

M I C R O - O R G A N I S M S

In The Air

Of

RAILWAY CARRIAGES

By

ROBERT NICOL^h BREBNER.

M. B., Ch. B. (Edin.)



Micro-organisms in the Air of Railway Carriages.

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Frequent investigation has demonstrated the presence of micro-organisms in the air everywhere and under all conditions except over the open sea and in the Polar regions (1) while they are most commonly found in the larger cities, especially under poor hygienic conditions (2).

Their occurrence in air cannot be regarded as otherwise than temporary (3-4) because of the greater specific gravity of the organisms or of the dust particles to which they may be attached.

They do not multiply in the air as they do in the soil from which the majority of those found in air are derived. They increase in number in a dry season, lessen in number when the surface soil is moist and immediately after rain are also fewer in number. They do not pass to the air independently but are transferred to it by dust (5).

The air does not supply them with the moisture and organic nutriment necessary to their development; at the same time some species may retain vitality for

for days or even weeks, as proved by experiment on the communication of infectious diseases through the air. (6)

It is estimated that there are from one to two hundred bacteria in a thousand litres of air almost all harmless. The bactericidal action of sunlight, desiccation and the diluting action of the atmosphere on noxious substances lessen the risk of direct aerial infection. Direct sunlight is fatal in at most a few hours to Tubercle, to Typhoid and Diphtheria in from half-an-hour to two hours, to spores of Anthrax in three and a half hours, the blue rays having most germicidal effect. (5)

In the open air in London one organism was found to every 38,300,000 dust particles present in the air, in room air one in 184,000,000. In a London suburb there were 20,000 dust particles in one cubic centimetre of air, and in a yard in the centre of London there were 500,000 particles in a cubic centimetre of air. (5)

In air almost entirely free from dust--the cellar of Montsouris Observatory--only one out of ten tubes showed the presence of bacteria.

From other observations made at the Montsouris

Montsouris Observatory during several years the average number of micro-organisms per cubic metre were (7):-

For City air (Paris)	5,400	Bacteria	1700	Moulds
Country air	290	-	190	-

As might be expected, the summer months give a much higher number than the winter months.

Paris air	July	21,000	Bacteria
	December	350	-
Country air	July	1000	-
	December	72	-

If the presence of micro-organisms in air has no greater significance it may be said to indicate facility for transportation of which advantage may be as well taken by pathogenic forms as by the more common so-called harmless ones.

Just what relations these so-called harmless forms bear to the diseases of men and animals it is impossible, with the knowledge at present available, to say, but doubtless the inhalation of these with dust may set up irritation in the respiratory passages and so predispose to actual disease. That dust has effect in producing pathological conditions of the lungs has been proved by Canfield (8) and the well-known conditions of Anthracosis and Silicosis show that dusty occupations predispose to tuberculosis (9).

tuberculosis (9).

But the nasal mucous membrane has been found to destroy inspired micro-organisms (10)-(11). Because of the great prevalence, however, of mouth-breathing this natural means of preventing infection is, to a very great extent nullified, and prolonged breathing, even through the nose, of a dust-laden atmosphere, would in time reduce the sensibility and activity of the mucous membrane and infection might follow.

That pathogenic bacteria do exist in the air is undeniable. Their detection in it is almost hopeless and their elimination from it an impossibility, but precaution may be taken to prevent their access to it by the destruction or disinfection of all infectious material.

There can be no doubt whatever as to the presence of tubercle bacilli expectorated in the sputum of phthisical patients, the sputum becoming dried and pulverised and the bacilli set free. This may take place in the streets and in the houses of the lower classes and the resulting dust inhaled, so affording a means of spreading the disease.

In Brompton Hospital Williams found the bacilli

bacilli in the air in the ventilating shaft and Klein found that guinea-pigs kept in the shaft became tubercular.

Clean streets would promote cleanliness of boots, of conveyances and of buildings but in the present conditions of city streets there is much room and need for improvement. It is manifestly an impossibility to keep floors and seats in public conveyances free from dust and therefore from micro-organisms. Each passenger brings in from the streets on boots and clothing a fresh supply and the dried product is raised into the air by every movement of the passengers and by every draught of air.

The examination of air for bacteria has been undertaken in many ways; in most cases the air has been drawn by means of an aspirator:-

- (a) Through glass tubes lined with nutrient gelatine or agar.
- (b) Through a sterile fluid.
- (c) Through Petri's sand filter or some modification of it.

Hesse (12) used a straight round tube lined with gelatine or agar through which air was drawn.

Pawlowsky (13) found that with the straight tube some organisms were carried through by the air current, and substituted for the straight tube a tube bent

bent four times each angle being 45 degrees, each limb 15 centimetres long, the air tube being lined with nutrient gelatine. Air was passed through at the rate of one litre per hour. Most of the organisms were caught at the first and third angles of the tube. Colonies were then counted as in Esmarck's roll tube. William (14) used a spiral tube lined with Lævulose. After the passage of air the medium was washed out with a definite quantity of sterile water and this resulting fluid was plated with gelatine --a rather expensive and more difficult method.

Miquel (15) and Kammerer (16) used an apparatus by which the air was aspirated through a bulb containing sterile fluid. After aspiration the resulting fluid was plated with gelatine.

Straus and Wurtz aspirated through liquid gelatine which was then plated.

Petri introduced the sand filter for the analysis of air, the air being aspirated through two layers of sand in a glass tube, the sand being retained between two pieces of wire cloth. After the passage of the air the sand is placed on a Petri's plate and covered with gelatine or agar. To some extent the resulting colonies are obscured by the grains of sand. This

This may be obviated by dividing the sand among several plates.

Another means of overcoming the obscuration has been suggested, that of using a soluble medium in place of the sand in the filter.

Miquel (17) used sodium sulphate.

Frankland (18) and Sedgwick (19) used pulverised sugar but with these there is the disadvantage of possible variation in the plating medium.

Ficker (20) used finely granular glass in place of the sand, the transparency of the glass overcoming the difficulty of the obscuration of the colonies caused by the sand. In comparison with the other methods glass has been found superior to all, Petri's sand filter showing next best results.

Firth (21) used a Petri's sand filter, the sand being in a two inch layer. After the passage of air the sand was emptied into a tube of liquid gelatine and after rolling the tube so as to thoroughly wash the sand in the gelatine, the gelatine was poured off into a plate leaving the sand in the tube. This was again washed in liquid gelatine which was again poured off. After a third washing the sand was poured out with the gelatine. Firth tested the

the efficacy of the sand filter and found that only 4% of the micro-organisms passed through the filter. Instead of the usual method of aspirating the air by water displacement or air pump Firth used a Prudden's aspirator, a metallic cylinder which was exhausted of its air, then the filter was attached to a stopcock and when the stopcock was turned the air passed in leaving the organisms in the filter. The cylinder fitted into a small handbag and could be carried and used without attracting much attention.

One great advantage of this method is the easily handled apparatus and its use disturbs the air to a very slight extent in the immediate neighbourhood where the test is being made.

In my own observations as to the occurrence of micro-organisms in the air of railway carriages I have used Petri's plates using agar agar and glycerine agar agar as media.

The plates had been carefully prepared each time they were used, being first well washed, then sterilised in the hot-air chamber at 170°C . for two hours then plated with newly sterilised agar agar and glycerine agar and then the whole sterilised in the steam steriliser for half-an-hour. Each was then enclosed

enclosed in a sterilised box which was sterilised after each exposure, so that in carrying to and from the laboratory they were fully protected from possible contamination by draughts. The boxes were always carried so that the plates were right side up and horizontal.

In the railway carriages the plates were exposed while the train was in motion, with closed windows so that ordinarily there was no disturbance caused other than that which would occur by the entry of a passenger with light parcels. In some cases, as will be noted in observations in connection with the plates, the seat cushions were lightly struck once and after a definite period of time to allow the dust to settle, the plates were exposed, the time of exposure being carefully noted in each case. This method furnished more organisms than were found at any other time, the number being sometimes too great for the ready clearing up of the plate.

One fact worthy of note is the contrast between the numbers of colonies on the plain agar agar and those on the glycerine agar agar, in every case the glycerinated medium having fewer colonies due probably to the germicidal action of the glycerine. In every

every case the conditions of exposure were exactly the same.

For examination of colonies a two-third inch objective was used; for films one-twelfth inch oil immersion with No.3 ocular.

Agar cultures were incubated at 37°C.
Gelatine cultures at room temperature about 20°C.

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EXPOSURE I. - Date. - 21st January 1901.

Agar and glycerine agar plates were exposed to the air in the central compartment of carriage No. 613, 3rd Class Suburban line of North British Railway.

Time and exposure. - Five minutes.

Conditions. - Train in motion: windows closed

Results. -

Glycerine Agar Plate.

21st January 1901.	Incubated		
22nd	-	Do.	No growth
23rd	-	-	8 Colonies
25th	-	-	10 -
28th	-	-	11 -
30th	-	-	11 -

Of these eleven colonies one was a bacillus, all the

the others cocci.

Bacillus I. - G.A. 21.1.01.

Plate colony was small, round, and about 2mm. in diameter. At first it was cream-coloured changing to light brown after several days incubation.

Under Low Power. - Colony was opaque and had a finely granular appearance with clean edges.

On Sloped Agar. - Growth was of light brown colour thinly covering whole surface of medium. Surface of growth was glistening.

Gelatine Stab Culture. - Growth not very profuse; along needle track of a yellowish brown colour; some small isolated colonies at lower end of needle track. Growth was most profuse in a crack in the gelatine. There was no liquefaction of gelatine and no formation of gas in plain gelatine or in lactose gelatine.

In Peptone Broth. - Became cloudy with a slight deposit. In the medium growth gave an extremely bad odour. On testing for indol a very faint rosy colour was developed.

Milk was coagulated in 48 hours.

Potato. - There was slight yellowish brown growth with glistening surface.

surface.

Under the microscope. -

Organism is rod-shaped varying in length from 3-7 microns. Ends are rounded. It is non-motile. Some curved forms are seen. No spores were found.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

Two varieties of cocci were found.

Coccus I. - G.A. 21.1.01.

Plate colony was small, round, and about 3mm. in diameter, of white colour.

Under Low Power. - . opaque, granular, edges clean.

On Sloped Agar. - A thin white growth covered the whole surface of the medium with glistening surface.

Gelatine Stab Culture. - Along the needle track a profuse whitish yellow growth with a few isolated colonies towards the lower end of the track. On the surface a few yellowish isolated colonies. There was no liquefaction of gelatine.

Peptone Broth. - Became very cloudy with slight white deposit.

Milk was not coagulated in fourteen days.

days.

On Potato. - Growth was slight and of yellowish white colour.

Under the microscope. -

Organism is a coccus somewhat smaller than Staphylococcus pyogenes aureus. It is found singly and in groups.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

Of this coccus there were four colonies.

Coccus II. - G.A. 21.1.01.

Plate colony was about 5mm. diameter, round, and of white colour.

Under Low Power. - Opaque, finely granular, clean edges.

On Sloped Agar. - Thick creamy growth extending about 2mm. on each side of needle track.

Gelatine Stab Culture. - Profuse white growth along the needle track. On surface of gelatine a small circular growth of white colour. No liquefaction of gelatine.

gelatine.

Peptone Broth. - Became cloudy and there was a thick white deposit.

Milk was coagulated in twenty-four hours.

On Potato. - A dense whitish growth with dull rough surface.

Under the Microscope. -

Organism is a coccus about the size of Staphylococcus pyogenes aureus. It occurs singly and in groups. A few short chains of four or five are seen.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

Of this coccus there were six colonies.

Summary.

Bacillus I.	1
Coccus I.	4
Coccus II.	6

Agar Plate.- 21st January 1901.

21st January 1901		incubated	
22nd	- - -		7 Colonies
23rd	- - -	12	-
25th	- - -	13	-
28th	- - -	20	-
30th	- - -	20	-

Of these two were bacilli, the balance cocci and sarcinae.

BACILLUS I. A. 21. 1. 01.

Plate Colony was of oval shape about 2 x 3 mm. light brown in colour.

Under Low Power.- Colony was finely granular, translucent, with clean edges.

On Sloped Agar.- A thin light brown film with dull surface covered most of the medium; a few small isolated colonies of same colour were present.

Gelatine Stab.- Slight growth of light brown colour along the needle track and slight surface growth of same colour. There was considerable liquefaction of gelatine and no gas formation.

