

Protection from microbial contamination in a room ventilated by a uni-directional air flow

O. M. LIDWELL AND A. G. TOWERS*

Cross-Infection Reference Laboratory, Colindale Avenue, London, N.W. 9

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There are a number of medical situations in which the protection of a patient from infection is of particular importance. For example, patients receiving cytotoxic drugs, suffering from extensive burns or being nursed in intensive care units after serious operations or major injury are unusually likely both to become infected and to suffer from the more serious clinical consequences of infection. The absolute and relative importance of the many possible sources and routes of infection for these patients is largely unknown and certainly varies for different categories of patients and in different environments. Total isolation from all extraneous sources of micro-organisms has been attempted by enclosing the patient in a plastic tent, carrying out all manipulations through glove ports, and using double-ended hatches for passing in sterile materials and removing wastes. The extensive use of systems of this kind lead to substantial nursing difficulties and to psychological problems for some patients. (Levitan *et al.* 1968).

In so far as the infective agent is airborne and is either inhaled by the patient or settles on a sensitive area, the risks of infection can be reduced by ventilation. The traditional methods of ventilating a space produce or permit sufficient turbulent circulation of the air to distribute any contamination dispersed into the air relatively evenly throughout the room and subsequent elimination follows a logarithmic course, i.e. successive fractional reductions in the contamination level take equal times and absolute removal theoretically requires infinite time. Since sedimentation is unaffected by the ventilation process, increases in the amount of ventilation produce a less-than-proportionate increase in the rate of clearance. To take a simple example, if the dispersed particles have a sedimentation rate of 1 ft./min.—a common average value for naturally dispersed airborne particles carrying bacteria—the time for the initial numbers to fall to one-tenth will be 23 min. in an unventilated room 10 ft. high, 12 min. if ventilation is provided at 6 air changes/hr. and about 5 min. if this is increased to 20 air changes/hr. If, however, the ventilating air could be induced to move across the space in one direction only, without any turbulent mixing, complete removal would be attained in the time taken for the air to traverse the space once. Attempts to apply this principle to the ventilation of surgical operating rooms with the limited volumes of ventilating air considered practicable proved to be ineffective, since the translational velocities that could have been produced by the introduced air, between 2 and 5 ft./min., were smaller than the turbulent velocities produced by thermal

* Working on a Ministry of Health Research Grant.

effects and the movement of persons (Lidwell, Richards & Polakoff, 1967). If however, the ventilating air volumes can be increased by a factor of 10 or more the effect of these disturbances is largely eliminated.

Industrial requirements for dust-free spaces, stimulated by the demands of the U.S. space programme, have led to the development in recent years of directed-flow ventilation systems employing both vertical and horizontal air flow in the range 50–100 ft./min. These are often described as 'laminar flow' systems, but as they do not fulfil the aerodynamic conditions for genuine laminar flow it seems desirable to avoid using this term as far as possible. The turbulent air movements in such systems are, however, sufficiently small for practically complete removal of air-dispersed particulates to be obtained with a single air change. The disturbance of the air flow by objects introduced into the room does not significantly detract from their performance in this respect so long as the dimensions of these objects are small in relation to those of the room. By nursing patients in a room ventilated in this way it should be possible practically to eliminate the risk of airborne infection. In addition to the possible clinical benefit that this might bring, such an environment would greatly facilitate the study of other routes of infection. If the potentially ubiquitous airborne route could be eliminated, details of the paths of contact transfer in a nursing situation could be more easily evaluated. With a view to exploring these possibilities, the Ministry of Health in 1967 acquired a clean room, based on an industrial model, with horizontally directed air flow.

We report here the results of a preliminary investigation carried out in this room using artificially generated bacteria-carrying airborne particles and exploring the dispersal of these with different air-flow velocities and with various degrees of active movement in the room. We were able to introduce very large numbers of particles at any one point, usually about 30 million in a single experiment. It was then possible to detect as few as 1 in 10^7 of the dispersed particles reaching a square foot of horizontal surface exposed anywhere within the room.

