of 1·0 c.c. to two injections of 1·5 c.c. at weekly intervals.

As it was impracticable to produce sufficient quantities of vaccine of the required strength by means of the serologically more desirable method of using suspension of surface culture, the broth culture method was continued with, and once a sufficient stock of vaccine had been obtained by the more rapid procedure of concentration described above, it was found practicable to maintain the stock by means of the slower process of allowing the formalinized cultures to sediment naturally, the supernatant broth being then decanted and the sediment resuspended in the required smaller volume of carbol saline. Production of the vaccine in this way has been very efficiently carried out by the laboratory assistant Mr W. A. Doust since the writer's retirement in October 1935, to meet the steady demand for this prophylactic in East Africa.

It seems that, for prophylaxis of pneumonia by means of a group-valent vaccine to be continuously successful, it is essential that the work of typing pneumonia cases should be carried out continuously to keep a check upon the relative and varying prominence of the different types of pneumococci and also of other bacteria, such as bile-insoluble streptococci, as causatives of pneumonia. A considerable proportion of native pneumonia cases yielded only bile-insoluble streptococci, some of which were found to be fermenters

of inulin; while from time to time the method of mouse culture of sputum adopted yielded only "Friedländer's bacillus", *B. mucosus capsulatus*, which was always of a non-dulcite fermenting strain, and sometimes caused pneumonia of a highly fulminating type, fatal in about 48 hr., and resembling—and occasionally mistaken for—pneumonic plague in the thin, haemorrhagic sputum produced by the patient. It has been shown by Lister & Ordman (1935) that as a result of prophylactic inoculations certain strains of pneumococci, notably the South African type A above mentioned, were immunized out of the mines labour population, the place of these types being taken by other strains or by bacteria other than pneumococci. In Nairobi the bacteriological laboratory assistant, Mr M. de Souza, has made a gallant attempt, under the pressure of much other work, to continue this routine of typing pneumonia cases, which routine the writer, while he was in charge of the bacteriological laboratories, found it possible to carry out single handed. But under such discouraging circumstances it is unlikely that prophylactic immunization against pneumonia will receive in Kenya that fair and extended trial which the experiences of the originator of *Pneumococcus* group vaccine, F. S. Lister, in South Africa have shown that the method so amply merits.

Therapeutic use of Pneumococcus *vaccine.* The use of Lister's group-valent prophylactic *Pneumococcus* vaccine for treatment of pneumonia patients in doses of 8000 millions for each injection is recommended in the *Handbook* of the South African Institute for Medical Research, and there can be no doubt that the method may be strikingly successful in favourable cases where the first injection can be given within the first 48 hr. of the pneumonia attack. Reliance is here placed on the speedy non-specific action of the heavy dose of vaccine; specific effects, which may be extremely valuable, may be expected to follow soon afterwards, except, of course, in cases where the infecting type of *Pneumococcus* is not present in the vaccine. With regard to chemotherapy, it seems reasonable to suggest that vaccine therapy in pneumonia, the prime value of which has so often been demonstrated, should not be abandoned in favour of chemotherapy, but that the two methods should be complementary. The experiments mentioned in section C in treating African pneumonia patients with the full 8000 million dose of the Nairobi prophylactic group vaccine revealed the extremely low toxicity of pure *Pneumococcus* vaccine. The very low toxicity of pure *Pneumococcus* suspension was demonstrated also by the writer's procedure for preparing agglutinating *Pneumococcus* serums. By this method, which is described in more detail below, rabbits were injected intravenously with not less than 5·0 mg., sometimes 6 or 7 mg., of killed *Pneumococcus* type suspension made by washing off 18 hr. surface cultures on blood agar; these injections were repeated on three consecutive days, before and after an interval of 5 or 6 days, and in no case was any sign of illness noticeable in the animals.