Gelatine Stroke.- Slight brown coloured growth along needle track.

track.

Peptone Broth.- Became cloudy; there was no deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- A dense dirty white growth elevated at centre and of brownish colour.

Under the Microscope.-

Organism is rod shaped varying in length from forms which are little more than cocci to others about 5 microns long. Ends are rounded. Some curved forms are seen. It is motile. No spores were found.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

BACILLUS II.- A. 21. 1. 01.

In all respects corresponded with Bacillus found
Glycerine
on Agar Plate.

COCCUS I.- A. 21. 1. 01.

Plate Colony.- White, round, about 6mm. in diameter, with irregular edges. Has the appearance of an overflow from the centre.

Under Low Power.- Opaque and granular. Edges

Edges are irregular but clean.

On Sloped Agar.- A thick copious growth of dirty white color covering most of the surface.

Gelatine Stab.- Copious white growth along needle track followed by almost complete liquefaction of the gelatine and at bottom a dirty white deposit.

Peptone Broth.- Became very cloudy with considerable white deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- A copious whitish growth, surface having a granular appearance.

Under the Microscope.-

Organism is a coccus somewhat smaller than Staphylococcus pyogenes aureus. Occurs singly and in groups. Occasionally in short chains of three to eight.

Staining Reactions.-

Stains well with ordinary stains, but does not retain Gram's stain.

COCCUS II.- A. 21. 1. 01.-

Plate Colony.- Faintly white, round, about 4mm. in diameter.

Under Low Power.- Translucent, faintly granular. Clean edges.

On Sloped Agar.- Growth a thin film, faintly

faintly white, covering most of surface; a few isolated colonies also seen. Surface glistening.

Gelatine Stab.- Considerable white growth along line of puncture. Much liquefaction of gelatine with dense deposit at bottom, of white color.

Peptone Broth.- Became cloudy. Considerable white deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- Copious white growth. Surface granular and glistening.

Under the Microscope.-

Organism is a coccus about the size of Staphylococcus pyogenes aureus. Occurs singly and in groups.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

COCCUS III.- A. 21. 1. 01.

Plate Colony.- White, round. About 3mm. in diameter.

Under Low Power.- Opaque and granular. Edges clean.

On Sloped Agar.- Copious white growth covering the greater part of surface of medium.

medium.

Gelatine Stab. - Considerable white growth along the needle track, with slight liquefaction of gelatine.

Peptone Broth. - Became slightly cloudy. No deposit.

Milk. - Was coagulated in twenty-four hours.

On Potato. - Copious growth of a dirty white colour.

Under the Microscope. -

Organism was a coccus about the size of Staphylococcus pyogenes aureus. Occurs singly and in groups.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

SARCINA LUTEA.

Plate Colony. - Of this organism there were seven colonies of various sizes, canary yellow in colour.

Under Low Power. - Opaque and granular; edges irregular but clean.

On Sloped Agar. - Copious growth of canary yellow colour along needle track, slightly raised at the

the edges, surface glistening.

Gelatine Stab.- Copious growth along needle track paler than that on agar. Gelatine is liquefied: there is a slight surface scum and heavy deposit at bottom of the liquid gelatine.

Peptone Broth.- Became cloudy and a slight deposit formed.

Milk.- Was coagulated in forty eight hours.

On Potato.- Profuse growth of canary yellow colour, raised at centre, covering most of surface: surface of growth glistening.

Under the Microscope.-

Organism is a sarcina occurring singly, in pairs, fours, cubes and masses.

Staining Reactions.-

Stains well with ordinary stains and retains

Gram's stain.

Summary

		Colonies
Bacilli	2	
Sarcina Lutea	7	-
Coccus I	1	-
Coccus II	2	-
Coccus III	4	-
Coccus II G.A.	4	-

EXPOSURE II. -

Date. - 25th January 1901.

Agar and glycerine agar plates were exposed to the air in the central compartment of carriage No.202, 3rd Class N. B. R. Suburban line.

Time of exposure. - Three minutes.

Conditions. - Train in motion; windows closed.

Results:- Glycerine Agar Plate.

25th January 1901. - Incubated

26th	-	-	Do	1 Colony
28th	-	-	-	4 Colonies
29th	-	-	-	5 -
30th	-	-	-	5 -

All were cocci, corresponding to those already described.

Summary.

Coccus II G.A. -	21. 1. 01.	3 Colonies
Coccus II A.	21. 1. 01.	1 -
Coccus III A.	21. 1. 01.	1 -

Agar Plate.

25th January 1901	incubated	
26th	-	5 Colonies
28th	-	11 -

and one mould which covered nearly the whole plate, as it was in fruit stage, plate was destroyed.

Mould was a variety of Mucor.

Only two of the colonies were examined--a bacillus and a coccus. Of the remainder three had the plate colony appearance of Sarcina Lutea.

Lutea.

BACILLUS I.- A. 25. 1. 01.

Plate Colony.- White, round about 3mm. diameter.

Under Low Power.- Opaque and granular: edges clean.

On Sloped Agar.- A thin white film covered the whole surface of the agar: dull surface.

Gelatine Stab.- Copious white growth along the needle track. On surface a thick growth of whitish colour. No liquefaction of gelatine.

Lactose Gelatine.- No gas formation.

Peptone Broth.- Became slightly cloudy and there was a very slight whitish deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- A thin growth of dull white colour.

Under the Microscope.-

Organism is rod shaped: slender with rounded ends, and is non motile. Length from 3 - 5 microns. No spores were found.

Staining Reactions.-

Stains well with ordinary stains and does not retain Gram.

COCCUS I.- A. 25. 1. 01.

Plate Colony.- White, round about 5mm. diameter

diameter, of creamy consistence.

Under Low Power.- Opaque and granular, edges clean.

On Sloped Agar.- A thin film, white, covered most of the surface. Isolated colonies similar to that on the plate were seen: surface of growth glistening.

Gelatine Stab.- Slight growth along needle track, with beaded appearance at edges due to the many isolated colonies. No liquefaction of gelatine.

Peptone Broth.- Became cloudy. Slight white flocculent deposit.

Milk.- Not coagulated in fourteen days.

On Potato.- Slight white growth, glistening surface.

Under the Microscope.-

Organism is a coccus about size of *Staphylococcus pyogenes aureus*, though some smaller forms are seen.

Staining Reactions.-

Stains well with ordinary stains and retains

Gram's stain.

Summary

Bacillus I A.-	25. 1. 01.	1	Colony
Coccus I A.-	25. 1. 01.	1	-
Mould		1	-
Not examined		8	Colonies

EXPOSURE III. - Date. - 29th January 1901.

Agar and glycerine agar plates were exposed to the air in west end (Waverley Station) Smoking compartment of carriage No.945 3rd Class N.B.R. Suburban line.

Time of exposure. - Three minutes.

Conditions. - Train in motion: windows closed: seats lightly struck and plates exposed ten minutes later.

Results:- Glycerine Agar Plate.

29th January 1901.-	Incubated		
30th	-	-	7 Not incubated
31st	-	-	12 from 2nd Feby
1st February	-	-	14
2nd	-	-	17
5th	-	-	19
6th	-	-	19

Of these, two were bacilli, the remainder being Cocci and Sarcinae.

BACILLUS I. - G.A. and A. 29. 1. 01.

Plate Colony. - Agar. A large irregularly shaped colony about 20 by 15 mm. of a dull white colour: edges raised and whiter in colour: moist and viscous. Surface dull but studded with drops like dew of about $\frac{1}{2}$ mm. in diameter.

Glycerine Agar. - Growth was not so rapid nor so copious and was more translucent.

Under Low Power. - Translucent and finely granula:

granular: edges very irregular but clean.

Appearance of the colony much resembled that of Friedlander's pneumobacillus.

On Sloped Agar.- Surface was completely covered in twenty-four hours: appearance similar to that of plate colony, but gradually became wrinkled and dry.

On Sloped Gelatine.- Growth was very slow, there being very little seen till about ten days: then growth became more rapid. At four weeks was creamy white in colour: surface much wrinkled, thin and not wide spread.

Gelatine Stab.- Growth even slower than on the sloped gelatine. At four weeks there was a thin wrinkled film on the surface: along needle track dense white growth and a few small isolated colonies at lower end of needle track. There was very slight liquefaction.

Lactose Gelatine.- Shake culture. There was no gas formation.

Peptone Broth.- Not cloudy. A thick buff-coloured surface scum formed sufficiently strong to support the broth when tube was inverted: it extended up the sides of the tube for nearly a quarter of an inch above the broth. Surface of scum was much wrinkled and on it were clear dew-like drops.

Milk.- Was coagulated in twenty-four hours.

hours.

On Potato.- Profuse buff-coloured growth much wrinkled covering whole surface.

Under the Microscope.-

Organism is rod shaped, freely motile: ends rounded, showing great variation in length from about 4 microns to long filaments: some chains also seen: they showed great tendency to adhere to each other.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain. No spores were observed.

BACILLUS II.- G.A. 29. 1. 01.

Plate Colony.- A thin film of dull white colour about 4mm. diameter.

Under Low Power.- Finely granular translucent clean edges.

In subcultures no bacilli were found: probably due to mismanagement in transferring: the resulting cultures showed cocci only, as next described.

Under the Microscope.-

Organism is rod shaped: varies greatly in length: occasional short chains of three or four granules of protoplasm are seen at various parts of the organism but most frequently were terminal.

terminal.

COCCUS I.- G.A. 29. 1. 01.

From colony as described under Bacillus II. G.A.

Plate Colony.- A thin film of dull white colour about 4mm. diameter.

Under Low Power.- Finely granular translucent clean edges.

On Sloped Agar.- A very thin film extending about 2mm. each side of needle track with a few isolated circular colonies scattered over the surface, growth almost colourless and transparent.

Gelatine Stab.- Copious creamy white growth along the needle track. At lower end, small isolated globular colonies of the same colour are seen. There was no liquefaction.

Peptone Broth.- Became cloudy and fine white deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- A thin glistening growth covering most of surface: almost colourless.

Under the Microscope.-

Organism is a coccus about the size of Staphylococcus py^ogenes aureus. Occurs singly and in groups.

Staining Reactions.-

Stains unevenly with ordinary stains and retains

retains Gram's stain.

Summary. -

Bacillus I. - G.A. -	29. 1. 01.	}	1
Bacillus II. - G.A. -	29. 1. 01.		
Coccus G.A. -	29. 1. 01.		1
Sarcina Lutea			5
Coccus II. - G.A. -	21. 1. 01.		4
Coccus II. - A. -	21. 1. 01.		2
Coccus I. - A. -	25. 1. 01.		1
Coccus I. - G.A. -	29. 1. 01.		3

Agar Plate. -

29th January 1901. - Incubated

30th	-	-	-	11	Cols.	Not incubated after 2nd February.
31st	-	-	-	19	-	
1st February	-	-	-	21	-	
2nd	-	-	-	23	-	
5th	-	-	-	25	-	
6th	-	-	-	25	-	

Of these, two were bacilli, the balance being Cocci and Sarcinae.

BACILLUS I. - A. 29. 1. 01.

Already described, see Bacillus I. G.A. 29. 1. 01.

BACILLUS II. A. 29. 1. 01.

Plate Colony. - Was round, about 5 mm. diameter: almost transparent, of slightly darker colour than the agar.

Under Low Power. - Finely granular, transparent: edges clean.

On Sloped Agar. - Buff-coloured moist growth covered the whole surface: surface glistening.

Gelatine Stab. - A thin buff film covered the surface: similar coloured growth along the needle track with a few isolated colonies at the deeper part.

in three weeks. No gas formation.

Peptone Broth.- Became cloudy, with slight flocculent deposit.

Milk.- Was coagulated in twenty four hours.

On Potato.- A thin film almost colourless, surface glistening, covered the whole surface of potato.

Under the Microscope.-

Organism is rod shaped, thick, of various lengths 2 - 6 microns. Both straight and curved forms are seen, some showing clubbing at one or both ends which are rounded. In some the protoplasm is unevenly distributed, some parts not staining. On staining with Carbol. fuchsin, counter staining with Methylene Blue, no spores were observed. The organism is motile.

Sarcina Lutea was present in 9 colonies.

Summary.

Bacilli	2
Sarcinae Lutea	9
Coccus II G.A.- 21. 1. 01.	5
Coccus I A.- 21. 1. 01.	6
Coccus I G.A.- 29. 1. 01.	3

EXPOSURE IV. - Date. - 1st February 1901.