METHODS

Description of the room and ventilating system

A diagram of the room in section is given in Fig. 1, together with a schematic representation of the ventilating and air conditioning plant. The floor, covered with vinyl sheet, was 16 ft. 8 in. long by 10 ft. wide. The room was 8 ft. 2 in. high and one end was entirely composed of a bank of filters through which the ventilating air entered at a uniform velocity over the whole wall area. This velocity could be adjusted up to 100 ft./min. by controlling the recirculating fans. The air was extracted from the opposite end through a series of louvres. These covered the entire wall surface and could be adjusted so that the outflow velocity was also uniform over the whole area. The two long walls were covered with Formica, except for the windows, and the ceiling, below the lights, was formed by double-walled panels covered with thin polyvinyl sheet. Tests with smoke generated from a stick dipped in titanium tetrachloride and with a vane anemometer showed that, at air velocities of 60 or 100 ft./min. air flow was nearly horizontal and at a con-

stant velocity throughout the room. At a velocity of 35 ft./min., however, when the refrigeration unit was in operation, uneven temperature distribution in the incoming air resulted in significant departures of the air flow lines from the horizontal. At 20–25 ft./min. these disturbances were much more marked and back flow could be detected in some regions. All tests done at these low velocities were therefore carried out with the refrigeration unit switched off. Refrigeration is normally necessary in a room of this type in order to remove the heat generated by the recirculating fans. The divergence of the smoke tracks showed that under all conditions small-scale turbulence was present, but the included angle of the visible smoke trail, averaged over some minutes, was not normally as much as 10° .

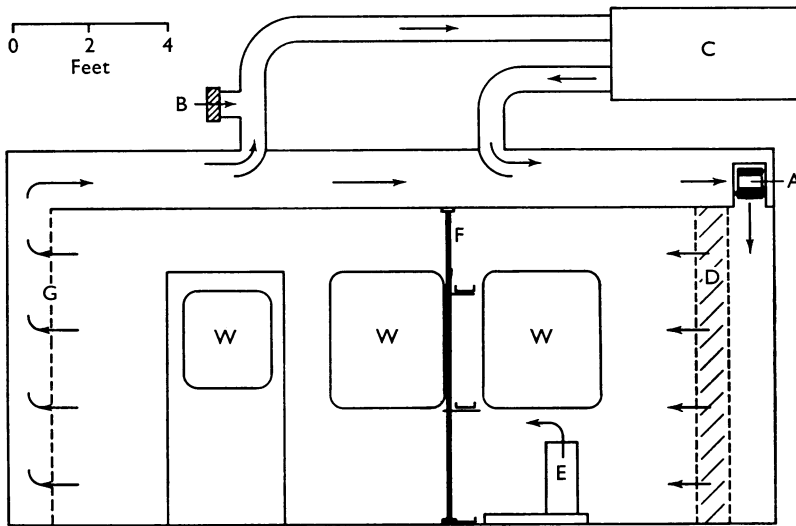


Fig. 1. Section of room. A, Recirculating fan; B, make-up air inlet; C, air-conditioning unit (temperature and humidity control, including a refrigeration unit); D, filter bank (prefilter followed by high efficiency filter); E, airborne particle generator; F, stand with sampling plates; G, return grille.

Production of airborne bacteria-carrying particles

In order to simplify calculations involving both the numbers of particles in a given volume of air and the numbers settling into a specified area, it is convenient to use particles of uniform size. Studies in a variety of environments have shown that a common value for the median diameter of bacteria-carrying particles in occupied environments is around 13μ , assuming unit density for the particles, or a settling rate in air under normal ambient conditions of about 1 ft./min. (Noble, Lidwell & Kingston, 1963). The most satisfactory method of producing such particles is the air-driven spinning top (May, 1949). If this is operated at an air pressure of about 7 lb./sq.in., the primary droplet produced is of the order of 40μ in diameter. When spraying a fluid such as nutrient broth with a solids content of some 2.5%, the diameter of the resulting 'dried' particle will be rather over 10μ . At 50% relative humidity drying is not complete and the particles resulting from spraying a broth suspension of *Bacillus subtilis* spores had an average settling rate

in air of about 1.1 ft./min., or an estimated diameter, assuming that the particles were of unit density, of 13.5 μ .

