SOIL FUNGI AND SOIL FERTILITY

2nd Edition

S. D. GARRETT

PREFACE

SINCE the first publication of this book in 1963, dramatic advances have been made in our knowledge and understanding of the way in which micro-organisms live and function, both under controlled conditions in the laboratory and in the natural environment of the soil. These advances have necessitated a replanning as well as a rewriting of this book, so that seven of the nine chapters are largely new. As before, this account of soil fungi is intended primarily as an introduction to soil microbiology for university undergraduates, and to show how the living ecosystem of the soil functions in the maintenance of soil fertility for crop production.

It is a pleasure to acknowledge the help I have had from others, by their critical reading and valuable comments on four chapters. Firstly, to my former research student Professor D. M. Griffin, of the Australian National University, for reading Chapter 2; in the subject that he has made so much his own, he has now been the teacher and I the taught. Secondly, to my colleague Dr. John Rishbeth, F.R.S., for reading Chapter 3 and also for all that I have learned from him in our common field of research on pathogenic root-infecting fungi. Thirdly, to my colleague Dr. H. J. Hudson, for reading Chapters 4 and 5; from his work and writings on fungal saprophytism I have learned much and now pass some of it on to others.

I am especially grateful to Mr. Brian Golding for three original drawings, now reproduced as Figs. 3, 5 and 8 and also for drawing the graphs shown in Fig. 11. I am indebted to authors, editors and publishers for permission to reproduce Figs. 1, 6, 9, 16, 17 and 18, sources of which are acknowledged individually in the text. Lastly I thank Mrs. Ruth Hockaday for the great skill and care with which she typed out my manuscript.

CONTENTS

CHAPTER 1

INTRODUCTION

THIS book will be an introduction to the microbial ecology of the soil, using soil fungi as examples. Fungi have various advantages for ecological studies. They are the easiest micro-organisms to recognize and identify, which fits them for quantitative as well as qualitative investigations in soil. The fundamental study of soil fungi has been greatly encouraged by the economic importance of two groups of root-infecting fungi. The mycorrhizal fungi live on and in the roots of their host plants in a *symbiosis* that benefits both partners, because these fungi are more efficient in absorbing plant nutrients, especially phosphates, from infertile soils than are non-infected root systems. The pathogenic root-infecting fungi cause widespread and sometimes serious losses of yield in almost every crop all over the world. Nevertheless, even crop pathogens have their use as censors of bad agricultural practice. When I was working on the take-all disease of wheat in South Australia, in the early 1930s, it became obvious that intensive growing of wheat on the poorer, sandy soils was causing not only widespread losses from this disease, but also a decline in soil fertility and consequent wind erosion of these soils. Again in 1948, when working on Panama disease of bananas in Jamaica, I declared that the widespread abandonment of large tracts of land for banana growing, caused by this disease, had come in time to save much further loss of surface soil from the mountainous areas due to devastating water erosion. This opinion was not well received, nor had I expected it to be; blessings in disguise are not readily recognized by those who have suffered both financial loss and disruption of their traditional cropping practices.