Agar and glycerine agar plates were exposed to the air in west end (at Princes St) compartment of carriage No.6 3rd Class Caledonian Railway, Leith Branch.

Time of exposure. - Three minutes.

Conditions. - Train in motion: windows closed.

Results:- Glycerine Agar Plate.

1st February 1901.	-	Incubated		
2nd	-	-	7.	Not incuba-
5th	-	-	incubated 10.	ted 2nd to
7th	-	-	-	15. 5th February.
9th	-	-	15.	

Of these one was bacillus, the remainder being Cocci and Sarcinae.

BACILLUS I. - G.A. - 1. 2. 01.

Plate Colony.- A very large irregular colony covering almost half the plate (four inch) thin fawn coloured, with glistening surface. It obscured five white circular colonies which were not examined but which presented the low power appearance of Cocci (three), Sarcinae Lutea (two).

Under Low Power. - Translucent and faintly granular: edges irregular but clean.

On Sloped Agar. - Thin fawn coloured growth

glistening.

Gelatine Stab.- Surface is cupped: the cupping central and including almost whole surface: a narrow rim of gelatine remaining. The growth in the cup is filmy and fawn coloured: under surface brownish colour and slight brown growth along needle track, at the upper part of which many small globular brown colonies are seen.

There was no liquefaction and no formation of gas.

Peptone Broth.- Became cloudy, a dense dirty white deposit and a slight surface film.

Milk.- Was coagulated in twenty-four hours.

On Potato.- Thin glistening film covered whole surface, of fawn colour.

Under the Microscope.-

Organism is rod shaped and shows great variation in length. Is motile.

Spores were present on fourth day: large oval.

Staining Reactions.-

Stains faintly with Thionin Blue and other ordinary stains. Stained by Gram's method without decolourizing: some forms took up the stain very well: spores also stained fairly well except centrally. Some of the bacilli stained feebly by this method. Staining for spores by Ziehl-Nielsen method they were not affected by the Carbol Fuchsin.

Fuchsin.

With Gram's method some retained the stain while others and the spores completely decolourized.

COCCUS I.- A.G. 1. 2. 01.

Plate Colony.- A small round colony about 4mm. diameter, white and moist: somewhat viscid.

Under Low Power.- Opaque and granular: clean edges.

On Sloped Agar.- A thin white growth, moist, viscid and glistening, covered the whole surface

Gelatine Stab.- Profuse white growth along the needle track: very little surface growth: many isolated colonies at deeper parts.

No liquefaction of gelatine.

Peptone Broth.- Slightly clouded and very slight white deposit.

Milk.- Was coagulated on fourth day.

On Potato.- Centrally raised white growth, around it thin glistening growth covering whole surface.

Under the Microscope.-

Organism is a coccus about the size of Staphylococcus pyogenes aureus: occurs singly and in groups.

Staining Reactions.-

Stains rather unevenly with ordinary stains and

and retains Gram's stain.

Summary.

Bacillus I. - G.A. - 1. 2. 01.	1
Coccus I. - 1. 2. 01.	1
Not examined	5
Sarcinae Lutea	5
Coccus II. - G.A. - 21. 1. 01.	2
Coccus I. - A. - 21. 1. 01.	1

Agar Plate. - 1st February 1901.

1st February 1901	incubated		
2nd - -		16.	Colonies Not incubated 2nd to 5th.
5th - -	incubated	18.	
7th - -	-	21.	
9th - -		21.	

Of these two were bacilli, the remainder Cocci and Sarcinae.

BACILLUS I. - A. - 1. 2. 01.

Plate Colony. - A many pointed star shaped colony, greatest diameter about 10mm., yellowish white colour: surface moist and wrinkled.

On Sloped Agar. - Growth covered the whole surface in forty eight hours: translucent. Surface was wrinkled, the raised parts being whiter than the rest and dry.

Gelatine Stab. - There was considerable irregular cupping of the gelatine with a very little liquefied gelatine: a thin white filmy growth lined the cup:

cup: along the needle track there was a copious white growth, small globular colonies at edges giving it a beaded appearance, at the bottom of the puncture a few globular white isolated colonies were seen. There was no gas formation either in this or in Lactose Gelatine.

Peptone Broth.- Became slightly clouded and a heavy deposit. On surface a thin buffy wrinkled scum not adherent to the side of the tube as in case of Bacillus I A.- 29. 1. 01.

Milk.- Was coagulated in forty eight hours.

On Potato.- Thin wrinkled filmy growth yellowish white in colour covered whole surface in twenty four hours.

Under the Microscope.-

Organism is rod shaped, straight and curved, ends rounded: shows considerable variation in length from three to ten microns, some filaments and some short chains also seen. The organism is motile. In film preparation tends to clumping: - No spores were observed.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

stain.

The bacillus in growth in Broth and on Agar to some extent resemble that of Bacillus I. A.- 29. 1. 01., but growth in gelatine, under same conditions, is much more rapid and shows slight liquefaction and cupping, neither of which occurred in gelatine culture of Bacillus I. A.- 29. 1. 01.

BACILLUS II. A.- 1. 2. 01.

Plate Colony.- Small, round, about 2mm. diameter, faint white colour: translucent.

Under Low Power.- Finely granular, transparent, crenated edges.

On Sloped Agar.- A very thin, almost colourless film along needle track about 2mm. width: edges crenated, slightly raised, white colour: surface of growth glistening.

Gelatine Stab.- Growth along needle track copious, white colour: a few isolated colonies at deeper part of the puncture. There was no liquefaction and no gas formation.

Peptone Broth.- Became slightly cloudy and a slight white deposit was formed.

Milk.- Was not coagulated in fourteen days.

On Potato.- A very thin filmy growth with

with glistening surface covered almost all the potato surface.

Under the Microscope.- Organism is rod-shaped.

Is rod shaped bacillus, non motile, with great variation in length: some very short and thick and some very long, the latter not always uniform in thickness, ends are rounded. Some curved forms are seen.

On the film some cocci are seen, singly, in pairs and groups. These I found impossible to separate from the bacilli.

Staining Reactions.-

Stains well with ordinary stains but does not retain Gram's stain.

Summary.

Bacilli	2
Sarcinae Lutea	10
Coccus II. G.A.- 21. 1. 01.	5
Coccus I. A.- 21. 1. 01.	1
Coccus I. G.A. 1. 2. 01.	3

EXPOSURE V.- Date.- 6th February 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from east end (at Waverley Station) of carriage No. 174 3rd Class N.B.R. Suburban line.

Time of exposure.- Three minutes.

Conditions.- Windows closed: train in motion.

Results:- Glycerine Agar Plate.

6th February 1901.- Incubated				
7th	-	-	-	1 Colony
8th	-	-	-	2 Colonies
9th	-	-	-	3 -
12th	-	-	-	3 -

Of these one was *Sarcina Lutea*, the other two both Cocci, the same as *Coccus II G.A.*- 21. 1. 01.

Agar Plate.- 6th February 1901.

6th February 1901.- incubated				
7th	-	-	-	4 Colonies
8th	-	-	-	7 -
9th	-	-	-	10 -
12th	-	-	-	10 -

Of these one was bacillus; remainder were cocci and sarcinae.

BACILLUS I.A.- 6. 2. 01.

Plate Colony.- Brownish white, about 8mm. diameter, round, surface granular and glistening:

glistening: edges crenated.

Under Low Power.- Edges are crenated but clean, central part of colony more dense than towards edges and coarsely granular.

On Sloped Agar.- Growth thick, greyish white, moist, spreading over most of the agar surface. Edges are crenated, surface is glistening.

On Sloped Gelatine.- Copious growth at first bluish white, moist: later became greyish white. Spreads over most of the gelatine surface.

Gelatine Stab.- Along needle track a chain of small globular colonies at first white later becoming light brown: on surface a thick greyish white growth: gas bubbles formed in the medium at first along the needle track later invading the rest of the gelatine. There was no liquefaction.

Gelatine Shake.- Gelatine became slightly turbid and numerous gas bubbles formed in twenty four hours throughout the whole of the medium. Small whitish colonies were present on the surface.

Lactose Litmus Gelatine.- Gas formation very marked and production of acid shown by change of colour, the medium becoming red in less than forty eight hours.

Peptone Broth.- Became turbid in twenty four hours and gave distinct acid reaction.

reaction.

On doing Indol reaction, pink colour was noted.

Milk.- Was coagulated in forty-eight hours.

On Potato.- Growth was profuse, moist and raised of dirty white colour: surface glistening.

Under the Microscope.-

Organism is rod shaped, motile, showing variation in length, some forms much resembling large cocci, other forms are very long and filamentous: most often is a short thick bacillus with rounded ends.

Staining Reactions.-

Stains well with ordinary stains and does not retain Gram's stain.

Stained by the Pitfield-Muir method, Flagella are seen faintly stained.

In every way this organism resembles Bacillus Coli Communis, so is probably a kindred organism.

Summary.

Bacillus	1
Sarcina Lutea	4
Coccus II. G.A.- 21. 1. 01.	2
Coccus I. A.- 25. 1. 01.	3

EXPOSURE VI. - Date. - 12th February 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from West end (at Princes Street) of carriage No. 352 3rd Class Caledonian Railway, Leith Branch.

Time of exposure. - Two Minutes.

Conditions. - Train in motion: windows closed.

Results:- Glycerine Agar Plate.

12th February 1901.	Incubated
13th - -	- 1 Colony
14th - -	- 2 Colonies
15th - -	- 2 -
18th - -	- 2 -

One was a coccus already described, *Coccus III. A.* - 21. 1. 01., the other *Sarcina Lutea*.

Agar Plate. - 12th February 1901.

12th February 1901.	incubated
13th - -	- 7 Colonies
14th - -	- 12 -
15th - -	- 16 -
18th - -	- 17 -

Of these one was mixed *Sarcina* and *Bacillus*, nine *Sarcina Lutea* and seven Cocci.

One colony when examined showed *Sarcinae* and numerous bacilli.

The bacilli were slender rods, varying in length from

from three to eight microns, straight and curved forms. They stained much more feebly with Thionin Blue than did the Sarcinae.

On making subculture, the bacilli were not again found either from growth on Agar or Gelatine.

Plate Colony I.- Was white, round, about 4mm. diameter, with crenated margins.

Under Low Power.- Opaque and granular: edges clean.

On Sloped Agar.- A dense ivory white growth spreading irregularly over the greater part of the surface: edges crenated: surface glistening. Many small isolated colonies were seen.

On Sloped Gelatine.- Growth was pure white, hardly so copious as on agar, extending about 1mm. each side of needle track: edges crenated: surface glistening, isolated colonies also present.

Gelatine Stab.- Considerable growth along needle track: a beaded appearance caused by many small colonies at edge of growth and many isolated colonies, white and globular seen at lower end of needle track. On surface there were three small white colonies, round with glistening surface. No liquefaction of gelatine.

Peptone Broth.- Became cloudy: no surface film but a plentiful white deposit at bottom of tube.

tube.

Milk. - Was not coagulated in fourteen days.

On Potato. - Profuse white growth with glistening surface.

Under the Microscope. -

Organism is a sarcina seen in pairs, fours, eights and masses. The Bacillus already described.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

From the colour of the colony and the fact that there is no liquefaction of gelatine the organism is probably either *S. Pulmomum* or *S. Ventriculi*.

COCCUS I. - A. - 12. 2. 01.

Plate Colony. - Is round, of pale yellow colour, about 6mm. diameter.

Under Low Power. - Opaque, granular: edges clean.

On Sloped Agar. - Growth is profuse and thick, pale yellow colour covering almost the whole agar surface, edges crenated: surface glistening, growth is of creamy consistence.

Gelatine Stab. - Growth along needle track slight, faint yellowish white in colour, almost transparent,

transparent, at edges many small isolated colonies. On gelatine surface many small isolated colonies of faint yellow colour. No liquefaction of gelatine.

Peptone Broth.- Became very cloudy: slight surface scum of yellowish white colour and dense deposit of same colour.

Milk.- Was not coagulated in fourteen days.

On Potato.- Profuse growth of pale yellow colour, raised at centre, surface glistening.

Under the Microscope.-

Organism is a coccus about same size as Staphylococcus pyogenes aureus: occurs singly, in pairs and groups.

