MOVEMENT OF BACTERIA IN MOIST, PARTICULATE SYSTEMS*

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The role of the soil moisture regime in the ecology of soil bacteria is poorly understood. The response of the metabolic rates of bacteria to changes in matric suction has been investigated (Bhaumik and Clark 1947; Dommergues 1962; Clark 1967) but there is no precise information on the influence of the moisture regime on bacterial movement by flagellar activity or Brownian movement.

The following are suggested as defining some of the limiting conditions for the physical regime which will permit movement of bacteria over appreciable distances:

(1) Bacteria will depend upon continuous water pathways for their movement to, and spread upon, substrates because they lack a hyphal system to bridge air spaces.

(2) Even though pores are water-filled, bacteria will not be able to move appreciably if the relevant pore necks are too small. For a rod-shaped bacterium, a pore neck of radius 1–1·5 μ is likely to restrict severely the rate of passage, whether by Brownian or flagellar movement. Such a pore neck radius is equilavent to a suction of pF 3·0–3·2 (Griffin 1963*a*) and if the matric suction exceeds this value, there will be no water-filled pores in the soil sufficiently large to permit easy passage of the bacterium.

(3) To permit appreciable movement of the bacterium, it is not sufficient for there to be some water-filled pores of the requisite size; there must be enough to provide a continuous pathway. In turn, a continuous pathway will depend on the pore size distribution within the matrix.

(4) Even though the pores of relevant size may be drained, movement of bacteria may be possible if lenses of water, associated with the contact points of the soil particles, are themselves in contact. Such movement is likely to be of limited extent, however, for continuous water pathways of this type are likely to be short.

This paper reports studies on movement of bacteria in particulate systems under controlled moisture regimes and provides evidence in support of the suggestions outlined above.

Materials and Methods

(i) *The Bacterium.—Pseudomonas aeruginosa* Migula, a bacterium with rods 0·5 by 1·5–3·0 μ and motile by one to three polar flagella, was used because of its motility and characteristic pigment on meat-infusion agar. The bacterium was prepared by overnight growth in meat-infusion broth at 30°C. Large loopfuls of the broth were streaked onto freshly poured meat-infusion agar plates which were then incubated at 30°C for 8–12 hr. The bacteria on the plate culture were tested for motility in a hanging-drop preparation.

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(ii) *Particulate Systems.*—Particulate systems were provided by one natural soil and a range of aluminium oxide grits, the advantages of which have been previously discussed (Griffin 1963*b*). The grits used were "TP" Aloxite (Carborundum Co. Ltd. Australia). Their relevant physical properties are given in the following tabulation:

Mesh	60	100	220	320	600	Levigated
Mean particle diameter (μ)	254	122	63	25	16	3
Representative radius of pore neck (μ)*	48	24	9·4	4·5	1·8	—

The grit mixture had the composition by weight of 35% 100-mesh grit, 40% 220-mesh grit, 12% 320-mesh grit, and 13% levigated Aloxite. The soil was a lateritic red earth of sandy-clay-loam texture and pH 5·5 (measured in 0·1M $CaCl_2$).

Moisture characteristics (drying boundary curves) of the various systems are shown in Figure 1. Because the density of the soil and grits differed considerably, the water contents of the systems are expressed on a volume, rather than weight, basis to facilitate comparison. The bulk densities of the soil, 60-mesh grit, 600-mesh grit, and the grit mixture were 1·5, 2·48, 2·13, and 2·15 g/cm^3 respectively. Although some shrinkage occurred in the soil on draining this was insufficient to cause cracking in the soil pats used in the experiments.

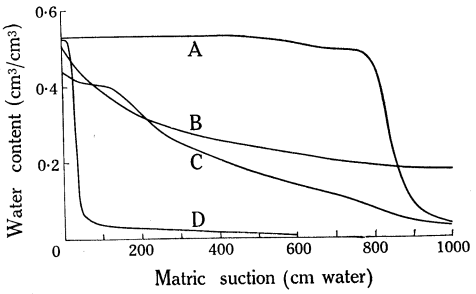

Fig. 1.—Moisture characteristics (drying boundary curves) of the particulate systems. *A*, 600-mesh grit; *B*, soil; *C*, grit mixture; *D*, 60-mesh grit.

(iii) *General.*—The soil and grits were placed in sintered-glass Büchner funnels (porosity grade 5) and matric suctions between 0 and 800 cm water were imposed using a method previously described (Griffin 1963*b*). To establish suctions in excess of 800 cm water, a small pressure membrane apparatus was used. All components of both systems were sterilized before assembly and use. The systems were allowed to come to equilibrium with the imposed suction before they were inoculated.