In their mode of nutrition, fungi contrast with all green plants, which are

able to fix solar energy as carbohydrates through their possession of chlorophyll pigments. All such plants, from the largest forest trees down to micro-organisms like the blue-green photosynthetic bacteria (formerly styled the blue-green "algae"), are termed *autotrophic*, i.e. self-nourishing. Similarly, various groups of chemosynthetic bacteria, which derive their energy from the exothermic oxidation of sulphur and iron compounds, can also be called autotrophic. Fungi are called *heterotrophic*, because they derive their energy from the exothermic break-down of organic substrates produced by other organisms, either by the original autotrophs or by another organism along the food-chain. Fungi are not producers of energy; they are consumers. But they play a useful part in the maintenance of terrestrial life through their widespread activity on and in the soil. If soil organisms and micro-organisms did not play their essential role in the breaking-up and decomposition of dead vegetation on and within the soil, the soil surface would become littered with a deep layer of plant refuse, of little value as a source of nutrients for the root-systems of higher plants. Members of the soil fauna break up the plant remains into smaller fragments, which are thus more quickly degraded by the heterotrophic soil bacteria, which are surface-feeders. But it is the soil fungi that are typically the pioneer colonizers of dead plant tissues. They are fitted for this pioneering role by their physical organization into a network of *mycelium*, composed of branching, rigid tubes (*hyphae*), filled with protoplasm. Such hyphae are able to penetrate cellulose walls in plant tissue, even those strengthened by lignification, by a combination of enzyme action and growth pressure exerted by the hyphal apices, supported by a rigid mycelium. The unicellular bacteria do not have this mechanical advantage for penetration of intact though dead plant tissues, and so their cellulase enzymes can cause only surface erosion of cellulosic plant tissues; nevertheless, they can later enter dead plant tissues through punctures made by fungi, and through the more extensive breakage and comminution caused by the soil fauna.

So a corpus of dead plant tissue is gradually broken down by the commensal activities of the saprotrophic soil fauna and the heterotrophic soil fungi and bacteria. This word *commensal* means feeding "together at the table" and it implies no more than this. It must be carefully distinguished from the term *mutualistic symbiosis*, which means "living together" by two (occasionally more) partners with *mutual* advantage, e.g.

lichens, which are composite plants formed by association between fungi and green algae. The final result of this commensal break-down of dead plant tissue is the liberation of the essential mineral nutrients needed by higher plants in a soluble form, i.e. nitrates, phosphates, sulphates and salts of potassium, calcium and magnesium, which can be taken up from the soil by their root systems. So the essential mineral nutrients needed by plants, animals and micro-organisms, N, P, K, Ca, Mg, S and others in smaller quantities, are kept in circulation by the heterotrophic soil micro-organisms. We have to note, however, that successive generations of green plants cannot reclaim the carbohydrate energy-reserve that is locked up as cellulose, etc., within the skeleton of dead plant tissue. But these energy reserves indirectly benefit the green plant, because they provide the wages for the microbial work-force that reclaims in soluble form the mineral nutrients orginally locked up in the dead plant tissues.

So far we have emphasized the commensal nature of these break-down activities, partly because in the past, soil microbiologists have sometimes ignored the important part played by the soil fauna. Sometimes this has been done for research convenience, quite deliberately. The soil fauna of insects and other small arthropods, etc., add one more factor to an already complicated situation, and various kinds of insect larvae, for example, devour fungal mycelium and its fructifications on which identification of the species depends; it is possible to minimize this unwelcome activity by the simple expedient of air-drying soil or plant tissue before experimental use. But while recognizing the universal importance of commensal action in the soil, it is opportune to point out that fungi and bacteria typically occupy somewhat different *ecological niches* in the soil, at least as pioneer colonizers of dead plant and animal tissues, when their respective roles are more easily distinguished. In this respect, the supply of oxygen, i.e. its partial pressure, within the soil or inside a corpus of tissue, plays a differentiating role. In general, the fungi are strict *aerobes*; their activity is often limited by oxygen supply, though the yeast fungi constitute a well known exception. Some bacteria are as oxygen-demanding as the majority of fungi; others, known as *micro-aerophiles*, grow best in very low oxygen tensions, and there is a large and important group of *obligate anaerobes*, which will not grow at all in the presence of oxygen.