Staining Reactions.-

Stains very unevenly with Thionin Blue, some cocci hardly taking up any stain, retains Gram's stain.

Summary.

Mixed Bacillus and Sarcina.	1
Sarcina Lutea	9
Coccus I. A.- 12. 2. 01.	3
Coccus I. A.- 21. 1. 01.	1
Coccus III A. 21. 1. 01.	3

EXPOSURE VII. - Date. - 18th February 1901.

Agar and glycerine agar plates were exposed to the air in central compartment of carriage No.540, Caledonian Railway, Leith Branch.

Time of exposure. - One minute - exposure being made five minutes after seats were lightly struck to raise dust.

Conditions. - Train in motion; windows closed; seats "dusted" as above noted.

Results:-

18th February 1901.-	Incubated.		
19th	-	-	7 Colonies
20th	-	-	9
21st	-	-	13
22nd	-	-	17
24th	-	-	17

Of these, two were bacilli - two were moulds, five *Sarcina Lutea*, and 8 Cocci.

BACILLUS I. - G.A. - 18. 2. 01.

Plate Colony. - Was thin and colourless - transparent at edges, somewhat darker centrally; in shape oval about 5 by 7 mm.; at edges irregular loops of growth extending out over the medium and doubling back again into the colony without a break; surface glistening.

Under Low Power. - Transparent except at the

the centre: faintly granular. Does not exhibit the striated appearance of plate colonies of Anthrax Bacillus. Edges show the looping well and many loose wavy ends. Not unlike those seen in plate colonies of B. Anthracis but are coarser.

On Sloped Agar.- There was little growth except at point of inoculation, where there was a small colourless colony: of much the same appearance as plate colony.

On Sloped Glycerine Agar.- Growth was more extensive, covering most of the surface of the medium. Thin and transparent except at the darker centre. Edges showing the same looping as a plate colony. Surface glistening.

Incubated at 50° C. for forty eight hours: there was no growth.

On Sloped Gelatine at room temperature.- No growth whatever.

Gelatine Stab.- No growth whatever after three weeks, at room temperature.

Incubated at 37° C. A cirrus-like ball formed in forty eight hours, at the upper part of the liquid gelatine. This growth did not become markedly larger in three weeks and did not reach to the

the surface.

Peptone Broth. - Growth was very similar to that in the liquid gelatine after incubation. The growth, however, went on rapidly and formed a light deposit at the bottom of the tube.

Milk. - Was not coagulated in fourteen days.

On Potato. - There was considerable growth. Almost colourless. With glistening surface.

Under the Microscope. -

Organism is rod shaped; motile. It varies very greatly in length. Variation is most marked in the impression film (q.v.) ; in this, short forms of two to four microns are seen and long chains and filaments running out from the centre of impression to form loops which turn back to centre again. The impression film was made from a small subculture. In ordinary film; the variation in length is not so marked, but here again chains are seen. Ends of bacilli are rounded. In some cases protoplasm is entirely absent, the sheath being faintly stained. In other bacilli protoplasm is present in small granules in the sheath; these staining much better than the sheath.

No spores were observed though specially stained for

for at different stages of growth.

In impression film some spore-like forms are seen, but these are not found in any other preparation.

Staining Reactions. -

Stains well with ordinary stains except when protoplasm is absent, and retains Gram's stain.

The plate colony resembled that of Bacillus Anthracis very much, but the failure to grow in gelatine at room temperature and the fact that the organism is motile, differentiate it from that organism.

BACILLUS II. - G.A. 18. 2. 01.

Plate Colony. - A small somewhat oval shaped colony about two by three mm. of bluish white colour; edges irregular.

Under Low Power. - Opaque, granular; edges irregular, but clean.

On Sloped Agar. - Growth covered about half the surface of the medium. Surface wrinkled and whitish in colour. Growth was drier than that on Glycerine Agar.

On Sloped Glycerine Agar. - Growth was more rapid than on plain Agar; more moist and viscous. Surface not so much wrinkled nor so white. A few isolated colonies are seen. Circular, with glistening

glistening surface.

Gelatine Stab.- On surface considerable growth: thick and raised. Copious growth along the needle track, of white colour; with many isolated white globular colonies near the needle track and towards the bottom of the puncture. Surface growth was moist and glistening. There was no liquefaction and no gas formation.

Peptone Broth.- Became slightly cloudy: on surface a light scum of yellowish white colour: slightly wrinkled: slight flocculent deposit.

Milk.- Was coagulated in twenty-four hours.

On Potato.- Considerable growth: white and wrinkled.

Under the Microscope.-

Organism is rod shaped, showing considerable variation in length. Chains of three or four bacilli also seen. Some curved forms are seen and in some there is a slight clubbed appearance at one end of the bacilli. In others protoplasm is at end only the middle portion being clear. There is some variation in breadth some forms being much more slender than most. No spores were observed though specially examined for at various stages of growth.

Staining Reactions.-

Stains well with ordinary stains and retains

retains Gram's stain.

This organism resembles Bacillus I. A.- 29. 1. 01. but growth in gelatine is more rapid and profuse. It also resembles Bacillus I. A.- 1. 2. 01. but differs from it in that gelatine does not become cupped.

Glycerine Agar Plate.- 18. 2. 01.

The Moulds.- There were two colonies about twenty mm. in diameter. A dense white fluffy growth with colourless branching mycelium extending one eighth of an inch beyond the edge of the white growth.

On Sloped Agar.- Growth was the same.

Gelatine Stab.- Surface was covered with dense white fluffy growth. Throughout medium individual colonies can be seen with almost colourless rays radiating in all directions. There was considerable liquefaction of the gelatine.

On Potato.- Growth was similar: white and fluffy.

Under the Microscope.-

The moulds presented the appearance of Penicillium Glaucum, but the growth had not the blue or green

colour of that mould.

<u>Summary</u>		
Bacilli		2
Moulds		2
Sarcina Lutea		5
Coccus II. G.A.- 21.1.01.		4
Coccus III. A.- 21.1.01.		2
Coccus I. A.- 25.1.01.		2

Agar Plate.- 18.2.01.

Exposed one minute as already mentioned. Plate was incubated. On afternoon of 19th February about twenty-two hours after exposure, the whole plate was covered with a thin brownish white film, moist, with a glistening surface. This growth covered eight other colonies both white and yellow none of which were examined. A peculiar fetid odour was noticeable on raising the cover from the plate.

Under Low Power.- Translucent and granular.

On Sloped Agar.- The growth covered the whole surface in twenty hours, and was of the same colour except at edges where agar touched the glass where growth was whiter than elsewhere. Surface was moist and glistening.

Gelatine Stab.- In this medium growth at first was white, after a few days becoming brown at the deeper parts. Isolated white colonies are seen which also became brown. About ten days after inoculation the gelatine became cracked for about an inch in depth, from the sides of this and farther down the needle track, gradually including the whole needle track, the growth assumed a peculiar form--Gas (?) bubbles growing into the medium in form of an oxborn, the

the small end outwards, the tip of the horn being represented but a small brown knob which pushes its way into the gelatine. The "bubbles" are lined with whitish growth. The large end opens into the crack into the gelatine and lower down into the open space caused by the disappearance of the gelatine.

These horn-like processes curve in all directions after extending into the medium a short distance. When they near the glass, they turn again and follow round the tube. They are longest at the upper part becoming very small towards the bottom of the needle track. This peculiar appearance of the gas bubbles gradually invaded the whole needle track, until eight weeks after inoculation, the whole tube was affected.

A second tube was inoculated later on; with the idea that the gelatine in first tube might have been too stiff but in this second tube the gelatine cracked the ninth day and the pitting of the gelatine assumed the same appearance. In this tube there was no liquefaction of gelatine.

Gelatine Stroke. - Along the needle track, a dense white growth gradually changing to brown which has "eaten" in the gelatine, a channel which gradually became wider and deeper. In this tube gelatine

gelatine became slightly liquefied and a small quantity of fluid accumulated at the bottom with a dense brownish white deposit. Over the remaining gelatine surface a thin bluish white film formed.

Gelatine (Lactose).- The medium became somewhat cloudy and many small whitish colonies were also seen but there was no formation of gas throughout the medium

The surface is much pitted as if the gelatine had been suddenly cooled when the surface was frothy. Cloudiness is most marked immediately below the pitted surface.

Peptone Broth.- Became very cloudy at first with slight brownish deposit: later deposit became very dense, thick and slimy. The broth otherwise fairly clear. There is no surface film.

Milk.- Was coagulated in twenty-four hours.

On Potato.- A thin glistening film of dirty white colour- moist - covered the whole surface.

Under the Microscope.-

An impression preparation of the culture made twenty four hours after exposure, showed that the organism is rod shaped with spores at one end. In some cases it seems terminal: in others not actually terminal. The organism much resembling the Bacillus

Bacillus of Quarter-evil. There is some variation in length, three to nine microns.

The spores are large and oval, nearly twice the thickness of the bacillus and about two microns in length. Some forms of the bacillus exhibited slight motility, other than Brownian movement, which was also present. In ordinary film preparations, spores are seen lying free and stain well with Carbol fuchsin. The bacilli counter staining well with Methylene Blue.

Staining Reactions. -

Stains well with ordinary stains and retain Gram's stain.

The eight colonies covered by the growth were not examined.

EXPOSURE VIII. - Date. - 20th February 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from East end (at Waverley Station) of carriage No.936, N.B.R. 3rd Class.

Time of exposure. - Three minutes.

Conditions. - Train in motion; windows closed.

Results:- Glycerine Agar Plate.

20th February 1901. - Incubated

21st	-	-	-	2 Colonies
22nd	-	-	-	2 -
23rd	-	-	-	3 -
26th	-	-	-	3 -

Of these, one was *Sarcina Lutea* - the other two being Cocci not previously described.

COCCUS I. - G.A. 20. 2. 01.

Plate Colony. - Oval about two by four microns of pale yellow colour; moist with glistening surface.

Under Low Power. - Translucent; coarsely granular; edges irregular but clean.

On Sloped Agar. - Copious pale yellow growth covering the greater part of the surface of agar. At edges very irregular. A few isolated colonies are seen. Growth is moist with glistening surface.

Gelatine Stab. - On surface considerable

considerable yellowish white growth with cupping and slight liquefaction of the gelatine. Along the needle track, growth is also considerable. Just below the surface it has a brownish tinge, but at deepest part is almost white. There are numerous isolated colonies near needle track, especially at the lowest part.

Peptone Broth.- Became very cloudy; with very slight whitish surface growth and dense yellowish white deposit.

Milk.- Was not coagulated in fourteen days.

On Potato.- Profuse yellow growth with very irregular glistening surface.

Under the Microscope.-

Organism is a coccus about the size of Staphylococcus pyogenes aureus. Some smaller forms are also seen. Occurs singly, often in pairs and in groups of many.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

COCCUS II.- G.A. 20. 2. 01.

Plate Colony.- Small, round, about two mm. diameter, of faint pink colour: edges translucent.

translucent.

Under Low Power. - Finely granular; translucent at edges but opaque at centre; edges regular and clean.

On Sloped Agar. - Very irregular and rather copious growth of pinkish colour. In parts very thin in others thick, so causing variation in colour. Surface moist and glistening.

Gelatine Stab. - Considerable growth of orange red colour, not unlike a drop of red sealing wax. Surface glistening. Along the needle track considerable growth lighter in colour than that on the surface. At lower end of needle track many isolated colonies are seen, of still lighter colour. There is no liquefaction of gelatine.

Peptone Broth. - Became cloudy with slight pinkish deposit.

Milk. - Was not coagulated in fourteen days. The scum adhering to sides of tube above milk surface assumed an orange colour.

On Potato. - Considerable growth with irregular glistening surface like a drop of red sealing wax.

Under the Microscope. -

Organism is a small coccus about .7 micron in diameter. Occurs singly, in pairs, threes, fours

fours and groups of many. Non motile.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

From colour of growth this organism seems to be Micrococcus Cinnabareus.

Summary.-

Sarcina Lutea	1
Coccus I. G.A. 20. 2. 01.	1
M. Cinnabareus	1

Agar Plate.- 20. 2. 01.

20th February 1901.	Incubated		
21st	-	-	4 Colonies
22nd	-	-	6 -
23rd	-	-	8 -
26th	-	-	8 -

Of these, one was a bacillus; two were cocci; one a Sarcina, white colony; and four Sarcina Lutea.

BACILLUS I.-A. 20. 2. 01.

