accurate; it falls, in many of the tests, between the tenth and the fifteenth day following the first instillation, or between the seventh and the tenth day after the last instillation.

The striking precision of the phenomenon may have an important bearing on the still discussed question of the mode of infection in epidemic poliomyelitis. No other means of producing the infection in monkeys gives a corresponding, regular result; and no other experimental method of inoculation, through an external portal, gives reliable results at all. The digestive organs in monkeys are impervious to the virus, or the virus penetrating the tract is destroyed quickly. Not only is infection almost never produced by artificial feedings of virus, but monkeys which have resisted repeated artificial feedings are found to be as susceptible as normal or control animals to the nasal instillations.

The regularity and simultaneity with which experimental infection can be induced by dropping the virus of poliomyelitis into the nares of monkeys, afford additional support for the view that the portal of entry of the virus in human beings is the upper respiratory mucous membrane.

SIMON FLEXNER

THE ROCKEFELLER INSTITUTE FOR MEDICAL RESEARCH NEW YORK, N. Y.

PRIMITIVE OR FILTERABLE FORMS OF BACTERIA

IN a previous note¹ we reported the general occurrence of filterable forms of bacteria in such substances as soils, hay infusions, decomposing manure, human feces and milk. More recently, Sherman, Safford and Brueckner² have recorded a quantitative approach to this subject, which indicates that these primitive forms of bacteria are usually present in milk and milk products in greater numbers than are bacteria of familiar type.

The quantitative method in use in this laboratory was based on the assumption, later shown to be correct, that these primitive forms should occur in many substances in greater number than ordinary bacteria which are detected and enumerated by conventional methods. Serial dilutions of the substance under examination are made into glucose-beef infusion broth. These dilution cultures, representing from 10^{-1} to 10^{-13} gram of the original material, are incubated one or two days at 37° C. and then from two to three weeks at 30° C. After incubation, the higher dilutions, which contain no growth of ordinary bacteria, are seeded on the surface of glucose-infusion agar

¹ SCIENCE, 73, 448, 1931.

² Proc. Inter. Dairy Congress, Copenhagen, July, 1931.

plates. The plates are incubated two days at 37° C. and, if necessary, longer at 30° C., after which they are examined for the delicate growth which Hadley has termed the "G" type colony. Growth has been recorded as positive when on microscopic study, after further culturing if necessary, definite minute cells which would be recognized as "bacteria" were found.

Aside from the quantitative approach, the most significant modification in this technique as compared with those previously used by Hauduroy,³ Hadley, Delves and Klimek,⁴ and others, is the longer incubation of the broth cultures. This prolonged incubation in broth appears to be desirable in order to get definite growth on the first agar plates seeded therefrom.

The numbers of primitive bacteria found by the application of this method are somewhat surprising. Nine samples of soil representing a wide range of productivity have yielded estimates ranging from 10^7 to 10^{12} per gram. Other materials have shown: Fresh human feees, two samples, 10^{12} ; sour milk, two samples, 10^{11} ; and raw market milk, six samples, 10^8 to 10^{12} per gram.

While the occurrence of these primitive microorganisms in such large numbers may be difficult to believe in the light of previous knowledge of "bacterial counts," they are perhaps not unreasonable. Ordinary bacteria are not infrequently found in numbers approaching and exceeding one billion per gram in certain types of decomposing and fermenting materials. When probable relative sizes are considered, the presence of bacteria in the filterable stage in numbers approximating one trillion per gram appears plausible. In this connection it is of interest to note that Clifton, Schultz and Gebhardt⁵ have recently determined the size of the virus of poliomyelitis as probably less than "50 µµ in diameter."

With reference to the microscopically definite cells which are found in "G" type cultures, it should be remembered that workers in this field appear to be unanimous in the belief that these organisms represent a partially transformed state between the filterable (perhaps ultramicroscopic) and the non-filterable stages of the bacteria. Hadley, Delves and Klimek⁴ have furnished rather definite evidence that this is in fact the case.

One of the most interesting points about the primitive forms of bacteria is their inertness on ordinary bacteriological media. When these forms are cultivated in the laboratory until they appear microscopically as true bacteria, they still make only very meager growth on agar and do not cause the familiar

³ 'Les ultravirus et les formes filtrantes des microbes,'' Paris, 1929.