Such obligate anaerobes are the chief decomposers of mammalian corpses, the tissues of which quickly become anaerobic after death, when

the ventilating action of heart and lungs has ceased. The inoculum for decomposition is already there within the corpse, because the intestines harbour a large and active microflora of anaerobic bacteria. The high water content of the material in an animal's alimentary tract provides a favourable habitat for bacteria and the anaerobic condition of the intestinal contents limits the *active* microflora to anaerobic bacteria. Ruminant herbivores, such as cattle, have a highly evolved fermentation chamber in the anterior part of their alimentary tract; this is known as the *rumen*. The animal itself does not secrete cellulase enzymes but members of the bacterial population in the rumen do so and thus they can degrade the cellulose cell-walls in the well chewed fragments of herbage into soluble sugars, part of which is absorbed by the host animal after microbial transformation into fatty acids, together with digestion products of proteins, etc.; in this way, the herbivorous ruminant benefits by extracting a much higher proportion of the nutrients than it could do if unaided by the bacterial microflora of its digestive tract. This association is clearly a symbiosis; the benefits to the host animal of cellulose digestion are obvious, and the bacterial population is provided both with a food substrate and optimum conditions for its decomposition. Evolution of the rumen seems to be a remarkable adaptation to a herbivorous diet; the wild rabbit has no rumen but it regularly devours its own dung balls, thus submitting them to a further extraction of residual nutrients.

These examples have been chosen to show that sometimes a single master factor, in this case oxygen supply, may broadly differentiate between several groups of micro-organisms, all of which may be at least potential competitors for a corpus of dead plant or animal tissue providing a *substrate*, i.e. food material for micro-organisms. Usually, however, many factors in both substrate and environment act together in selecting the micro-organisms best fitted for colonization of a particular ecological niche. Substrate factors comprise physical organization, chemical composition and relative availability of nutrients. Factors of the soil environment include temperature, degree of aeration, availability of water, chemical composition and availability of mineral nutrients, and hydrogen ion concentration (pH value). But before we can understand the complex organization of microbial activities within the soil, it is essential to understand, at least in broad outline, the physical framework and chemical

composition of the soil. This phrase "to understand" is here used in a relative (or optimistic) sense and it will soon appear that much remains to be found out about the behaviour of soil by physicists, chemists and microbiologists.

CHAPTER 2

SOIL AS A HABITAT

The shortest definition of "soil" that I have seen was produced by G. V. Jacks (1954) in his book *Soil*: "Soil is what plants grow in". To explain clearly the physical and chemical framework of soil as a habitat for organisms, it is unfortunately necessary to subdivide aspects of the subject that are not naturally divisible; there are no sharp demarcations in the natural world but they are still necessary for the exposition and administration of science.

PHYSICAL CHARACTERISTICS OF SOIL

Soil texture

This term expresses the distribution of the ultimate particles of a soil within a range of conventionally determined sizes, as shown in Table 1.

TABLE 1. GRADING OF SOIL MINERAL PARTICLES

Grade	Particle diameter (mm)
Gravel	>2
Coarse sand	2–0.2
Fine sand	0.2–0.02
Silt	0.02–0.002
Clay	< 0.002

The size distribution of the particles is determined by a method known as *mechanical analysis* of soil. Particles are separated by sieving for grades

down to and including coarse sand, the weight of which in the soil sample is determined after drying. Separation of the finer grades depends on the fact that the finer a mineral particle, the more slowly does it fall through water. The time taken by spherical particles to sediment through a standard vertical column of water at a standard temperature can be calculated from Stokes's equation. Although the mineral particles are far from spherical and their mean density has to be conventionally assumed as 2.6, yet these assumptions do not invalidate the method for comparison between different soils. The method of analysis begins with crushing the soil, so that as much as possible will pass through a sieve of 2 mm mesh. The organic matter is first removed by boiling the soil in a solution of hydrogen peroxide; the organic matter content is determined on another sample of soil. Calcium carbonate is then removed by leaching with dilute hydrochloric acid; this has to be done because calcium salts tend to flocculate the mineral particles, i.e. cause them to cohere into aggregates. Any soil aggregates still remaining are finally dispersed by shaking the soil sample in a dilute alkali solution, after which the fraction of coarse sand is separated off by wet sieving and the remainder is suspended in a vertical column of water. At suitable intervals, the density of the soil suspension at a fixed depth is determined, most quickly and easily with a hydrometer, the use of which for this purpose was introduced by G. J. Bouyoucos in 1927. From the results, the distribution of the ultimate mineral particles between the various grades can be calculated.