Plate Colony.- Round; about five mm. diameter, dirty white in colour; surface glistening: edges crenated.

Under Low Power.- Translucent. Finely granular. Edges irregular but clean.

On Sloped Agar.- Growth covered the whole surface with a thin glistening film of dirty white colour

colour. Translucent.

On Sloped Gelatine. - Growth covered whole surface thicker than on Agar and whiter in colour; less translucent. At edges growth was thinner than at centre giving a fringe-like appearance. Surface was glistening.

Gelatine Stab. - Surface was slightly cupped. The surface growth was very similar to that on sloped gelatine. Growth along needle track slight. Some isolated globular colonies at deeper part of needle track, of whitish colour.

There was no liquefaction of gelatine and no gas formation in the medium, though gelatine cracked to the depth of an inch four weeks after inoculation.

Peptone Broth. - Became very cloudy. Slight white surface film and dense white deposit.

Milk. - Was not coagulated in fourteen days.

On Potato. - Profusedirty white growth over most of the surface, with irregular glistening surface.

Under the Microscope. -

Organism is rodshaped; motile. Shows great variation in length from short forms of about three microns length to long filamentous forms; some curved forms are seen. Protoplasm is unevenly

unevenly distributed, many showing clear spaces at various parts. About the centre of some bacilli from young cultures, a large spore-like body is seen which stains fairly well with thionin blue but when stained with Carbol Fuchsin - heated - did not take up the stain at all.

In old cultures these bodies are not seen. The bacilli are longer and the uneven distribution of the protoplasm is more marked. When stained with Carbol Fuchsin - heated - counterstained with Methylene Blue, no spores were seen, the organisms staining equally well with both stains.

Does not retain Gram's stain.

Summary. -

Bacillus I. A. 20. 2. 01	1
Sarcina Lutea	4
Sarcina Pulmonum (?)	1
Coccus II. G.A. 21. 1. 01.	2

EXPOSURE IX. - Date. - 25th February 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from east end (at Princes Street) of carriage 617 Caledonian Railway, Leith Branch, 3rd Class. Carriage had no back cushions.

Time of exposure. - Two minutes.

Conditions. - Train in motion: windows closed.

Results:- Glycerine Agar Plate.

25th February 1901.-	Incubated		
26th	-	-	4 Colonies
27th	-	-	6 -
28th	-	-	7 -
2nd March			7 -

Of these, two were bacilli; two cocci; and two *Sarcina Lutea*. There were two colonies of one bacillus.

BACILLUS I. - G.A. - 25. 2. 01.

Of this bacillus there were two colonies. It corresponded in every way with Bacillus described as Bacillus I. - G.A. - 18. 2. 01. (q.v.).

BACILLUS II. - G.A. - 25. 2. 01.

Plate Colony. - A round colony about six mm. diameter; almost colourless; irregular edges.

Under Low Power. - Finely granular. Transparent. Edges irregular but clean.

On Sloped Agar. - The growth covered the whole

whole surface; dense, buff coloured. Surface smooth and glistening.

Gelatine Stab. - A thin filmy whitish growth on the surface of the liquefied gelatine; at the bottom of this a dense buff coloured deposit. In the still solid gelatine, copious buff coloured growth paler than the deposit. Along the needle track, at the edge of which are many isolated colonies; light buff and globular.

Gelatine is liquefied to the depth of an inch. There was no formation of gas.

Peptone Broth. - Became clouded with slight flocculent deposit.

Milk. - Was coagulated in forty-eight hours.

On Potato. - Potato was covered with thin, moist film of buff colour with glistening surface.

Under the Microscope. -

Organism is rodshaped. Varies both in length and thickness. In length from about three microns to long forms of ten and more. Some forms are nearly double the thickness of the average. Curved forms are seen and the shorter forms are often in pairs. Bacilli show tendency to grouping. No spores were observed.

Staining Reactions. - Stains well with ordinary stains

stains and retains Gram's stain.

Summary. -

Bacillus I. - G.A. 18. 2. 01	2
Bacillus I. - G.A. 25. 2. 01	1
Sarcina Lutea	2
Coccus I. - G.A. 20. 2. 01	2

Agar Plate. - 25. 2. 01

25th February 1901	Incubated	
26th - - -		3 Colonies
27th - - -		7 -
28th - - -		10 -
2nd March		10 -

Of these, one was a bacillus; four were cocci; and five Sarcina Lutea.

BACILLUS I. A. 25. 2. 01.

Plate Colony. - Round; about 5mm. diameter; white with raised centre of brownish tinge.

Under Low Power. - Opaque; granular, edges rather translucent, crenated but clean.

On Sloped Agar. - Growth is white and moist covering most of the surface. Is thinner at edges than centre, giving a fringe-like appearance. Surface of growth gradually became drier.

Gelatine Stab. - Growth along needle track spreads out in filmy branches of about equal length. Many of these became twisted together assuming a some-

somewhat spiral appearance.

There was rapid liquefaction.

Peptone Broth.- Became clouded rapidly. Surface scum formed: at first white, later becoming brown. Considerable dirty white deposit.

Milk.- Was coagulated in five days. The whey separated. No solid curd was formed, the appearance being that of a syrupy precipitate.

On Potato.- Profuse growth, white and slimy.

Under the Microscope.-

Organism is rod shaped with rounded ends from two to three microns in length. Chains of various lengths are seen and some spores. The organism is motile.

In old cultures many spores are seen - oval in shape - rather more than one micron in length and about half a micron in breadth.

Staining Reactions.-

Stains well with ordinary stains but does not retain Gram's stain.

The spores stain well with Carbol Fuchsin - heated - bacilli counterstained with Methylene Blue.

This organism in growth on the various media and in microscopical appearance resembles *Bacillus Mycoides*.

COCCUS I.- A. 25. 2. 01.

Plate Colony.- Round, about three mm. diameter

diameter, dirty white colour, centre raised and slightly darker in shade. Surface glistening. Growth is moist and viscid.

Under Low Power. - Opaque, granular: edges irregular but clean and somewhat translucent.

On Sloped Agar. - Copious moist viscid growth of white colour about five mm. in width, centre raised and of yellowish tinge. Edges crenated. Some isolated colonies are seen, round and white. Surface of growth glistening.

Gelatine Stab. - On surface, thick, raised, viscid growth of yellowish colour. Along the needle track, thick white growth with many isolated globular colonies just outside needle track and at deeper part of the medium.

No liquefaction of gelatine.

Peptone Broth. - Became slightly cloudy and a slight white deposit formed.

Milk. - Was not coagulated in fourteen days.

On Potato. - Copious white growth; moist and viscid; surface granular and glistening.

Under the Microscope. -

Organism is a coccus about the size of *Streptococcus pyogenes*; occurs singly and in pairs but most frequently in fours. Occasionally a faint

faint capsule surrounding a tetrad can be seen but this does not stain at all well.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

This organism in growth on the various media and in microscopical appearance, groups of four, corresponds with *Micrococcus tetragonus*.

Summary. -

Bacillus (<i>Mycoides</i>) I. - A.25.2.01.	1	Colony
Micrococcus (<i>Tetragonus</i>) do.	2	Colonies
Sarcina Lutea	5	-
Coccus I. - G.A. 20. 2. 01	<u>2</u>	-
	10	-

EXPOSURE X. - Date. - 4th March 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from east end (at Waverley station) of carriage No.1099 N.B.R. 3rd Class.

Time of exposure. - One minute - seats were lightly struck to raise dust and plate was exposed five minutes later.

Conditions. - Train in motion; windows closed; seats "dusted" as already described.

Results:- Glycerine Agar Plate.

4th March 1901.	-	Incubated	
5th	-	-	2 Colonies
6th	-	-	4 -
7th	-	-	6 -
9th	-	-	6 -

These were all Cocci and Sarcinae.

Sarcina Lutea		2 Colonies
Coccus II. - G.A. 21.1.01		3 -
Coccus I. - A. 25.1.01		1 Colony

Agar Plate.

4th March 1901	Incubated	
5th	-	8 Colonies
6th	-	11 -
7th	-	16 -
9th	-	16 -

Of these, two were bacilli (of one four colonies); two were moulds; five Sarcina Lutea; and three cocci.

BACILLUS I. - A. 4. 3. 01.

Plate Colony. - There were four colonies in one

one group; smallest about two mm. diameter; the largest about five mm. diameter. These gradually joined forming a mass about fifteen by ten mm.

Isolated, they were round; of light brown colour at first and translucent.

Under Low Power. - Granular; translucent with a circular central "core" from which "rays" grew out towards circumference; the general appearance being not unlike that of a section of the stem of a dicotyledonous plant.

Colonies gradually became drier and the "core" more superficial, finally assuming a crateriform appearance. Centrally light brown in colour; round this a circle of a lighter shade and the edges of same shade as centre. Edges were very irregular but clean.

On Sloped Agar. - Growth was very profuse almost covering the whole agar surface. Thick and of light brown colour. Surface of growth very uneven and glistening. Edges very irregular. Growth extended down between agar and glass for about an eighth of an inch from surface.

Gelatine Stab. - Profuse growth along the needle track, of faint brown colour. Very slight surface growth of same colour. A few isolated colonies are seen at deepest part of the needle track. There was

was no liquefaction of gelatine and no gas formation.

Peptone Broth. - Became cloudy and a dense whitish brown deposit formed.

Milk. - Was coagulated in forty-eight hours.

On Potato. - Whole surface was covered with dense growth of very light brown colour. Surface much wrinkled and glistening.

Under the Microscope. -

Organism is rod shaped; non motile with considerable variation in length, short forms of two to three microns to long forms of eight microns or more; many curved forms are seen.

In old cultures very many spores are seen, about one micron long, rather more than one half micron in width; oval in shape. In one or two cases spores seem to be formed at ends of bacilli.

Staining Reactions. -

Stains well with ordinary stain but does not retain Gram's stain.

BACILLUS II. - A. 4. 3. 01.

Plate Colony. - Round, about four mm. diameter of light fawn colour with glistening surface.

Under Low Power. - Opaque, granular; edges irregular but clean.

clean.

On Sloped Agar.- Growth was not very profuse extending about one mm. on each side of the needle track. Colour same as that of plate colony. Several isolated colonies are seen having same appearance as the plate colony.

Gelatine Stab.- On surface considerable growth of very faint fawn colour with glistening surface. Along needle track considerable growth almost white in colour. Many small globular isolated colonies at edges of needle track and in deeper parts.

There was no liquefaction and no gas formation.

Peptone Broth.- Became slightly clouded and a dense flocculent whitish deposit formed.

Milk.- Was not coagulated in fourteen days.

On Potato.- There was no growth.

Under the Microscope.-

Organism is rod shaped with rounded ends. Considerable variation in length: some forms little more than cocci, while others are about eight microns in length. The shorter forms are often seen in pairs. Some filamentous forms are seen. It is non motile.

In old culture bacilli show even greater variation in length, some very long forms being seen. In these

these long forms, a number of minute clear rounded areas are seen, irregularly placed in the bacilli. These do not stain with ordinary stains nor with Carbol Fuchsin (heated).

Spores are present being about twice the breadth of the bacilli; oval in shape and vary somewhat in length - one to two microns.

Staining Reactions.-

Stains well with ordinary stains but does not retain Gram's stain.

COCCUS I.- A. 4. 3. 01.

Plate Colony.- Round, white colony about five mm. diameter. Centre darker than edges. Edges are irregular.

Under Low Power.- Coarsely granular; opaque: edges are irregular but clean.

On Sloped Agar.- Profuse whitish yellow growth covering almost the whole agar surface: moist with glistening surface. Numerous isolated colonies are seen similar to the plate colony.

Gelatine Stab.- Growth on surface slight, of yellowish white colour. Profuse whitish growth along the needle track. Many isolated globular colonies at edges of needle track and at deep part of

of tube.

There is no liquefaction of gelatine.

Peptone Broth. - Became cloudy and there was slight flocculent deposit of yellowish white colour.

Milk. - Was coagulated in eight days. The whey separated but curd was syrupy rather than solid.

On Potato. - Copious whitish yellow growth with irregular glistening surface.

Under the Microscope. -

Organism is a coccus, generally about the size of Streptococcus Pyogenes. Some larger forms are seen which stain more deeply than others.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

The two moulds were the same as that described under Glycerine Agar Plate of 18th February 1901.

