

SPECIAL ARTICLES

THE LAWS OF DISTRIBUTION OF PARTICLES IN SUSPENSION

THE motion and distribution of fine particles suspended in a fluid are subject to laws which have been derived by statistical methods by Einstein and extensively studied experimentally by Perrin. Their results are well known and frequently cited. I wish here to point out a very much simpler derivation than Einstein's of a slightly more general law than Perrin's. The reasoning is thermodynamic rather than statistical.

Throughout any aggregate of particles in thermal equilibrium, the kinetic energy of thermal agitation is everywhere RT/M calories per gram (R gas constant, T absolute temperature, M molecular weight), otherwise there would be a net flow of heat which is contrary to the assumption of uniform temperature. The size or kind of particle is assumed to be immaterial and it may be either molecular or microscopic. The pressure corresponding to this energy, which we shall call the kinetic pressure

$$(1) \quad \alpha = CJRT/M \text{ dynes/cm}^2$$

is the integrated kick of all the molecules or particles of a given class (defined by M and concentration C) or of all classes. For microscopic particles it is the pressure due to Brownian movement which is caused by and is in equilibrium with the thermal agitation of the molecules of the suspending fluid.

The weight of such an aggregate of particles is

$$(2) \quad W = Ckg \text{ dynes/cc}$$

where C is concentration in grams/cc, $g = 980$ dynes/gram and $k = 1 - \rho_1/\rho_g$, ρ_g being the density of the grains and ρ_1 that of the suspending fluid.

The pressure gradient $d\alpha/dz$ depends upon the variation of the ratio CT/M with depth. If this ratio is the same at all depths, the kinetic pressure must be uniform and the gradient zero. In a true solution C and M are both constant if the temperature is uniform and $CT = \text{constant}$ (Soret effect) if it is not. Even in a suspension of fine particles CT/M may be constant if, at various levels, the concentration is proportional to size of particle, *i.e.*, if the number of particles in unit volume is constant. In general, an aggregate of particles will be in equilibrium distribution if the pressure gradient upward just equals the weight of suspended material in unit volume

$$(3) \quad d\alpha/dz = dw$$

or, by substitution from (1) and (2), if

$$(4) \quad \frac{d \log(CT/M)}{dz} = \frac{Mkg}{JRT}$$

which is the most general equilibrium condition. In the special case of uniform mass of particle M and uniform temperature T , (4) reduces to $d \log c/dz = Mkg/JRT$ which may be integrated giving

$$(5) \quad \log(C/C_0) = Mkgz/JRT$$

the integration constant yielding C_0 , the concentration at the surface where $z = 0$. (5) is the equivalent of Perrin's equation.

If the pressure gradient in a suspension is greater than its immersed weight then it will diffuse upward to the surface; if less than the weight it will settle according to the generalized Stokes' law with the velocity

$$(6) \quad v = \frac{2}{9} \frac{r^2 \rho_g}{\eta C} \left(Ckg - \frac{JRT}{M} \frac{dC}{dz} \right).$$

The expression bracketed is the resultant force acting on all the particles in unit volume. When the concentration gradient $dC/dz = 0$, (6) reduces to the ordinary form of Stokes' law. This and other equations above may be put in different forms by using the substitutions $C = mN$, $M = V\rho_g = M$ times the mass of the hydrogen atom, 1.66×10^{-24} gram.

From the form of (6) it is evident that velocity of fall is very sensitive to size of particle. It may readily be zero or even negative (upward) for the smaller particles. As each size of particles falls, smaller particles are not only left behind but tend to diffuse upward.

The kinetic pressure theory of suspensions here developed assumes clean reflecting walls for the containing vessel. A soft or muddy bottom, for example, quite actively assists in pulling down a suspension. Light pressure also effects settling, fine suspensions depositing on the far side wall of a tube exposed to strong horizontal illumination.

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THE OCCURRENCE OF VIABLE COTTON ROOT-ROT SCLEROTIA IN NATURE

A TRUE sclerotial stage of the cotton root-rot fungus, *Phymatotrichum omnivorum* (Shear) Duggar, was first observed in Arizona by C. J. King and H. F. Loomis in September, 1928, in laboratory cultures.¹ These observations were later confirmed by the writer in Texas, and such sclerotia have also been discovered in the soil, first in northern Texas at the U. S. Cotton Breeding Field Station near Greenville, in Hunt County, and later in southern Texas at the U. S. Field Station near San Antonio. As the sclerotia formed readily and abundantly in culture jars containing layers of sterile soil, sand and cotton roots,

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after intervals of from thirteen to fourteen days, it was believed that careful examination of the soil of fields infested by the fungus would ultimately reveal the true sclerotia under natural conditions. Although various sclerotium-like bodies similar in their general morphology to the sclerotia had been found in the soil from time to time during the winter and spring months, attempts to recover the fungus in culture from such bodies were not successful. Accordingly, it was decided to make deeper and more careful excavations of infested soil areas.