The results of such mechanical analyses of soils are needed particularly by pedologists in their work on the evolution and classification of soils. But the experienced soil surveyor can often make quite a good guess at the mechanical composition by merely examining and handling the moist soil, just as an experienced statistician can run his eye over a table of data and assign a probable degree of significance for differences between means. Those who enjoy country walks learn more about soil texture from their feet than they may consciously realize. Thus it is possible to run across a field of bare sandy soil without much effort and without getting the legs dirty; if a similar traverse is made across a field of wet clay, especially if recently ploughed, the first such experience will not soon be forgotten. Such a sandy soil is often referred to as "light-textured" or "light", whereas a clay soil is similarly called "heavy-textured". These fairly ancient terms have nothing to do with what is called "bulk density" of a soil; they refer to

the number of horses required to pull a plough through the moist soil, which would have been only one for most sandy soils and up to four for a clay soil. But the draught of a plough is not determined by soil texture alone in any particular soil at any particular time, because it is affected by the state of aggregation of the soil particles to give the *soil structure* (see next section). The individual farmer has to decide when a heavy clay soil is at the optimum moisture content for ploughing, i.e. when the power required to draw the plough is least; through the correct use and timing of his cultivations he can create and preserve an optimum soil structure, with resulting economy in cost of power. Lastly, we should note that soils intermediate in texture between sands and clays are usually described as *loams*. A loam soil usually contains not more than *ca.* 30 % clay along with 40–50 % sand, but sub-divisions are often made, not always accurately I suspect, into light loams, medium loams and clay-loams.

Soil structure

Most soils have a definite and visible structure; although various types of soil structure have been described, especially for natural soils under wild vegetation, that which is deemed best for agriculture is a granular or *crumb structure*. The soil crumbs, like the ultimate mineral particles, vary over quite a wide size-range (optimum 1–5 mm diam.) in different types of soil, and also within one soil at different times, according to management by the farmer. In each crumb, the coarse particles are usually surrounded by fine particles, which cement the crumb together through the surface forces associated with fine particles of clay and humus. A cultivated soil in good *tilth*, as the farmer would say, is made up of an assemblage of variously sized soil crumbs, separated by a network of intervening *soil pores*. In a soil at medium moisture content, the soil pore space is occupied by air and water (more strictly speaking, by the soil solution) in roughly equal proportions. Micro-organisms live both within the pore spaces and also in the interior of the crumbs, which provide a microbial environment somewhat different from that in the pore spaces; the centre of a water-saturated crumb is likely to be anaerobic if the crumb radius exceeds *ca.*3 mm. The total population of bacteria in a soil at any time is probably limited not only by available substrates providing nutrients, but also by the total internal surface area, both within and outside the soil crumbs, per unit

volume of soil. This is because bacteria, much more than fungi, are restricted to growth in films of liquid covering internal soil surfaces. As noted above, the structure of a soil can be much influenced, for better or for worse, by a farmer's crop-husbandry operations. Grassland is one of the best makers and preservers of good crumb structure and so a good arable rotation generally includes a temporary pasture, or ley, usually made up of one or more grasses along with a legume, for one or more years. The mechanical effects of the growth of a mat of fine grass roots may contribute to this effect, in addition to the fact that roots produce an external coating of mucilage. Young grass roots, like young roots in general, exude a solution that serves as a more or less complete nutrient medium for growth of micro-organisms. This increased population of micro-organisms around the young, active roots is known as the *rhizosphere*. Many species of soil bacteria produce a sheath of polysaccharide substances around their cells and such gummy excretions can help the building up of soil crumbs, as can the binding effect of fungal mycelium and the organic residues left when the mycelium dies and autolyses. Such possible effects of soil micro-organisms on crumb structure have been critically reviewed by Griffiths (1965).