Summary. -

Bacilli I. & II. A. 4. 3. 01.	5 Colonies
Moulds	2 -
Sarcina Lutea	5 -
Coccus I. A. 4. 3. 01.	1 -
- II. G.A. 21. 1. 01.	2 -
- I. G.A. 20. 2. 01.	<u>1</u> -
	16 -

EXPOSURE XI. - Date. - 8th March 1901.

Agar and glycerine agar plates were exposed to the air in east end compartment (at Princes Street) of carriage No.7 Caledonian Railway, 3rd Class.

Time of exposure. - One minute.

Conditions. - Train in motion; windows closed.

Results:- Glycerine Agar Plate.

8th March 1901.	-	Incubated		
9th	-	-	-	2 Colonies
11th	-	-	-	4 -
12th	-	-	-	5 -
14th	-	-	-	5 -

These were all cocci.

COCCUS I. - G.A. 8. 3. 01.

Plate Colony. - About three mm. in diameter.

Yellowish white at first becoming orange the second day. Surface smooth and glistening.

Under Low Power. - Opaque, granular; edges somewhat irregular but clean.

On Sloped Agar. - Abundant growth along the needle track; with smooth glistening surface. Bright orange colour. Edges irregular not unlike a streak of paint.

Gelatine Stab. - Growth along the needle track seen the day after inoculation. Whitish colour. Liquefaction began at the surface on the fourth day

day and proceeded rapidly, the growth falling to the bottom as a flocculent deposit of bright yellow colour. A slight yellow film formed on surface. Fluid gelatine is turbid. Liquefaction finally extended to whole diameter of tube and gelatine was almost entirely liquefied in ten days.

Peptone Broth. - Became very turbid and a dense yellowish deposit formed.

Milk. - Was coagulated in forty eight hours.

On Potato. - Considerable orange coloured growth with glistening surface.

Under the Microscope. - Organism is a coccus about the size of Staphylococcus Pyogenes Aureus; occurs singly, in pairs and in large groups of many cocci. A few short chains are seen - five to eight cocci.

Staining Reactions. -

Stains readily with ordinary stains and retains Gram's stain.

So far as can be judged from cultural and microscopical appearances, this seems to be Staphylococcus Pyogenes Aureus.

Summary. -

Coccus I. G.A. 8. 3. 01.	1 Colony
- II. G.A. 21. 1. 01.	3 Colonies
- II. A. 25. 2. 01.	2 -

AGAR PLATE. - 8. 3. 01.

8th March 1901.- Incubated				
9th	-	-	-	2 Colonies
11th	-	-	-	5 -
12th	-	-	-	6 -
14th	-	-	-	6 -

Of these, one was a bacillus; two *Sarcina Lutea* and three cocci.

BACILLUS I. - A. 8. 3. 01.

Plate Colony. - A large irregular colony about fifteen by five mm. Almost colourless except at centre where it was raised and of brownish colour.

Viscid in consistence. Surface glistening.

Under Low Power. - Transparent except at centre; finely granular; edges irregular but clean.

On Sloped Agar. - Surface almost covered with growth, very similar to that on plate. Centre part not so dark as on plate. Transparent except at centre.

Viscid.

Gelatine Stab. - Very slight growth along the needle track, of faint white colour. No surface growth. No liquefaction of gelatine and no gas formation.

Peptone Broth. - Became slightly cloudy and a slight white deposit formed.

Milk. - Was not coagulated in fourteen days.

days.

On Potato. - There was slight, filmy, almost colorless growth; surface glistening.

Under the Microscope. -

Organism is rod-shaped; motile. In length varies from three microns to filamentous forms. Occasional chains are seen. In these bacilli appear to have square ends, while single bacilli have rounded ends. Some curved forms are seen. In some bacilli protoplasm is entirely absent, the sheath alone staining faintly. In others, small areas of protoplasm are seen. No spores were observed.

Staining Reactions. -

Stains fairly well with ordinary stains. Some forms stained very faintly. Does not retain Gram's stain.

COCCUS I. - A. 8. 3. 01.

Plate Colony. - There were two about three mm. in diameter; round; faintly white in colour; edges crenated; surface glistening.

Under Low Power. - Transparent; faintly granular; edges crenated but clean.

On Sloped Agar. - Copious whitish growth over

over almost whole surface. Many isolated colonies are seen; moist and slightly viscid; surface glistening.

Gelatine Stab. - Copious growth along the needle track of light fawn colour with many small globular isolated colonies at edges and at deeper part of tube. Gelatine is cupped to the depth of a quarter inch. Inside the cup, the growth is whiter than along the needle track, No liquid gelatine and no gas formation.

Peptone Broth. - Became very cloudy and a dense white deposit formed.

Milk. - Was not coagulated in fourteen days.

On Potato. - Profuse white growth with glistening surface.

Under the Microscope. -

Organism is a coccus about the size of Staphylococcus Pyogenes Aureus; occurs singly, in pairs, threes, and groups of many.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

Summary. -

Bacillus I. A. 8. 3. 01.	1
Coccus I. A. 8. 3. 01.	2
Sarcina Lutea	2
Coccus II. A. 25. 2. 01.	1

EXPOSURE XII. - Date. - 14th March 1901.

Agar and glycerine agar plates were exposed to the air in second compartment from the east end (at Waverley Station) of carriage No. 939 N.B.K. Suburban line 3rd Class.

Time of exposure. - One minute.

Conditions. - Train in motion; windows closed.

Results:- Glycerine Agar Plate.

14th March 1901. - Incubated	
15th	1 Colony
16th	3 Colonies
18th	5 -
20th	5 -

One was *Sarcina Lutea*, the others Cocci.

<i>Sarcina Lutea</i>	1
Coccus II. A.G. 21. 1. 01.	1
- I. A.G. 1. 2. 01.	1
- II. A.G. 20. 2. 01.	2
(<i>Micrococcus Cinnabareus</i>)	

Agar Plate. - 14. 3. 01.

14th March 1901. - Incubated	
15th	4 Colonies
16th	6 -
18th	8 -
20th	8 -

Two were bacilli. One not previously described; the other, *Bacillus I.* Agar Plate 25. 2. 01 (*B. Mycoides*).

Two were *Sarcinae Luteae*; one white *Sarcina*; two Cocci not previously described and Coccus II. A.G.

A.G. 20. 2. 01.

BACILLUS I. - A. 14. 3. 01.

Plate Colony. - Small white irregularly spread: edges very irregular: surface glistening.

Under Low Power. - Opaque, granular: edges irregular but clean.

On Sloped Agar. - Growth about two mm. in width along needle track: white with glistening surface. A few small isolated colonies are seen.

Gelatine Stab. - Very slight, faint white growth along the needle track. A few very small globular colonies are seen. There was no liquefaction of gelatine and no gas formation.

Peptone Broth. - Became slightly clouded and a slight deposit formed.

Milk. - Was not coagulated in fourteen days.

On Potato. - Slight filmy growth; almost colourless glistening surface.

Under the Microscope. -

Organism is a bacillus varying in length from little more than coccus to long forms of eight or ten microns; there is also considerable variation in thickness. Some forms show transverse division of protoplasm; some are clubbed at one or both ends not unlike involution forms of *Bacillus Diphtheriae*.

Diphtheriae.

Staining Reactions. -

Stains fairly well with ordinary stains; some forms very faintly. Does not retain Gram's stain.

BACILLUS II. - A. 14. 3. 01.

Corresponds in every way with Bacillus I. A.

25. 2. 01. (B. Mycoides.)

COCCUS I. - A. 14. 3. 01.

Plate Colony. - Large, oval; about seven by ten mm.; of bright orange colour; translucent; surface glistening; edges irregular.

Under Low Power. - Translucent; finely granular; edges irregular but clean.

On Sloped Agar. - Growth along needle-track about two mm. wide; cream coloured; surface glistening; very viscid.

Gelatine Stab. - Surface growth - orange coloured; along the needle-track considerable growth of faint orange tint. Many isolated colonies are seen along the needle-track and at deeper parts of the tube.

Gelatine liquefied to the depth of one ^{inch}~~eighth~~ in two weeks. The liquid gelatine was very turbid.

Peptone Broth. - Became very cloudy with consider-

considerable deposit.

Milk. - Was coagulated on sixth day.

On Potato. - Thin filmy growth of orange colour; viscid; surface glistening.

Under the Microscope. -

Organism is a coccus rather smaller than Staphylococcus Pyogenes Aureus; occurs singly, in pairs, threes, and groups of many. Some short chains are seen.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

COCCUS II. - A. 14. 3. 01.

Plate Colony. - Round, about three mm. diameter; almost colourless; translucent; edges irregular; viscid consistence.

Under Low Power. - Transparent; finely granular; edges irregular but clean.

On Sloped Agar. - Growth along needle-track about three mm. wide; faintly white in colour. Isolated colonies are seen; surface glistening; very viscid consistence.

Gelatine Stab. - No growth apparent after fourteen days.

days.

Peptone Broth. - Did not become cloudy;
slight white deposit formed.

Milk. - Was not coagulated in fourteen days.

On Potato. - Very slight filmy growth; colour
less; viscid consistence.

Under the Microscope. -

Organism is a coccus rather larger than one
micron, though smaller forms are seen. Is most
frequently seen in pairs: in some cases these assume
an oval shape. Is seen also in short chains and in
groups of many.

Staining Reactions. -

Stains well with ordinary stains and retains
Gram's stain.

Summary. -

Bacillus I. - A. 14.3.01.	1
Bacillus II. - A. -	1 (B. Mycoides)
Coccus I. - A. -	1
Coccus II. - A. -	1
Sarcina Lutea	2
Sarcina Pulmonum (?)	1
Coccus II. - A. G. 20.2.01.	1

EXPOSURE XIII. - Date. - 19th March 1901.

Agar and glycerine agar plates were exposed to the air in west end compartment (at Princes Street Station) of carriage No. 473, 3rd Class, Caledonian Railway, (Leith Branch).

Time of exposure. - One minute.

Conditions. - Train in motion: windows closed.

Results:- Glycerine Agar Plate.

19th March 1901. -		Incubated	
20th	- -	-	7
21st	- -	-	8
22nd	- -	-	9
23rd	- -	-	10
25th	- -	-	10

Of these, two were bacilli - of one there were two - colonies including as one a mass of many small ones; of the other, there were five colonies - one *Sarcina Lutea*, one coccus and a mould *Penicillium Glaucum*.

BACILLUS I. - G.A. 19. 3. 01.

Plate Colony. - There were many small ones uniting to form a mass about eight by fifteen mm. and one isolated colony about five mm. diameter: transparent: almost colourless: surface glistening; moist and viscid.

Under Low Power. - Finely granular; transparent; edges irregular but clean.

On Sloped Agar. - Growth faintly white covering

covering the whole agar surface and extending down between the agar and glass: growth moist and viscid: surface smooth and glistening.

Gelatine Stab. - Surface growth white; in thicker parts has a faint brownish tinge. Gelatine is cupped to the depth of a quarter inch, from sides of the cup one or two small projections of growth into the gelatine assuming the oxhorn appearance described under Agar Plate of 18th February 1901. Along the needle-track there is slight growth. Many small isolated colonies are seen especially at deeper parts, white and globular.

There is no liquid gelatine in the "cup" and no gas formation.

Peptone Broth. - Became cloudy and a considerable white deposit formed.

Milk. - Was coagulated in seven days.

On Potato. - Growth filmy, faintly white with glistening surface: viscid consistence.

Under the Microscope. -

Organism is rod-shaped with rounded ends, varying in length from, some forms little more than cocci to forms of six microns, rarely longer than this. Is thick in proportion to length, being somewhat less than one micron in diameter. Often seen in pairs,

pairs, end to end. Some single bacilli show a slight constriction as if in process of division. A few slightly curved forms are seen. The organism is motile. On examination of old culture, no spores were observed.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

BACILLUS II. - G.A. 19. 3. 01.

Plate Colony. - Of this organism there were five colonies; round, from two to five mm. diameter: opalescent in colour: translucent: at edges whiter than centrally and slightly raised: surface moist and glistening: edges regular.

Under Low Power. - Coarsely granular; translucent edges regular and clean.

On Sloped Agar. - At first opalescent, later becoming faintly brown. Growth covered the whole surface with a thin film. Surface smooth and glistening: of slightly viscid consistence.