After studying the infection maps for the previous years, excavations were made on May 24 near the center of a station plot designated as D-7. This particular plot was planted to cotton in 1928 and is fallow this season. The infestation over this entire area last season was exceedingly heavy and more or less continuous, with little if any well-defined individual spots.

Soil samples were taken at depths of twelve, sixteen and thirty-two inches and the soil particles carefully examined under suitable hand lenses. Several sclerotia were found free in the soil, unattached to any plant material, at different depths. One sclerotium showed even under the low power of the microscope the characteristic strand hyphae and acicular branches of the root-rot fungus. The sclerotia were mostly single, averaging slightly less than two millimeters in diameter, but some were attached or paired. All the sclerotia obtained from these excavations were placed in small vials and immediately removed to the laboratory for tests of viability. Cultures were obtained from sclerotia collected at depths of twelve and sixteen inches.

The sclerotia were surface sterilized for forty-five seconds in a 1:1000 solution of mercuric chloride, dipped for a brief interval in 50 per cent. alcohol, washed in sterile distilled water and cultured on acidulated corn-meal agar slants in test-tubes and Petri dishes. Two days later they were removed to similar tubes and Petri dishes containing non-acidulated corn-meal agar. On May 29, three days after the cultures were made, evidence of germination was apparent in two of the tubes, the young hyphae appearing first white in color, somewhat rigid and bristly. On June 4 mycelial strands were plainly visible on the sides of the tubes, extending several millimeters above the media, of a yellow to light ochraceous buff color, such as characterizes root-rot hyphal strands after a few days' growth on a carbohydrate medium. Microscopical examinations were made of the cultures on June 6, and the large-celled, Rhizoctonia-type of mycelium was observed, with two-forked and three-forked branches arising from the large cells and becoming flexuous and delicate as growth continues. On June

10 transfers were made to flasks containing dead sterilized cotton roots and a slight excess of moisture. On this medium typical strand hyphae, acicular hyphae and pseudosclerotia were formed in eight to ten days, with the latter arranged in a typical tree-like pattern and attached to the strands on the walls of the flasks. This form of growth is typical of *Phymatotrichum omnivorum*.

On June 15 transfers were also made of the mycelium which the sclerotia had produced on the agar slants to wide-mouth pint bottles containing alternating layers of sterile soil, sand and cotton roots. These cultures were incubated at 29.5° C., and, after a growth period of thirteen days, produced true sclerotia identical in their morphology to those produced in other cultures of the fungus isolated from the roots of infected cotton plants at the Greenville Station during the season of 1928.

One of the first sclerotia obtained, at a depth of twelve inches, was fixed in chromo-acetic acid solution, dehydrated, imbedded in paraffin and longitudinal sections prepared eight microns in thickness. These were stained in Flemming's Triple Stain and in iron-alum hematoxylin. They reveal the same general type of structure as in other sclerotial fungi and in the cotton root-rot sclerotia which have been produced heretofore in culture. They are round, oval, ellipsoid, reniform or variously constricted, and often flattened or depressed around the strand connections. The sections reveal a distinctly cellular structure, usually with two layers of smaller thick-walled epidermal cells, whereas the cells toward the center are parenchyma-like, with thin walls and of irregular shape.

Additional histological and morphological studies of sclerotia formation by the root-rot fungus, and infection experiments with cultures originating from the sclerotia obtained from the soil are in progress.

The function of the sclerotia as the hold-over stage of the fungus is indicated. The first sclerotia produced in cultures at Greenville were recorded on January 3, 1929, and are still viable in July, 1929, after remaining seven months in the incubator.

SUMMARY

Viable sclerotia of the cotton root-rot fungus, *Phymatotrichum omnivorum*, have been found under natural conditions in the soil of infested cotton fields in Texas, showing that this fungus is a soil organism not restricted to living roots of susceptible host plants, but having an independent means of over-wintering and dissemination.

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