Soil water content

It is quite easy to determine the moisture content of a sample of soil, by weighing it before, and again after, it has been air-dried or oven-dried. For any collection of soil to be used for experimental work in soil microbiology, it is necessary to calculate moisture content on an air-dry basis, because soil is denatured by oven-drying and is then quite useless for microbiological studies. Anyone who looks after potted plants in a glasshouse soon learns from experience when plants need watering; by the appearance and feel of the soil one can say whether it is too dry, about right or too wet, and for this purpose no precise determination of soil moisture content is needed. But a statement that an unknown soil contains a certain percentage of water is usually meaningless, because the weight of water that can be taken up and held by 100 g air-dry soil at saturation usually varies over the range 25–80 g. From about the year 1920 onwards, therefore, microbiologists determined the saturation capacity of any stock of air-dried soil to be used in laboratory experiments, employing the *perforated box method* described by Keen and Raczkowski (1921), or some modification of it. Rectangular

boxes with a perforated bottom, lined with a sheet of thin filter-paper, were weighed before and after filling with air-dried soil, and then stood in a shallow layer of water until the soil was saturated; then the boxes were re-weighed. From the figures thus obtained, the weight of water held by 100 g air-dry soil could be calculated. From this figure, the volume of water required to bring *ca.*200 ml air-dried soil to a medium moisture content, of 50–60 % saturation, could be calculated. Cylindrical glass jars of *ca.*300 ml capacity were used as soil containers; if the required volume of water was added not too quickly to the surface of the air-dried soil, it would percolate downwards, and soil moisture would become evenly distributed within 24–48 h, as shown by determining actual moisture content of consecutive slices of soil. Those employing this method of preparing moist soil in small containers usually experienced no difficulty in getting uniform packing of soil, previously passed through a 2 mm sieve, or uniform distribution of water down to a moisture content of 50 % saturation. Some types of soil might be difficult to handle and so were unsuitable for this kind of laboratory work. But at soil moisture contents below *ca.*50 % saturation, distribution of soil moisture was less good and failed to become even through the soil. So most workers soon realized that this method was unsuitable for study of the effect of soil moisture content upon microbial activity and other methods had to be found.

The procedure outlined above made it possible to bring a soil to a medium or high moisture content, and to maintain it there by periodical addition of de-ionized water to return soil containers to their original weights, i.e. to replace water lost by evaporation from the soil surface. So the effect of experimental treatments upon microbial activity in one particular soil could be compared, and experimental results were usually repeatable in consecutive experiments with the same stock of air-dried soil. But eventually it was realized that different types of soil held at the same moisture content, as expressed by percentage saturation, were not at equivalent moisture contents with respect to availability of water to plant roots and micro-organisms. To withdraw water from a soil at a moisture content of less than saturation, roots and micro-organisms have to exert a suction, and the magnitude of the required suction increases as the soil gets drier. The tenacity with which a soil holds water against the demand of roots and micro-organisms varies widely according to the type of soil; the finer the soil particles, the more strongly is water held by surface forces.

Thus a quartz sand holds only *ca.*20 g water/100 g sand at saturation, but only a low suction, of *ca.* −1 atm, is required to withdraw nearly all the water. A clay soil, on the other hand, may hold up to 80 g water/100 g soil at saturation, but a suction of −1000 atm will be needed to withdraw nearly all of the water. At a moisture content of 30% saturation, a suction of *ca.* −0.5 atm will be needed to withdraw water from the sand, whereas one of *ca.* −10 atm will be required to withdraw it from the clay soil. So it becomes clear that the expression of soil moisture content as percentage saturation gives little information about the availability of water to roots and micro-organisms and so it does not afford a valid basis for comparison between different soils over a range of moisture contents.