Autogenous vaccine treatment of pneumonia. The advantages of autogenous vaccine for treatment of pneumonia seem clear; the obstacle to its use is the

length of time that must elapse before an autogenous vaccine can be got ready for injection by ordinary methods of culture, when the time factor is of urgent importance. The writer accordingly experimented with the use as vaccine of the heavy suspension of pneumococci obtained by washing into carbol-saline the peritoneal cavities of mice which had been inoculated intraperitoneally with sputum from the patient to be treated; the advantage here being that the vaccine can easily be got ready for use a few hours after the first satisfactory specimen of sputum is obtained from the patient. A full description of this method and of therapeutic trials made of it has been given elsewhere (de Smidt, 1938).

D. TECHNIQUE OF SURFACE CULTURE SUSPENSIONS FOR SEROLOGICAL AND BILE-SOLUBILITY TESTS, AND FOR PREPARING SPECIFIC AGGLUTINATING SERA

The procedure used in typing consisted in mouse culture made by inoculating mice intraperitoneally with sputum, followed the next morning by culturing the heart's blood of the mice on to blood agar. Criticism has been levelled at the use of sputum in this way, on the grounds that a mouse might become infected by adventitious pneumococci which have come to adhere to the sputum during its passage of the upper respiratory regions. For this reason the elaboration of washing each specimen of sputum before emulsifying it for inoculation has been generally advised: a procedure which appears to the writer to be irrational, and has therefore never been used by him. The chance is sufficiently remote that a mouse would become infected by a few casual pneumococci picked up by the sputum from the upper respiratory passages, instead of by the vast numbers of virulent pneumococci brought up by the sputum from the alveoli and regularly diffused through its substance.

The primary cultures from mouse's heart were subcultured on to blood agar, heavily enough to produce confluent growths. After not more than 18 hr. incubation, the subcultures were washed off in about 5·0 c.c. of normal saline, or more, and the moderately dense suspensions so obtained used as described below for bile solubility and agglutination tests. The saline is only allowed to be in contact with the blood agar for the minimum time needed to remove the culture with the help of a platinum loop, and thus clean suspensions without noticeable blood staining are obtained. Suspensions of opacity about equal to those of the familiar Oxford Standards agglutinating suspensions of *B. typhosus*, etc., were found to give quite satisfactory results, and perfectly definite bile and agglutination tests were found to be obtainable with suspensions so light as to be only faintly opaque. After the bile solubility test had been done by the method described below, the suspension was formalinized by adding with a capillary pipette and teat one drop of a 10% solution of formalin in normal saline for roughly each cubic centimetre of suspension. Suspensions of pneumococci so killed and fixed preserve their specific agglutinating properties for an indefinite period at room temperature.

Bile solubility tests. The materials used consisted of Dreyer's agglutination tubes, of the old obtuse bottomed type; 1% solution of sodium desoxycholate in normal saline; a short capillary pipette with teat; normal saline in a beaker; an agglutination test rack, and water-bath at 37° C. Using two agglutination tubes, two drops of the bile salt are placed in tube 1, the pipette washed out, and two drops of saline placed in tube 2 for control. Four drops of suspension are next added to each of the tubes, their contents mixed by flicking the butt ends with the finger, and the result observed after incubating the tubes for 2 or 3 min. in the water-bath, by holding them up side by side against a window. Although the incubation is unnecessary for solution, it hastens the process and standardizes it for comparative purposes. Given a pure suspension of *Pneumococcus*, clarification is complete or so nearly so that when a control is employed as described the trace of residual haziness does not obscure the result even with a faintly opaque suspension. By the drop method, the actual concentration of bile salt obtained is less than 1/300, the drop of bile salt solution being smaller than the drop of saline from the same pipette. The two in six drops concentration is dependable for all purposes including centrifugalized top fluid of mouse peritoneal wash, and for cerebrospinal fluid cloudy with cocci; but good results are often obtainable with a one drop in five concentration of the bile salt in testing suspension from blood agar or serum culture.