To avoid uncertainties, due to the break-up of particles in a fluid medium, the density of the spore suspension was selected so as to give only a small probability that any sprayed particle would carry more than a single spore. The spore suspension used was prepared by growing *B. subtilis* var *niger* N.C.T.C. 10073, Detrick strain, according to the method of Beeby & Whitehouse (1965). The viable count of the stock suspension in distilled water was estimated by depositing with a calibrated pipette drops of suitable dilutions on nutrient agar plates. Characteristic pigmented colonies are formed after overnight incubation at 37° C. The suspension was stored in the refrigerator at 4° C. and diluted with nutrient broth

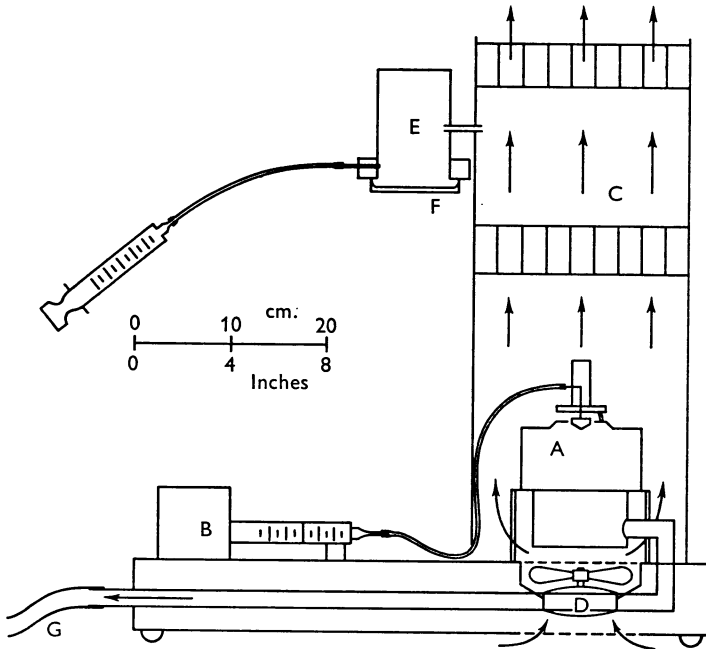


Fig. 2. Airborne particle generator. A, Spinning top (May, 1949); B, mechanically driven syringe for feeding suspension; C, vertical wind-tunnel with baffles to reduce turbulence; D, fan; E, sedimentation chamber for sampling dispersal cloud; F, sampling plate for cloud; G, tube to lead effluent air from spinning top, with satellite particles, out of room.

before each test-run to a final concentration of 6×10^6 colony-forming units per ml. Since a 40 μ diameter droplet has a volume of approximately 3×10^{-8} ml. the probability that any droplet will contain a viable spore is approximately 17%, and less than 10% of the infected droplets would be expected to carry more than one viable spore.

Dispersal of the bacteria-carrying particles

In order to introduce the particles into the room at a determined point they were generated at the base of a short vertical wind-tunnel. The diameter of this was made as small as possible consistent with avoiding impingement of the sprayed

droplets onto the walls, and air was introduced at the base at a sufficient rate to ensure an adequate air flow up the tunnel to carry the dried droplets with it and discharge them from the upper end of the tube. The arrangement used is shown in Fig. 2. The diameter of the tunnel was 9 in. (23 cm.) and air was blown in at 210 l./min. The compressed air supply to the rotor at 7 lb./sq.in. (0.46 kg./cm.²) was equivalent to 30 l./min. free air and the induced efflux from the apparatus was 70 l./min. The net flow up the tunnel was therefore approximately 170 l./min., giving a discharge velocity of 7 cm./sec. or about 14 ft./min. At this velocity the discharge did not significantly affect the air-flow lines in the room, and the effects on these of the apparatus itself were small. The spore suspension was fed on to the centre of the spinning disc at a constant rate, 0.35 c.c./min., by a mechanically