This then new concept about the state of water in soil and its measurement was embodied by R. K. Schofield (1935), a soil physicist at the Rothamsted Experimental Station, in a paper entitled "The pF of the water in soil". Schofield expressed the suction needed to withdraw water from a particular soil at a given moisture content (g water/100 g soil dried at 105°C) on a logarithmic scale, on which pF 0 = 1 cm water suction, pF 1 = 10 cm and pF 7 = 10^7 cm water suction. Expression of pF on a $\log_{10}$ scale is thus analogous to the expression of hydrogen ion concentration on the pH scale.

The new ideas concerning soil water content developed by Schofield and others took a long time to penetrate current laboratory practice in soil microbiology; soil physicists may well have said that it took a *very* long time. Adoption of a new technique, however, involves more than the assimilation and adoption of a new idea; it also means getting used to a possibly more difficult and laborious method. At that time, measurement of soil pF involved the use of a water tensiometer. Much was done by D. M. Griffin (1963) in a paper entitled "Soil moisture and the ecology of soil fungi" to persuade soil mycologists and microbiologists to use the new methods; since that time their assimilation has been quite rapid (Cook and Papendick, 1970).

The form of the relationship between the actual moisture content of a soil (g water/100 g oven-dry soil) and pF value is shown in Fig. 1, reproduced from Griffin's (1963) paper.

The curves shown for a sand, a loam and a clay soil in Fig. 1 are known as *moisture characteristic* curves; those for sand and clay represent the extremes of relationship between moisture content and soil pF, with the

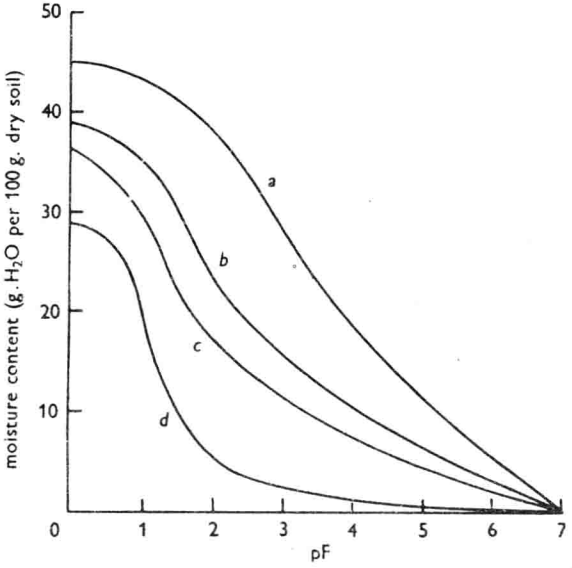

FIG. 1. Representative moisture characterisitics: *a*, drying boundary curve of a clay; *b*, drying boundary curve of a loam; *c*, wetting boundary curve of a loam; *d*, drying boundary curve of a sand (reproduced from D. M. Griffin (1963), *Biological Reviews*, Cambridge).

loam as an intermediate. As Griffin has explained, the moisture characteristic curves for collections of soil from one locality may vary with structure (a variable characteristic) as well as with texture, and also with the past history of wetting and drying of the soil. The latter point is illustrated by the two central curves in Fig. 1; *b* represents the drying boundary-curve for the loam soil drying out from saturation, and *c* the wetting boundary-curve for the same soil taking up water from the oven-dry condition. The difference between these two boundary curves, with possible intermediates, is due to a phenomenon termed *hysteresis*, which need not be explained here; its existence should be remembered, however, together with the fact that the drying boundary-curve is easier to determine, because of the greater speed of equilibration between soil and water.