Agglutination tests, and the use of formalinized suspensions. The materials used for agglutination tests were: Dreyer's obtuse-bottomed agglutination tubes; agglutination test rack, the holes of which were numbered or lettered in grease pencil for the various type sera to be used; water-bath at 37° C.; capillary pipettes and teat, the pipettes being cut short, normal saline, and the pneumococcal suspensions formalinized as above described. One drop of each type serum, or suitable dilution thereof, is placed in an agglutination tube, using a fresh pipette for each, and four drops of suspension are added, the contents of the tubes being mixed by flicking their bottoms with the finger. The control consists of one drop of saline and four drops of suspension, but it cannot be regarded as indispensable; auto-agglutination of surface culture suspension is such a rare occurrence that the risk of it may be ignored, and in any case the negative tests in each series form controls upon the positive test that may occur. Controls of normal rabbit serum are totally unnecessary. Naked eye agglutination is complete within 2 or 3 hr. at 37° C., where the serum is of not less than 1/10 titre; but a very weak serum has been found to need 24 hr. to produce a standard degree of agglutination. It appears to be immaterial whether a temperature of 37 or 56° C. is used for incubation. If the rack of tubes is left overnight in a room temperature of about 60° C. the results will be the same as if incubation had been performed in the usual way. Readings being taken in the usual manner against a shaded lamp, standard agglutination is regarded as complete clearing of the fluid surrounding the clumps, these consisting of dense, clear cut, refractile granules which sink

to the bottom of the tube and settle on its sides. With heavier suspensions, irregular but similarly clear cut and opaque larger masses, usually flattened, can be stirred up and broken into granules by flicking the bottom of the tube. Such beautifully clear and definite agglutination tests were invariably obtained with formalinized suspensions of pure cultures of pneumococcal types. Indefinite results produced by non-specific protein-agglutination were never seen, except when for experimental reasons the culture for agglutination had been greatly over-incubated so that autolysis of the cocci was advanced. Striking contrasts were obtained by comparing agglutination tests carried out as described with formalinized suspensions of young blood-agar cultures, and tests performed with deposits of young broth cultures resuspended in saline; while the agglutination with the former was always beautifully clear and sharply defined, that obtained with the broth culture deposits was always muddy and ill-defined, and some degree of non-specific agglutination always appeared with other sera than the homologous one. The reason for this at once becomes clear when Gram-stained smears are examined microscopically from blood-agar cultures of 18 hr. incubation, and centrifugalized broth culture of the same age; while in the surface culture smear all of the pneumococci are intensely stained and of exuberant appearance, in the broth culture smear the vast majority of the cocci have lost their power to retain Gram stain and are of a mushy appearance due to partial autolysis, and these characters are doubtless associated with a very considerable loss of type specificity. The great superiority of surface culture suspension over broth culture deposit for preparing specific pneumococcal agglutination sera, and probably also therapeutic sera, is therefore evident.

Practical advantages of formalinized suspensions. In the work of analysing serologically the Kenya *Pneumococcus* group IV, the group IV strains which did not agglutinate to any serum prepared up to date were stored in the form of formalinized suspensions in capped tubes, to be tested against sera subsequently to be made. It was thus easy to keep a record of the relative incidence of pneumococcal types during any previous period, without the trouble and difficulty of maintaining a collection of living cultures. Such suspensions of known types were preserved also for verifying and checking the potency of agglutinating sera when occasion demanded.

Formalinized suspensions were further found to be of value in the scheme of correlating Kenya types of *Pneumococcus* with those isolated in America and South Africa, as described in section B. The exchange of formol saline suspensions of *Pneumococcus* surface cultures by mail over long distances is a simpler and more certain matter than the similar exchange of cultures preserved in the living state by one of the accepted methods. The amount of formalin added by the writer to his suspensions to fix and preserve them, by the rough drop method described, was from 0·3 to 0·4%.