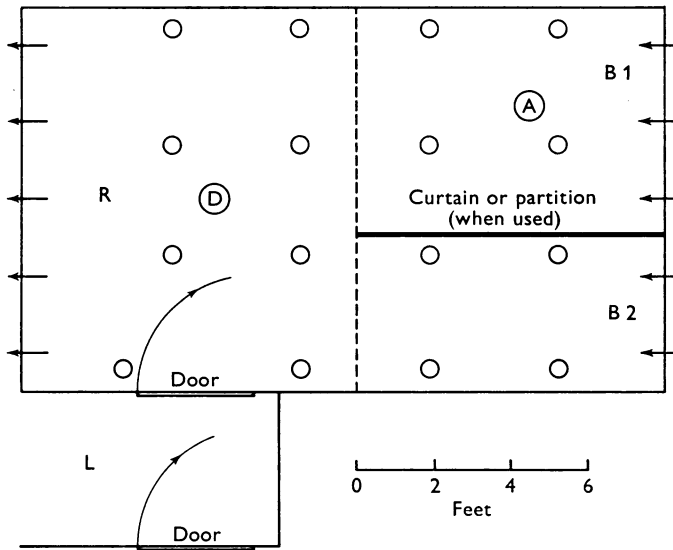


Fig. 3. Plan of room. B 1, B 2, Notional bed areas; R, rear area of room downwind of bed areas. A, dispersal point in B 1; D, dispersal point at rear of room. The unlabelled circles mark the position of the sixteen stands each carrying up to three 5½ in. diameter Petri dishes (F in Fig. 1).

driven syringe. As a check on the rate of dispersal of the spore particles, two 10 c.c. portions of the particle-laden air were withdrawn into the sedimentation chamber shown in the figure shortly after the start and shortly before the end of each period of spraying. The median number of colonies found on the settling plates from 35 tests was 186. This corresponds to a dispersal rate of $(186 \times 170 \times 10^3)/20 = 1.6 \times 10^6$ spore-bearing particles per minute, compared with the delivery rate of $0.35 \times 6 \times 10^6 = 2.1 \times 10^6$ viable spores per minute.

Sampling method

The use of a homogeneous bacterial cloud leads to an exact equivalence between sampling by volumetric or settling methods whatever the age of the cloud. In these circumstances, the exposure of open plates containing culture medium allows

samples to be obtained simultaneously from many positions in the room without disturbing the air-flow pattern by introducing large amounts of apparatus.

Petri dishes, $5\frac{1}{2}$ in. in diameter, containing nutrient agar were exposed at sixteen points disposed in a uniform grid over the plan of the room (see Fig. 3). Three plates were exposed at each point, one 6 ft. above the floor, one 3 ft. above the floor and one on the floor itself. The upper two of each set of three plates were supported on small horizontal platforms made of aluminium sheet attached to $\frac{1}{2}$ in. diameter vertical rods. The plates were uncovered, starting from the upstream end of the room at the beginning of each experiment, and normally covered again in the reverse order 5 min. after the conclusion of the spraying period. In order to check the rate of contamination due to manipulation, a total of 133 plates were exposed and recovered at various times during the course of the experiments without any spraying of spores. In all, fifty-three colonies of pigmented subtilis were found on twenty-seven of the plates after incubation. The expected rate of accidental contamination during the experiments was therefore 0.40 colonies per exposed plate.

The exposed surface area of each plate was approximately $\frac{1}{8}$ sq.ft., so that the total sampling area was 8 sq.ft. or about one twentieth of the total floor area of the room.

Experimental procedure

The room was cleaned initially and between each group of two or three trials by swabbing the floor with detergent solution, the walls and plate-carriers were similarly treated after any major rearrangement of equipment within the room. The two people who entered the room wore clean laboratory coats and plastic overshoes but took no other special precautions against contamination. Trials were carried out at four air-speeds measured by a vane-type anemometer across the central transverse plane of the room. The speeds used were 100, 60, 35 and 22 ft./min., and variations in time and space across this plane did not exceed $\pm 20\%$.