Gelatine Stab. - Surface growth at first almost colourless; then became whitish and slightly opalescent. There is slight cupping of the gelatine and a little liquid gelatine in the "cup", which is

is lined with thick growth. Just below the cupping the growth is of brown colour extending down the needle-track for one eighth of an inch. Below this the growth is white with many small isolated globular colonies especially at the deeper part.

There is no gas formation.

Peptone Broth. - Became very cloudy with copious white deposit.

Milk. - Was coagulated on sixth day.

On Potato. - A thin brownish filmy growth with glistening surface: viscid consistence.

Under the Microscope. -

Organism is rod-shaped: non-motile, with rounded ends: varies in length from two microns to forms of ten microns or more: diameter rather less than one micron, though in the longer forms diameter is not uniform. A few short chains are seen and some curved forms. Protoplasm is often partly and occasionally entirely absent, the sheath alone staining feebly. Spore-like bodies are seen occasionally at end of bacillus.

In old cultures spores are found, almost globular, rather more than one micron in diameter.

Staining Reactions. -

Stains well with ordinary stains and retains

retains Gram's stain.

Summary. -

Bacillus	I. G.A.	19. 3. 01.	2
-	II. G.A.	-	5
Coccus	II. G.A.	21. 1. 01.	1
Sarcina Lutea			1
Penicillium Glaucum			1

AGAR PLATE. - 19. 3. 01.

19th March 1901.-	Incubated		
20th - -	-	2 Colonies	
21st - -	-	7	-
23rd - -	-	11	-
25th - -	-	11	-

Of these, two were bacilli; one coccus and two Sarcina Lutea: the remaining six were covered by the colony of Bacillus I.- A. 19. 3. 01.

BACILLUS I. - A. 19. 3. 01.

Plate Colony.- Was very large, covering fully one half of the plate: thin and transparent: faint white colour: edges raised and whiter than central part of the colony: very irregular: surface glistening and moist.

Under Low Power. - Transparent; finely granular; edges very irregular; crenated and rough.

On Sloped Agar. - Growth covered almost whole of surface in twenty-four hours except at upper part where it was about four mm. in width with irregular edges.

edges. Surface was smooth and glistening: yellowish white in colour: of slightly viscid consistence.

Gelatine Stab. - Whole surface was covered with a thin whitish growth; the upper ~~one~~ eighth inch. was very turbid. The lower side of surface growth is of a yellowish tint. Along the needle-track growth was rather slight, of white colour. Many isolated colonies are seen along needle-track and at deeper part of tube. There was no liquefaction of gelatine and no gas formation.

Peptone Broth. - Became cloudy with a slight yellowish deposit.

Milk. - Was coagulated on fifth day.

On Potato. - Slight white filmy growth with glistening surface.

Under the Microscope. -

Organism is rod-shaped; motile with rounded ends. Varies in length from two to seven microns; diameter somewhat less than one micron. Short chains are seen. Is not always uniform in thickness. In a few of the longer forms a clear spot is seen about the centre of the organism.

In old cultures curved forms are seen, some almost forming a circle.

Many spores are seen, about twice the diameter of the

the bacillus; in shape oval; about two microns in length. The bacilli have the appearance of a thick capsule; stained faintly inside which are small, elongated particles of protoplasm, deeply stained. In many of the bacilli two such particles are seen.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

BACILLUS II. - A. 19. 3. 01.

Plate Colony. - Was round, about four mm. in diameter; almost colourless; transparent with regular edges.

Under Low Power. - Faintly granular; transparent; edges irregular but clean.

On Sloped Agar. - There was slight, filmy, faintly white growth about three mm. width along the needle-track with smooth glistening surface.

Gelatine Stab. - Very slight white growth along the needle-track. A few isolated globular colonies are seen. No surface growth. No liquefaction of gelatine and no gas formation.

Peptone Broth. - Became slightly cloudy and a very slight white deposit formed.

Milk. - Was coagulated on seventh day.

day.

On Potato.- Very slight, filmy, colourless growth with glistening surface.

Under the Microscope. -

Organism is rod-shaped with rounded ends; motile. Varies in length from two microns to long filamentous forms. Some curved forms are seen and some have clubbed appearance at ends. A few short chains are present.

No spores were found.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

Summary. -

Bacillus I.- A. 19. 3. 01.	1
- II.- A. -	1
Coccus II.-G.A. 21. 1. 01.	1
Sarcina Lutea	2
Not examined (covered by Colony of Bacillus I.)	6

EXPOSURE XIV.-Date.- 26th March 1901.

Agar and glycerine agar plates were exposed to the air in the central compartment of carriage No. 1100 3rd Class N.B.R. Suburban line.

Time of exposure.- One minute.

Conditions.- Train in motion; windows closed.

Results:- Glycerine Agar Plate.

26th March 1901.	Incubated
27th - -	- 2 Colonies
28th - -	- 3 -
30th - -	- 3 -

Of these one was a bacillus, the same as Bacillus I. A.- 25. 2. 01. (B. Mycoides). Two were cocci of which one corresponded to Coccus I.G.A.- 8. 3. 01. (Staphylococcus Pyogenes Aureus), the other not previously described.

COCCUS I.G.A.- 26. 3. 01.

Plate Colony.- Round, about 2mm. diameter, white with darker brownish centre.

Under Low Power.- Opaque and granular: edges clean.

On Sloped Agar.- Copious white growth covering most of the agar surface: edges very irregular but clean: surface moist and glistening.

Gelatine Stab.- Very slight surface growth, white

white colour: along the needle track considerable growth whitish yellow in colour: many isolated colonies at the edge of track and at the deeper part of the tube. No liquefaction of gelatine and no gas formation.

Peptone Broth.- Became cloudy, with considerable white deposit.

Milk.- Was coagulated in forty eight hours. By the fourth day the clot had almost entirely disappeared leaving a clear fluid, the upper part of which was frothy as if gas had been given off.

On Potato.- Copious white growth with glistening surface.

Under the Microscope.-

Organism is a coccus about the size of Staphylococcus pyogenes aureus: occurs singly, in pairs etc., and in groups of many. A few short chains are seen.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

Summary.

Bacillus I.A.- 25. 2. 01. (B. Mycoides)	1
Coccus I.G.A.- 8. 3. 01. (Staphylococcus pyogenes aureus)	1
Coccus I.G.A.- 26. 3. 01.	1

Agar Plate.- 26. 3. 01.

26th March 1901.		incubated	
27th	-	-	5
28th	-	-	10
30th	-	-	11
1st	-	-	11

Of these one was a bacillus whose growth covered seven other colonies which were not examined. The other three colonies were cocci, one of which corresponded to Coccus I.G.A.- 26. 3. 01, the other two not previously described.

BACILLUS I. A.- 26. 3. 01.

Plate Colony.- A very large colony covering almost one half of the plate, bluish white in colour, translucent, surface glistening: slightly viscid consistence: edges irregular.

Under Low Power.- Faintly granular, edges irregular but clean: transparent.

On Sloped Agar.- Thick viscid white growth covered the whole surface of the agar: surface smooth and glistening.

Gelatine Stab.- Along the needle track copious white growth: edges beaded by many isolated colonies globular. On surface growth is considerable,

considerable, raised up and rounded, the general appearance being that of a nail with rounded head. Two gas bubbles appeared along the line of growth.

On Sloped Gelatine.- Copious white growth, moist and raised.

Lactose Gelatine.- Copious white growth similar to that in plain gelatine: some gas bubbles formed. There was no liquefaction of gelatine.

Peptone Broth.- Rapidly became cloudy with a dense whitish deposit.

Milk.- Was coagulated in forty eight hours.

On Potato.- Copious whitish growth with glistening surface: viscid.

Under the Microscope.-

Organism is rod shaped, non motile, with rounded ends. Many forms little more than cocci are seen and others of about three and four microns.

Staining Reactions.-

Stains well with ordinary stains but does not retain Gram's stain.

From microscopic appearance and general manner of growth especially the nail-like growth in gelatine and the fact that it does not retain Gram's stain this organism seems to correspond to Friedlander's Pneumobacillus.

bacillus.

COCCUS I. A.- 26. 3. 01.

Plate Colony.- Was round, about 3mm. diameter, white colour with glistening surface: viscid.

Under Low Power.- Opaque and granular: edges clean.

On Sloped Agar.- Considerable white growth along the needle track, about 4mm. in breadth: very viscid: surface glistening and smooth, edges irregular but clean.

Gelatine Stab.- Surface was slightly cupped but no liquid gelatine present: considerable white growth along the needle track and many isolated colonies are seen especially at the deeper part. No gas formation.

Peptone Broth.- Became cloudy with slight white deposit.

Milk.- Was coagulated on the sixth day.

On Potato.- Considerable growth, faintly white in colour, with glistening surface: viscid.

Under the Microscope.-

Organism is a coccus about the size of Streptococcus pyogenes occurs singly, in pairs, threes etc., in groups and in short chains. In many cases a

a faint halo-like appearance as of a capsule is seen surrounding single cocci, also groups of three and four and in the case of chains of three or four. It does not stain.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

COCCUS II. A.- 26. 3. 01.

Plate Colony.- Small, about 1mm. diameter, white with brownish centre.

Under Low Power.- Granular and opaque with clean edges.

On Sloped Agar.- Many small white isolated colonies are seen outside the needle track along which the growth is about 3mm. in width: glistening surface.

Gelatine Stab.- Surface growth was slight and of white colour: along the needle track considerable growth of the same colour: has beaded appearance owing to numerous isolated colonies at edges of the growth. There is no liquefaction of gelatine and no gas formation.

Peptone Broth.- Not cloudy but slight white deposit formed.

Milk.- Was not coagulated in fourteen days.

days.

On Potato.- A thin filmy colourless growth with glistening surface.

Under the Microscope.-

Organism is a coccus, rather less in size than *Staphylococcus pyogenes aureus*, occurs singly, in pairs, threes etc., in groups of many and a few short chains are seen.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

Summary.

Bacillus I. A.- 26. 3. 01.	1
Coccus I.G.A.-26. 3. 01.	1
Coccus I. A.- 26. 3. 01.	1
Coccus II. A.- 26. 3. 01.	1
Not examined-covered by Colony of Bacillus I. A.-	7

EXPOSURE XV. - Date. - April 4th 1901.

Agar and glycerine agar plates were exposed to the air in the central compartment of carriage No. 300 Caledonian Railway 3rd Class (Leith Branch).

Time of exposure. - Two minutes.

Conditions. - Train in motion: windows closed.

Results:- Glycerine Agar Plate.

4th April 1901	incubated		
5th	-	-	7 Colonies
6th	-	Not incubated	10 -
8th	-	-	15 -

Of these two were bacilli, two cocci, one Sarcina white colony, and one mould which covered seven colonies. These were not examined. One of the cocci corresponded with Coccus I. - A. - 25. 2. 01: Micrococcus Tetragonus. Of the other coccus there were three colonies.

BACILLUS I. - G.A. - 4. 4. 01.

Plate Colony. - Was irregularly round, about six mm. diameter, yellowish white in colour, viscid: surface glistening.

Under Low Power. - Translucent, faintly granular, edges irregular but clean.

On Sloped Agar. - Profuse yellowish white growth covering almost the whole of the agar surface, moist

moist and viscid, surface smooth and glistening.

Gelatine Stab. - Considerable white growth along the needle track followed by rapid liquefaction of gelatine affecting the whole diameter of the tube. At lower part of needle track many isolated colonies are seen.

There was no surface film and no formation of gas.

Peptone Broth. - Became cloudy, with slight brownish deposit, a fawn coloured surface film wrinkled and dry.

Milk. - Was coagulated on the fourth day, the whey separated leaving a syrupy clot.

On Potato. - Copious fawn coloured dry growth covered the potato: surface of growth dull and wrinkled.

Under the Microscope. -

Organism is rod shaped, thick, with rounded ends, slightly motile, varies in length from some little more than cocci to long forms of about seven microns: often seen in pairs. Occasionally a faint halo-like appearance is seen around the organisms. In old cultures many spores are seen, in shape elongated oval about two microns in length and one in width.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

stain.

BACILLUS II. - G.A. - 4. 4. 01.

Plate Colony. - Was round, about three mm. in diameter, brownish white in colour, with dark brown centre.

Under Low Power. - Opaque, granular, edges clean and regular.

On Sloped Agar. - Growth covered the whole of agar surface, thin and filmy with faint orange tint, at the bottom of the tube a distinct orange coloured deposit formed, surface of growth was smooth and glistening, slightly viscid.