Preparation of agglutinating sera by means of surface culture suspensions. The advantage of the use of surface culture suspension for the preparation of

agglutinating sera, and no doubt also for preparing sera for therapeutic use, lies in the readily demonstrable fact that young surface culture of *Pneumococcus* is very rich in specific antigen, consisting as it does practically entirely of healthy cocci the type specificity of which is unimpaired; whereas by the time that a broth culture has reached its maximum growth, the great bulk of the pneumococci are found to be degenerate to the extent of having lost their power to retain Gram stain and to react in a definite specific manner to agglutinating type sera. Agglutinating sera, made by the easy and very reliable method of inoculating rabbits with heavy doses of killed surface culture suspension as described below, were found invariably to be completely type specific in low dilutions, and invariably to give beautifully definite agglutination tests with formalinized surface culture suspensions of pneumococci, or with peritoneal washings of sputum-inoculated mice.

To ensure type purity, culture derived from a single, well-isolated colony was always used. Four Roux flasks of fresh blood agar (made from whole sheep's blood drawn off from a sheep on the laboratory premises) were inoculated by pouring into each some 15 c.c. of saline suspension of a small blood-agar culture made from a single *Pneumococcus* colony, running the suspension over the whole surface of the agar in the Roux flask and then decanting the excess fluid. The flasks were then incubated for 17 hr. at 37° C., agar downwards. After examining Gram-stained smears from each flask, the cultures were washed off into 14 c.c. of 0·5% carbol saline, the whole of the growth in each flask being rubbed off by means of a long sealed capillary pipette bent into a right angle. The crude suspension was then poured into a tube through a filter funnel loosely plugged with cotton wool, to remove flakes of agar, and its strength estimated by opacity scale. After heating in a water-bath at 60° C. for ½ hr., the suspension was introduced for convenience of use into a small rubber-capped vaccine bottle. The strength of the suspensions so obtained was generally about 5·0 mg.

A rabbit received intravenous injections of 5–7 mg. of pneumococci, in 1·0–1·4 c.c., on three consecutive days, the injections being repeated for a further 3 days after a 5-day interval. After a rest of 5 clear days, a test sample of blood was taken from the rabbit in the morning, and if the agglutinating titre was satisfactory the main bleeding was done the same day into agar-lined tubes. Allowing for the variation in response of individual rabbits, sera of the sufficiently high titres of 1/25–1/150 were expected from six injections as described, the usual titre being between 1/50 and 1/100.

Summary and conclusions

A laboratory investigation of 776 cases of lobar pneumonia, mainly among Kenya Africans, over a period of 5 years, revealed a remarkable preponderance of infections due to group IV pneumococci.

Comparison of the total case mortality rate with the case mortality of

group IV pneumonia infections suggests that Kenya Africans are not specially susceptible to pneumonia, and that many of the group IV pneumococci which cause pneumonia in Kenya are of high virulence and penetrative power.

A considerable number of group IV pneumococcal types capable of causing severe lobar pneumonia were serologically isolated. A few of these were identified with pneumococcal types prevalent in New York and South Africa.

An experiment was made on a fairly large scale with prophylactic anti-pneumonia vaccine, made in the same way as Sir Spencer Lister's pneumonia prophylactic which is used with success in the Rand mines, but constituted of the pneumococcal types which had been found to be most prevalent in pneumonia in the Nairobi district. Extensive trials with this vaccine have been made on the labourers of the Kenya gold fields, and elsewhere, and very encouraging reports on the effects of this vaccine in reducing the incidence and case mortality of pneumonia have been received from medical officers who carried out the inoculations.

A technique is described for the use of surface culture suspensions of pneumococci for serological and bile-solubility tests, and advantages are claimed on specified grounds for the use of surface culture suspensions in preparing agglutinating and therapeutic pneumococcal sera.

A method is described for the use of formol-saline suspensions of surface cultures for preserving collections of pneumococcal types, and for correlating pneumococcal types prevalent in different countries.

In conclusion, I wish to express my gratitude to Mr W. A. Doust, Laboratory Assistant in charge of the Media and Vaccine Production Section of the Kenya Medical Research Laboratories, for the invariable excellence of his assistance in preparing materials of the finest grade for my work; also to Mr J. St A. M. de Souza, who was for many years my personal assistant in the bacteriological laboratories, for the highly skilful and trustworthy work for which I could always rely upon him.

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