In addition, comparative measurements were made by means of two systems that produced highly turbulent conditions of air movement in the room. The first was provided by a small fan unit placed in the centre of the room 7 ft. above the floor which recirculated the air through a high-efficiency filter at 130 cu.ft./min., equivalent to an air change rate of 6/hr. The recirculating fans were switched off when this system was in use, and the return grilles were blanked off with polythene sheet. The second was obtained from a portion of the recirculating plant: 1600 cu.ft. of air/min. were delivered into the room through a hardboard funnel arranged to produce a highly turbulent air-flow pattern in the room without excessive air velocities below the 6ft. 6 in. level. The return grilles were mostly blanked with polythene sheet, leaving two outflow apertures about 2×2 ft. each at the bottom corners. This rate of air flow corresponds to an air change rate of 75/hr.

As the rate of clearance of the air-borne spores was much slower with these systems than with the directed air flow, the period of spraying was reduced from the usual 20 min. to 1 or 2 min. only, and a substantial period (20 min. and

10 min. respectively) was allowed after the conclusion of spraying before the exposed plates were re-covered.

In addition to varying the ventilation arrangements, two positions were used for dispersal of the particles, and a curtain or a solid partition was used to divide up the interior of the room during some experiments. Figure 3 shows the position of these.

During the entire period of exposure of the plates, the two people in the room moved about according to one or other of four standardized patterns. These were: (1) moving about continuously in the rear half of the room only, (2) moving about continuously in this area and in the left-hand front quarter of the room, (3) moving about continuously over the whole of the room area, and (4) sitting quietly throughout at the end of the room by the return grilles. The movement consisted of a steady walk at about 2 m.p.h. with frequent turns and reverses on moving in and out of the various areas delimited by the position of the vertical rods and other equipment.

Expression of the results

The underlying intention of these experiments was to investigate the extent to which two or more patients in bed in such a room would be effectively isolated from each other in spite of the disturbance introduced by the movements of nurses and others in the room. For this purpose, the beds would be placed with their head ends close to the inlet filter wall and their length parallel to the direction of air flow (see Fig. 3). The room was therefore considered as three areas: B 1 and B 2, the two quarters curtaining the beds; and R, the rear half of the room down-wind from these two. Dispersal took place either in the quarter B 1 at point A or in the rear half R at point D, and the contamination reaching the other bed area B 2 or both the bed areas B 1 and B 2 when dispersal took place in the rear half of the room was assessed by adding together the colonies found on all the twelve plates exposed in the relevant bed area. The number of spore-bearing particles discharged in a 20 min. test was $1.6 \times 20 \times 10^6 = 3.2 \times 10^7$ and the aggregate area of the twelve plates was 2 sq.ft. The total number collected represents therefore the average contamination within the area per square foot of surface for a dispersion of 6.4×10^7 particles, and the results have been converted in the tables to the contamination per square foot per 10^8 particles dispersed. Since the particle-settling rate approximated to 1 ft./min., this is closely equivalent to the volumetric (e.g. inhaled) contamination rate per 10^8 particles dispersed per cubic foot of air per minute sampled or inhaled.

It is of interest to compare the results with those that would be expected if the ventilating air had been supplied at the same rate in such a way that the air was always completely mixed by turbulent motion in place of the directed air flow system. At the higher rates of air flow this would, of course, need intolerably high turbulent air velocities. Under these postulated conditions the rate of removal of dispersed particles would be exponential, and the total number that would settle on any square foot of horizontal exposed surface would be $(60NS)/VK$, where N is the total number of particles dispersed, S is the settling rate of the particles in feet

per minute, V is the volume of the room in cubic feet and K is the exponential die-away constant in hr.^{-1} .