In most experiments the soil and grit samples were *c.* 1 cm in depth, and they were inoculated at a point with a mass of bacteria scraped from the surface of an agar culture. After 24 and 48 hr particles were removed with fine forceps from the system at distances of 0, 0·5, 1, and 2 cm from the inoculum. The particles were spread on freshly poured plates of meat-infusion agar which were subsequently examined for colonies of *P. aeruginosa*. All experiments were performed at 25°C.

An alternative technique was necessary with the 60-mesh grit because at p*F* 2·0 the grit had drained and could not be inoculated without unduly disturbing the system. In this case, a layer of 220-mesh grit was placed on the sintered-glass surface of the funnel and the 60-mesh grit was placed on top of this, leaving a small central area of the finer grit exposed. The fine grit was inoculated and, because its pores remained saturated at p*F* 2, the bacteria spread through it. Vertical movement of the bacteria from the 220- into the 60-mesh grit was assessed by sampling the superficial layers of the 60-mesh grit. The depth of the 60-mesh grit in various funnels varied from 0·5 to 2 cm.

* Equivalent radius of pore neck at point of inflexion of the moisture characteristic (drying boundary curve).

In early experiments, the grits and soil were initially saturated with either meat-infusion broth or with water but, as the movement of bacteria in the two liquids was the same, later experiments were conducted with water only as the liquid. Most experiments were replicated three times.

Results

In general, no differences in movement of *P. aeruginosa* were detected either between replicates or between water and broth. The results have, therefore, been consolidated and one result only is given in Table 1 for each system at each matric suction. In subsidiary experiments, movement of bacteria was detected at 100 but not at 500 cm suction, in one light sandy loam and in one loamy sand.

TABLE 1

MOVEMENT OF BACTERIA UNDER VARIOUS MOISTURE REGIMES

Matrix	Matric Suction, h (cm water)	Water Content (%)	$\Delta\theta_h$ (cm^3/cm^3)*	Movement†
Soil	100	39·0	0·215	++
Soil	181	33·0	0·155	+
Soil	253	30·0	0·125	+
Soil	313	28·0	0·105	—
Soil	500	24·0	0·065	—
Soil	1000	17·5	0	—
Grit, 60-mesh	100	0·74	*c.* 0·007	—
Grit, 600-mesh	500	52·5	0·49	++
Grit mixture	100	40·5	0·375	++
Grit mixture	500	17·0	0·14	+
Grit mixture	1000	3·0	0	—

* Volume of water held in pores draining between h and 1000 cm water suction.
† ++ 2 cm in 24 hr; + 0·5 cm in 48 hr; — no movement in 48 hr.

Discussion

The rate of movement of the bacteria in the systems (2 cm in 24 hr) was similar to that reported by Thornton and Gangulee (1926). That movement was not detected at pF 3·0 supports the first two suggestions concerning limiting conditions as outlined in the Introduction. At other suctions, no clear pattern relating bacterial movement to suction or water content was evident (Table 1). The data were therefore analysed in terms of pore size distribution and the continuity of relevant pathways.

If it is assumed that a pore neck of radius *c.* 1·5 μ severely limits bacterial movement through the necks, then all water held at suctions greater than pF 3·0 can be neglected when considering rapid bacterial movement. The possibility of such movement in a given soil at a suction of h cm water will then depend upon the continuity of water-filled pathways with necks of radius r cm, where $0{\cdot}00015 \leqslant r \leqslant 0{\cdot}15/h$ (Griffin 1963*a*). In turn, this continuity will be reflected in the volume of water held in such pores.

Let the volume of water held in the pores draining between h and 1000 cm water suction be $\Delta\theta_h$ cm^3/cm^3. At 500 cm water suction, movement was detected

in the 600-mesh grit and in the grit mixture but not in the soil, the values of $\Delta\theta_{500}$ being 0·49, 0·14, and 0·065 cm^3/cm^3 respectively. The critical value for $\Delta\theta_h$ for these systems, therefore, lies between the last two values. Such limits are narrowed by the detection of movement of 0·5 cm in the soil at 253 but not at 313 cm water suction, $\Delta\theta_h$ being 0·125 and 0·105 cm^3/cm^3 respectively. These results demonstrate that a critical volume of water is necessary for appreciable movement of bacteria to occur. If the results have general validity, the movement of bacteria in most soils will be very restricted if the soil is much drier than field capacity.

The above arguments should not be taken to imply necessarily a general reduction in the rate of bacterial metabolism at these suctions because extensive movement may not be necessary for bacterial activity in many circumstances. Dommergues (1962) has shown that in some cases bacterial metabolism is not reduced until suctions considerably in excess of pF 3·0 have been developed. Such activity presumably depends on bacteria already situated on the substrate so that movement is of minor importance.

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