To complete this explanation of the behaviour of water in soil, and to bring it up to date, just a few further points have to be mentioned. The state of water in soil is now generally expressed not on the pF scale but on that of

a related scale, known as the *water potential*; in a soil less than saturated with water, the water potential has a negative value. Water, like heat, flows from regions of high to regions of lower energy. Pure, free water at atmospheric pressure is assigned a value of zero potential energy. Because work has to be done by applying suction, to withdraw water from soil at a moisture content less than saturation, the potential of the soil water has a negative value. Just so, pure free water with zero potential will flow into unsaturated soil with a negative potential. At one time, before the introduction of S.I. units (*Système International d'Unités*) water potential would have been expressed in terms of a negative value of atmospheric pressure, e.g. −1, −2, etc. atm. I have used this older terminology earlier in this discussion, because to many microbiologists it is still more familiar than the new units. The new S.I. unit is the *pascal* and 1 MPa (megapascal) = 9.87 atm. An alternative unit, the *bar* (1 bar = 0.987 atm.) has been employed until recently, but under S.I. recommendation should now be restricted to meteorology; its use in soil science is thus obsolescent.

Lastly, the forces that hold water in soil can be briefly categorized. Over the range of soil moisture content in which microbiologists are likely to be interested, i.e. from saturation down to air-dry in an arid climate, these are of two types. Firstly, there is an osmotic effect due to solutes in the soil solution associated with colloidal particles of clay and humus; this imparts a negative *osmotic potential* to the moist soil. In saline soils characteristic of irrigated areas in arid climates, this negative osmotic potential may be considerable, but it is often negligible in temperate climates except where an excess of chemical fertilizers has been applied. More usually, much the larger component of total water potential is *matric potential*. The negative value of matric potential is due to the forces that act at air-water and solid-water interfaces. These forces are sometimes called *capillary forces*, because in part they are similar to that causing water to rise within a vertical capillary tube. In part, however, matric potential is due to the forces that bind water to the mineral particles, aggregates and organic material that together make up the soil.

Methods for measuring and controlling water potential in soil have been described by D. M. Griffin (1972) in his book *Ecology of Soil Fungi*. Use of an instrument like the thermocouple psychrometer will eventually become as widespread in both field and laboratory studies as is now the use of a pH-meter. Griffin's book can be strongly recommended as an introduction to

soil physics for microbiologists. All physical factors are treated in detail and in depth, but Griffin's accounts are so arranged that those unable or unwilling to follow mathematical and physical theory can still read this book with much profit. These remarks can be kept in mind for the section now to follow.

The soil atmosphere

In a soil at medium moisture content, about half of the soil pore space will be filled with liquid and the remainder with the soil atmosphere. When soil is in a state of activity from the growth of roots and decomposition of organic substrates by heterotrophic micro-organisms, the average partial pressure of carbon dioxide in the soil atmosphere is likely to be somewhat higher than that of the air above ground, and the partial pressure of oxygen may be slightly lower. But around respiring roots and their rhizospheres of micro-organisms, these differences may be much accentuated. Griffin (1972) has pointed to an important distinction between soil fungi and bacteria in respect of soil location. By their organization as a rigid mycelium, fungi are enabled to grow across and to occupy air-filled pore spaces; bacteria are obliged by their unicellular organization to grow sessile on the internal surfaces of the soil, around and within the soil crumbs. For active growth, bacteria require a film of liquid covering soil surfaces, which must be deep enough to accommodate their cells, i.e. a minimum of 2 μm. Griffin has concluded that, in most natural soils, bacterial growth and activity is likely to be restricted at water potentials less than -0.5 to -5 bar, whereas many kinds of soil fungi can tolerate soil water potentials lower than this. Griffin (1968) had earlier shown, by microscopical observations of fungi growing in a translucent particulate matrix of glass micro-beads, that fungi such as *Cochliobolus sativus* and *Curvularia* sp. sporulated only in air-filled spaces large enough to accommodate their rather large spores. But another species, *Fusarium culmorum*, produced its masses of slime spores within the liquid films though adjacent to the liquid-air interface.

The question of the precise location of micro-organisms within the soil has aroused much interest in recent years, particularly with regard to composition of the soil atmosphere within micro-habitats such as the surface of growing roots. The whole situation has been much clarified in a