K includes the effects of both ventilation and settling and is given by $60(v + SA)/V$, where v is the volume of ventilating air in cubic feet per minute and A is the total area of exposed horizontal surface in square feet. For the room and particles used in these experiments, $S = 1.1 \text{ ft./min.}$, $V = 1360 \text{ cu.ft.}$ and $A = 167 \text{ sq.ft.}$ Hence $K = 0.044v + 8$ and the calculated settling is

$$0.049N(0.044v + 8) \text{ per sq.ft.}$$

RESULTS

Table 1 shows the results of the forty-eight individual experiments. The extent of transfer of air-dispersed contamination into the 'sensitive' area has been determined, as described in the previous section, as the number of particles settling per square foot of surface for 10^8 particles dispersed. The 'sensitive' area is defined as either both bed areas (B1 and B2), when dispersal took place at the rear of the room, or as the other bed area, B2, when dispersal took place in bed area B1.

Table 1. *Numbers of spore bearing particles settling per square foot of exposed surface in the 'sensitive' area for 10^8 particles dispersed: equivalent particle diameter 13μ*

	Ventilation conditions					
	(a)	(b)	(c)	(d)	(e)	(f)
Linear air-flow velocity (ft./min.)	100	60	35	22	Turbulent	
Rate of air supply (cu.ft./min.)	8200	4900	2900	1800	1600	130
Ventilation rate/hr.	360	220	130	80	75	6
	Number of particles settling					
Calculated settling for turbulent ventilation	1.32×10^4	2.18×10^4	3.63×10^4	5.57×10^4	5.90×10^4	35×10^4
Group	Experimental conditions					
1	D0-P, -C or -U	—	3, 10	—	—	—
	A0-P, -C or -U	—	6, 0	25	0	4.0×10^4
	D1-P, -C or -U	5, 3, 5, 10	—	13, 12	12, 207	3.2×10^4
	D2-P	—	5	—	—	—
	D2-C	2	8	—	—	—
	A1-P	2	2	—	—	—
	A2-P	7	0	10	2	—
2	D3-P, -C or -U	40, 90	—	255, 146, 104	590, 287, 500	—
		87, 65	—	222, 62, 85	900, 845, 557	—
	A2-C	100	47	57	600	—
	A3-P	149	—	119	445	—
3	A2-U	202	—	650	7200	—
	A3-U	162	380	1230, 1380	4150	4.1×10^4

The experimental conditions have been coded as follows. D, Dispersal at the rear of the room, at point D; A, dispersal in one bed area, B1, at point A (at the front of the room). 0, No movement during dispersal; 1, movement in the rear half of the room only; 2, movement in the rear half of the room and into the non-sensitive bed area only; 3, movement over the whole area of the room; P, a solid partition separated the two bed areas; C, bed areas were separated by a curtain; U, Communication between the two bed areas was unobstructed.

When dispersal was at point D the 'sensitive' area comprised both bed areas, B1 and B2. When dispersal took place at point A, only the area B2 formed the 'sensitive' area. The contamination level as determined in control exposures without dispersal was equivalent to eight particles settling in any test.

The numbers of spore-bearing particles recovered during the experiments with the two turbulent ventilating systems correspond reasonably closely to those calculated. With the directed flow system they are always much lower than this even at the lowest rate of air flow. The results obtained with the directed air-flow system fall into three clearly distinguishable groups according to the position of dispersal, extent of movement in the room during the particle dispersal and the presence or absence of the curtain or partition.

The first group comprises all those experiments with no movement of persons, those where dispersal took place at the rear of the room and movement was confined to the rear half of the room or movement extended into one of the bed areas only and this area was separated from the sensitive area either by a curtain or by a solid partition, together with those where dispersal took place in one bed area and movement was confined to the rear of the room or extended into the dispersal area and this area was separated from the sensitive area by a solid partition. With the exception of one experiment at the lowest air velocity (22 ft./min.) the numbers recovered do not significantly exceed the expected accidental contamination level due to manipulation obtained in control experiments; that is, contamination by settling does not exceed 10/sq.ft./ 10^8 particles dispersed and is probably much less. This is more than 1000 times less than the level that would be expected if the air in the room had been turbulently mixed. In this group, any contamination would have had to travel against the direction of air flow beyond the areas directly disturbed by movement of persons.