⁴ Jour. Infect. Diseases, 48, 1, 1931. ⁵ Jour. Bact., 22, 7, 1931.

changes in culture media which are characteristic of bacteria of ordinary type. In other words, it appears that morphological reversion runs very much ahead of physiological transformation. This has been noted by other workers. It apparently was the case in the excellent work of Smith and Jordan⁶ on the diphtheria organism. Dr. Hadley informs us (personal correspondence) that it has been true in all the work which he has done on this subject. Kendall,⁷ in his recent important contribution to this field of study, reports the same observation. In their exceptionally complete work on the Shiga bacillus, Hadley, Delves and Klimek⁴ succeeded in accomplishing a total physiological transformation of the organism through its various stages to the "adult" form of the laboratory culture with which the experiments were started. In this connection we wish to report that some of our cultures which have been obtained from the filtrates of various substances, after several months' culture in the laboratory, ferment sugar broths and give physiological tests in other media simulating those obtained with ordinary bacteria.

> J. M. Sherman, C. E. Safford

CORNELL UNIVERSITY

DIAMAGNETISM IN METAL CRYSTALS

ONE of the fundamental problems in the study of the constitution of solid metals is to find the cause of the magnetic susceptibility due to crystalline state, which property is for most crystals very different from the atomic susceptibility of the metal. As an example tin (Sn) may be mentioned, which is diamagnetic in the liquid state, paramagnetic in the "white" (tetragonal), and diamagnetic in the "gray" (hexagonal) modification. Hence, one must conclude that the magnetic properties of a metal crystal depend mostly on an action caused by the coexistence of a number of molecules (atoms) in a given geometric configuration.

One fashion to approach this problem in a way which gives reliable measurements is the study of the magnetic susceptibility in anisotropic metal single crystals in different directions with regard to the crystal. After the magnetic constants of a pure crystal are known, a small number of atoms of another metal are added to the original substance, and a new single crystal of the same orientation is produced. For most of the measurements bismuth (Bi) was used, since its crystalline susceptibility is 15 to 20 times larger than its atomic magnetism; it is, furthermore, one of the most diamagnetic substances known.

The results obtained concerning the influence of

foreign atoms on the magnetic properties of the crystal are the following:

(1) The presence of foreign atoms affects the magnetic properties of the crystal only if the atom goes into a solid solution. Inclosures or occlusion of heterogeneous substances do not affect the susceptibility. Thus, all metals which can not be dissolved in f. i. bismuth do not change its magnetic properties appreciably.

(2) If a soluble metal (Sn, Pb) is added in a quantity below the limit of saturation (which is very low—0.5 per cent. to 3 per cent.—for different enantiomorphous metals) the effect on the susceptibility is very large. Beyond the saturation the effect due to enclosures of eutectic mixtures is negligible.

(3) The effect of dissolved foreign atoms is surprisingly large and affects the crystal differently in different directions. (An atomic concentration of 1:10,000 alone changes the susceptibility several percent.)

(4) The influence calculated per added atom within the solubility limits in the crystal depends on their number, *i.e.*, the first few atoms have an effect which may be 100 fold larger than the atomic effect for larger concentrations.

(5) The dependence of the susceptibility of the crystal on the temperature is changed very much by foreign atoms such as to cause a large decrease with decreasing temperature. The decrease is different in different directions.

The effect mentioned under (3) works in all cases investigated as to *increase* the anisotropy of the crystal, *i.e.*, the ratio of the susceptibilities normal and parallel to the axis becomes larger due to the fact that the diamagnetism parallel to the axis decreases. This change is more distinct at lower temperatures, and it is thus possible to obtain a crystal saturated with 3 per cent. Sn which is below 270° K. paramagnetic parallel to the axis and diamagnetic normal to it. Since the x-ray analysis of such crystals does not show any difference from the normal Bi-crystal, it is evident that the atomic complexes within the lattice responsible for the diamagnetism must be of much larger sizes than the wave-lengths of the x-rays used.

To account for the exceedingly small amount of foreign atoms sufficient to influence the crystal diamagnetism it seems necessary to accept one of the two alternative conclusions:

(1) The distortion due to a foreign atom dissolved within the crystal lattice reaches very far (somewhat like 25 crystal-atoms in each direction.

(2) The foreign atoms are absorbed in discrete layers within the crystal, the total effect thus being due to an internal surface phenomenon.

The former assumption, of far-reaching influence in

⁶ Jour. Bact., 20, 25, 1930.

⁷ SCIENCE, 74, 129 and 196, 1931.