Gelatine Stab. - Slight white growth along the needle track, with many isolated colonies at the deeper part. No liquefaction of gelatine and no gas formation.

Peptone Broth. - Not cloudy, considerable flocculent deposit of faint orange tint.

Milk. - Was coagulated on sixth day.

On Potato. - Slight orange tinted growth with glistening surface, slightly viscid.

Under the Microscope. -

Organism is short thick rod shaped, with rounded ends: is non-motile. Some coccal forms are seen,

seen, and some little more than cocci but having one diameter distinctly longer than the other, often seen in pairs. Occasionally a halo-like appearance is seen surrounding the organisms.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

COCCUS I. - G.A. - 4. 4. 01.

Plate Colony. - There were three, in diameter three to five mm., round, light brown in colour.

Under Low Power. - Opaque and granular, edges clean.

On Sloped Agar. - Growth was yellowish white, with glistening surface, slightly viscid, covering almost the whole agar surface.

Gelatine Stab. - Slight surface growth, yellowish white and dull. Along the needle track considerable growth with very many isolated globular colonies at edges and at deeper part. There is no liquefaction of gelatine and no gas formation.

Peptone Broth. - Became cloudy and a slight yellowish white deposit formed.

Milk. - Was coagulated on the fifth day.

On Potato. - Considerable yellowish white growth with glistening surface.

surface.

Under the Microscope. -

Organism is a coccus about the size of Staphylococcus pyogenes aureus. Occurs singly, in pairs, threes, etc., and in groups of many.

Staining Reactions. -

Stains well with ordinary stains and retains Gram's stain.

THE MOULD. - G.A. - 4. 4. 01.

Plate Colony. - Was fluffy and white covering the greater part of the plate: mycelium was colourless and very much branched, sporangia round.

Gelatine Stab. - Surface growth similar to that of the agar plate colony. Along the needle track, colourless spikes radiated on all sides, straight into the medium. There was no liquefaction of gelatine.

Appearance under the microscope resembled Mucor Mucedo but the fact that the gelatine is not liquefied differentiates it from that mould.

Summary.

Bacillus	I. - G.A. - 4. 4. 01.	1
Bacillus	II. - G.A. - 4. 4. 01.	1
Coccus	I. - G.A. - 4. 4. 01.	3
Coccus	I. - A. - 25. 2. 01. (M. Tetragonus)	1
Sarcina (white colony)		1
Mould		1
Covered by mould and not examined.		7

AGAR PLATE.- 4. 4. 01.

4th April 1901.-		Incubated	
5th	-	-	12 Colonies
6th	-	-	20 -
8th	-	-	24 -
10th	-	-	24 -

Of these, three were bacilli, two having one colony each; the other three. Three were cocci, two having three colonies each; the other, two; all already described. Five were *Sarcina Lutea*; two *Sarcinae* with white colonies; two *Torula Rosacea* and two were covered by the colony of *Bacillus III*.

BACILLUS I.- A. 4. 4. 01.

Plate Colonies.- There were three; three and four mm. in diameter: whitish: translucent: viscid: edges irregular.

Under Low Power.- Faintly granular: translucent: edges very irregular.

On Sloped Agar.- Considerable growth with many isolated colonies covering most of the agar surface: white: in young culture very viscid, less so in old. Surface is glistening.

Gelatine Stab.- Considerable white growth along the needle-track with very many isolated colonies at deeper part. The gelatine became cupped the second day after inoculation and cupping proceeded along the

the track, till at the end of a week, it was an inch in depth. At surface the cup was one quarter inch in diameter; lined with white growth, which at the lower end, assumed a spiral appearance.

There was no liquefaction of gelatine and no gas formation in the medium.

Peptone Broth.- Became cloudy and a slight white deposit formed.

Milk.- Was coagulated on sixth day.

On Potato.- Growth covered whole surface, white and glistening.

Under the Microscope.-

Organism is rod-shaped; freely motile, with rounded ends. In length varies from two to five microns. Is seen occasionally in chains and a few filamentous forms are seen.

In old cultures many forms stain very slightly as if they were sheaths of bacilli without protoplasm. No spores are seen.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

BACILLUS II.- A. 4. 4. 01.

Plate Colony.- Was irregularly rounded, about

about eight mm. in diameter; white; translucent; edges very irregular; viscid.

Under Low Power.- Granular; translucent; edges very irregular with many fibrils spreading over the medium.

On Sloped Agar.- After twenty-four hours, incubation. The whole surface was covered with dull, white growth: surface, hard, dry and wrinkled.

Gelatine Stab.- Slight surface growth of faint white colour; slight white growth along the needle-track. Isolated colonies are seen at the deeper part.

There is no liquefaction of gelatine and no gas formation.

Peptone Broth.- Became cloudy and a dense, brownish deposit formed: a buff coloured film covered whole surface, adhering slightly to sides of the tube.

Milk.- Was coagulated on the eighth day: a surface film, faint buff coloured formed on the whey.

On Potato.- Was completely covered with dry buff coloured growth: very wrinkled.

Under the Microscope.-

Organism is rod-shaped, with rounded ends. Is non-motile. Varies in length from two to seven microns. Slightly curved forms are seen. Occasion-

Occasionally a halo-like appearance surround individual bacilli. Shows tendency to clumping.

In old cultures, many spores are seen; elongated ovals almost two microns in length and one in width. Some coccal forms are seen; frequently in pairs with halo-like appearance round both. Some of the bacilli are clear at one end as if spore was in process of formation.

Staining Reactions.-

Stains well with ordinary stains but does not retain Gram's stain.

BACILLUS III.- A. 4. 4. 01.

Plate Colony.- Agar. An irregularly rounded colony about ten mm. diameter: of faint white colour: transparent; edges very irregular: looped. Is extremely viscid.

Under Low Power.- Transparent: surface has appearance of being covered with fine lines: striated: edges are very irregular: loops and fine fibrils growing out over the plate.

Plate Colony.- Gelatine. On this medium there was very little growth in two weeks. Colony was almost colourless: transparent: edges irregular.

Under Low Power.- Faintly granular: transparent

transparent. Edges irregular but no loops nor fibrils are seen.

On Sloped Agar.- Along the needle-track there was considerable growth: almost colourless: transparent: edges very irregular, fibrils and loops being well seen with low power objective. Growth is very viscid and has glistening surface.

On Sloped Glycerine Agar.- Growth is more profuse: isolated colonies covering almost all the agar surface. Colour is more distinctly white. Loops and fibrils are well seen with low power objective.

Gelatine Stab.- In seven and a half per cent gelatine growth in two weeks was very slight, a faint white line being visible along the needle-track.

With low power objective, there seems to be slight spiking. The gelatine is slightly cupped on the surface but no liquid gelatine is present.

In ten per cent gelatine growth is hardly noticeable, there being very little change in appearance three weeks after inoculation. Slight cupping of surface is again present. When ten per cent gelatine was incubated, growth was again slight, small almost colourless masses of growth appearing forming a slight deposit.

On Sloped Gelatine.- Growth very slight and very

very little change in appearance in three weeks.

Peptone Broth.- Became slightly cloudy with slight whitish deposit.

Milk.- Was coagulated in four days.

On Potato.- No growth was visible after several inoculations.

Under the Microscope.-

Organism is rod-shaped with rounded ends. Is non motile. Varies in length from two to ten microns. Short chains are seen and occasionally long filaments. In impression preparations, the longer organisms, chains and filaments are better seen: at edges of colonies, are seen in chains lying side by side and chains are seen running out from the edge; in some cases turning back again to join the colony. In older cultures, many clear spots are seen in the organisms which do not stain well. When film is stained with Carbol-Fuchsin and Methylene Blue, the organisms stain very unevenly - the fuchsin staining the protoplasm in isolated masses: between these and on either side of the fuchsin stained masses, the Methylene Blue staining; the clear spore-like spots remaining unstained.

Staining Reactions.-

Stains well with ordinary stains and retains

retains Gram's stain.

TORULA ROSACEA.

Plate Colony.- Of this organism there were two colonies, about two and four mm. diameter: round: pink tinted: surface glistening.

Under Low Power.- Translucent: coarsely granular: edges crenated. At edges the individual organisms are seen.

On Sloped Agar.- Along the needle-track, copious raised growth of pink colour with glistening surface: at sides of needle-track, a thin filmy growth covered the remainder of the agar surface: at the bottom of the tube there was a thick, pink deposit.

Gelatine Stab.- Surface growth pink and glistening. Along the needle-track, copious white growth. Many isolated globular colonies are seen at the edge of the track and at deeper part.

Peptone Broth.- Became slightly cloudy with a dense pink deposit.

Milk.- Was not coagulated in fourteen days: a dense pink deposit formed at bottom of the tube.

On Potato.- Copious pink growth with glistening surface.

Under the Microscope.-

Microscope.-

Large oval cells up to about ten microns in length. Budding is seen in many cases. In centre of cells, granules are seen. Some very large cells do not stain as well as the others.

Staining Reactions.-

Stains well with ordinary stains and retains Gram's stain.

Summary.-

BACILLUS	I.- A. 4. 4. 01	3
-	II.- A. -	1
-	III.- A. -	1
Torula Rosacea		2
Coccus	II.- A. 21. 1. 01.	3
-	I.-G.A. 1. 2. 01.	2
-	I.-G.A. 4. 4. 01.	3
Sarcina Lutea		5
-	(white colony)	2
Covered by colony of Bacillus	III.	2

The total number of micro-organisms collected was 332, of which 289 were examined, the remaining 43 having been covered by the growth of moulds or of other organisms.

Sarcina Lutea was the most common individual organism, 88 colonies having been found. It was present in every collection, being absent in only four of the thirty plates; all four plates being Glycerine Agar. Five colonies of *Sarcina Pulmorum* (or *Ventriculi*) were found.

Cocci were found in 140 cases, all examined, a notable feature being the absence of streptococci.

Forty-five colonies were bacilli, of which eight were spore producing.

Seven were moulds and one *torula* (*Rosacea*) was found.

I had been led to understand that time would not permit the procuring of a license for inoculation experiments on animals, so that evidence as to the pathogenic nature of the bacteria is incomplete; but, so far as the appearance of Agar, Gelatine, Peptone Broth, Milk and Potato cultures, and of the organisms under the microscope, holds good, I have isolated four pathogenic organisms:-

organisms:-

Bacillus Coli Communis

Friedlander's Pneumo-bacillus

Staphylococcus Pyogenes Aureus

Micrococcus Tetragonus

I had fully expected to find the Tubercle Bacillus and have been agreeably disappointed in not doing so; but, in one or two railway carriages, I have seen sputum which looked suspiciously like that of a phthisical person.

Having thus demonstrated the presence of many micro-organisms in the railway carriages, the question arises as to what means are to be used to prevent their further entrance and to get rid of those already present.

Greater cleanliness can be obtained by more frequent and better cleaning. So far as I have observed, cushions which are removeable are sometimes taken out of the carriage and beaten outside, but very frequently the beating is done in the carriage, which is then brushed out and the woodwork and glass rubbed with a damp cloth, this does to some extent remove the dust but some of it must necessarily settle again on the cushions and on the luggage racks and then set

set moving when the train is in motion, by draughts and by movements of the passengers.

The abolition of all cloth window curtains and the substitution of leather for the plush and other rough material at present in use for covering seats and back cushions, would furnish less favourable lodgement for dust and organisms and greater facility for cleaning, but I very much doubt if such measures would find favour with the travelling public and certainly not with the railway companies, on the ground of expense alone.

Disinfectants in the form of spray might be used but Firth (21) mentions that Thymol solution had been used in this way on the matting in New York tram-cars but without appreciable effect, numerous organisms being found on the fibres of the matting.

Carbolic Acid to be effective would need to be of such strength that many would object to its use on account of the odour and many of the proprietary disinfectants at present on the market would have the same objection.

Formalin, according to its makers, would be the ideal germicide but there is doubt as to its efficacy unless used in solution of considerable strength and then the piquant odour might be objectionable.

objectionable.

To sum up:- Greater cleanliness is absolutely necessary and can only be secured by more frequent and thorough cleaning; the removal of all cushions and curtains to the open air for beating; occasional sterilization of these by hot dry air; the washing of windows and all woodwork with a suitable and reliable disinfectant; and, if possible, the substitution of leather for cloth fabrics, as coverings for seats and cushions.

Robert Nichol. Dobner.
M.B. Ch.B.