The second group showed contamination levels between about 100 and 1000 times less than those expected in a room where the air was turbulently mixed. This group comprises those experiments where dispersal took place at the rear of the room but movement of persons extended over the whole of the room, those where dispersal took place in one bed area and movement extended over the whole of the room but the sensitive area was separated from the dispersal area by a solid partition, and in addition those where dispersal took place in one bed area, movement was confined to this area and the rear of the room and the dispersal area was separated from the bed area by a curtain. In this group contamination had moved upstream against the direction of air flow in association with the movement of persons or had moved across the direction of air flow under the lower edge of the curtain when this was disturbed by persons moving alongside it. The colonies of *B. subtilis* recovered in these last experiments were confined to those plates placed on the floor within 1 ft. of the lower edge of the curtain. In this group the contamination increased as the velocity of air movement was reduced, but not significantly more than in proportion to the reduction in air velocity.

The third group includes the remaining experiments where the particles were dispersed in one bed area; there was neither curtain nor partition between this and the sensitive area and movement was either over the whole of the room or over the rear of the room together with the dispersal area. The contamination levels varied between 1/10 and 1/100 of those expected in a fully turbulently mixed room and increased more than proportionately to any reduction in the linear air velocity. In this group the contamination moved across the direction of air flow as a result of the movement of persons in the room.

The details of this process were examined in more detail by considering the colonies recovered on the plates exposed in the sensitive area in two groups: those recovered on the six plates exposed near to the centre line of the room, and those recovered on the six plates near to the wall and remote from the point of dispersal. The results of this analysis are given in Fig. 4. This shows, as would be expected, that many more colonies were found on those plates exposed near to the centre line. These were exposed little over 3 ft. from the line of particle dispersal and the number of colonies found depended little on whether or not movement of persons extend right over the sensitive area. Movement within the dispersal area in any

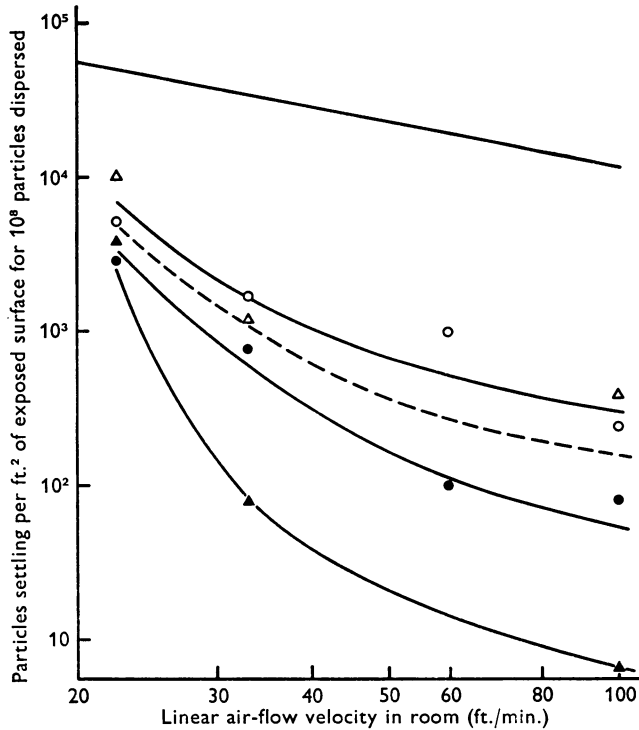


Fig. 4. Transfer across the direction of air flow. Dispersal took place at point A in area B 1. Triangular symbols, Δ and \blacktriangle , record experiments in which movement was restricted to the rear and area B 1 only (coded 2 in text and Table 1). Circular symbols, \circ and \bullet , record experiments in which movement extended over the whole of the room area (coded 3 in text and Table 1). Open symbols, Δ and \circ , record the spore-bearing particles recovered from the six plates exposed in area B 2, close to the centre-line of the room. Filled symbols, \blacktriangle and \bullet , record the spore-bearing particles recovered from the remaining six plates exposed in area B 2 close to the wall of the room. The upper straight line shows the numbers of spore-bearing particles that would have been recovered if the air had been completely mixed by turbulent movements. The lower curves have been drawn in by eye to illustrate the effect of the various conditions on the number of spore-bearing particles recovered.

case reached to within inches of these plates. If the air velocity was 35 ft./min. or over the numbers of particles reaching the plates exposed near to the room wall were between 10 and 100 times less than the number found on the plates near the

centre line when movement did not extend into the sensitive area, and between 2 and 10 times less when movement extended over this area. When the air velocity was reduced to only 22 ft./min., however, the numbers recovered differed little between the two positions whatever the nature of the movement during particle dispersal.

The effect of a curtain or partition and the distribution of contamination in the three room areas is shown in more detail in Table 2 for the experiments carried out at a linear air velocity of 35 ft./min. In particular, this includes the results of one experiment in which, although the two subjects walked over the whole area of the room, they restricted their movements as if a curtain or partition had been in place although no such barrier was in position. It is clear that this restriction had little if any effect on the figures obtained.

Table 2. *Numbers of spore-bearing particles settling per square foot of exposed surface for 10^8 particles dispersed*

Group	Movement	Partition or curtain	Number of particles settling in		
			Dispersal bed area, B1	'Sensitive' bed area, B2	Rear half of room, R
2	Rear of room and dispersal bed area only	Neither	15,500	650	22,000
		Curtain	21,000	57	53,500
		Partition	14,000	10	38,500
3	Over whole of the room area	Neither	6,500	1230	36,000
		Neither*	12,500	1380	34,000
		Partition	15,500	119	43,500

Equivalent particle diameter, 13 μ . Linear air velocity 35 ft./min. Dispersal took place in one bed area, B1.

* Although there was no partition or curtain in position during this experiment, the two subjects restricted their movements in the room as if such an obstruction had been there.

DISCUSSION

These experiments seem to establish to a high level of significance that little if any particulate contamination moves against the direction of air flow even when there is considerable movement of persons in the room. Persons moving from a contaminated region can, however, transport small numbers of particles in any direction. Lateral transport of particles across the direction of air flow, however, does not extend to any significant extent beyond a few feet from the area in which the movement is taking place so long as the velocity of linear air movement does not fall below 35 ft./min.

In setting up a room with a directed air flow for clinical studies it is convenient, especially in relation to controlling noise from the machinery, to use as low an air velocity as is compatible with efficiency. These experiments suggest that a horizontally directed air flow of 40 ft./min. with an inter-bed spacing of 5-6 ft. should be effective in eliminating airborne transfer of bacteria-carrying particles from one patient to another within the room. Protection from contamination generated at the down-stream end of the room should be even better. This ability to nurse more

than one patient in a single room and to allow unrestricted access to the downstream end of the space without impairing isolation from airborne infection should substantially ease the nursing and supervisory difficulties associated with isolation procedures and reduce the psychological problems that an isolation regime may induce in patients. The extent to which air isolation is important in practice for any class of patient is by no means clear, and a principal objective in future studies carried out in this room will be an exploration of infection transferred via nursing procedures in an environment free from the risk of airborne infection.

SUMMARY

Experiments have been carried out on the extent to which movement of persons in a room ventilated by a horizontally directed uniform air velocity can transport airborne bacteria from one position to another. More than 10^7 particles approximately 13μ in diameter (settling rate in air about 1 ft./min.) carrying spores of *Bacillus subtilis* var. *niger* were liberated in each experiment and the numbers reaching the different parts of the room were estimated by those recovered on exposed settling plates. At air velocities of 35 ft./min. and over, no particles could be certainly found to have moved against the direction of air flow except into areas actually entered by persons, and the numbers found in these areas were between 100 and 1000 times less than would have been expected in rooms turbulently ventilated with the same volume of air. There was some transport of particles transversely across the air-flow lines but at 4 ft. distance from the area where movement of persons was taking place the numbers had fallen substantially below 1/100 of those to be expected in a turbulently ventilated room.

The results recorded at an air velocity of 22 ft./min. were significantly less satisfactory.

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