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Atypical Acidfast Organisms.

MAX PINNER.

From the Laboratories, Desert Sanatorium and Institute of Research, Tucson, Ariz.

A number of acidfast organisms has been observed in this laboratory during the last year. As far as could be ascertained by a search of the literature, they have not been described. They fall into 3 distinctly different groups.

Group I. The organisms in this group are air contaminators. These colonies, on various agars, are round, transparent or creamy white, with a diameter of 0.01-1.0 mm. All strains show constantly smooth colonies, with the exception of one which is constantly rough. On smears they are diphtheroid-like, very pleomorphic rods and cocci. The rods are frequently granulated, beaded, barred or clubbed. The cocci vary in size from barely visible granules to coccoidal or diplococcoidal forms of the size of staphylococci. Frequently large single cocci are seen. With the Ziehl-Neelsen method only a part of each smear retains the carbolfuchsin. The relative proportion of the acidfast elements varies greatly with the different strains and with the age of the cultures. Acidfastness is enhanced by growth on egg media. Growth occurs within 24 hours on the usual agar media, in the usual broth media and on tubercle bacilli media; but the addition of dyes, commonly used for the cultivation of tubercle bacilli (gentian violet, malachite green) inhibits growth. In broth, growth occurs throughout, either by even clouding, or slightly granular. All smooth strains are poor fermenters, forming acid but no gas in dextrose only, or not at all. The rough strain ferments several sugars and liquifies gelatine. All strains are completely avirulent for guinea pigs and rabbits, even in massive and repeated doses.

Group II. This group consists of 6 strains, isolated from human material (sputum or urine) or from known strains of human tubercle bacilli. Their outstanding characteristic is their completely smooth, unpigmented colony formation. The colonies are white and glistening, having the appearance of drops of cream, a consistency of butter or cream cheese. They are easily suspended in water. Varying with the strains, the smears, stained by the Ziehl-Neelsen method, show acidfast rods, indistinguishable from typical tubercle bacilli, or mixed with such rods, tiny acidfast and non-acidfast

granules. They grow more rapidly than true tubercle bacilli, some develop visible colonies in 18 to 24 hours. Their growth is luxurious on the usual tubercle bacillus media, but the most rapidly growing strain prefers dextrose to glycerole. On subcutaneous injection in large dosage into guinea pigs, they cause the formation of local abscesses, which have a tendency to spontaneous healing. In addition, strictly localized lesions may occur in the various lymph nodes, in the liver, spleen, lung, omentum, testes and peritoneum. All these lesions—tubercular in gross appearance—have a marked tendency to spontaneous regression. Histologically they consist of a granulation tissue which simulates closely the early phases of tuberculosis; caseation, as a rule, is absent, but abscess formation occurs, particularly in the local lesion at the site of injection. During the first few weeks following the infection, numerous acidfast rods are present in the foci, and can usually be isolated from them in pure cultures; later on, they disappear, and cultures remain sterile more frequently.

It is believed that these strains are probably smooth human tubercle bacilli. Two of these strains were isolated from known tuberculous material, one was found in association with typical rough tubercle bacilli. The other 4 strains were derived directly from typical, known strains of human tubercle bacilli. One strain has on several occasions, reverted to a rough strain following animal passage.

Group III. This group consists of 11 strains, all of which were isolated from human secretions or excretions. Four were sent to me by Dr. J. F. Norton, Detroit, and one by Dr. H. S. Willis, Northville, Mich. These organisms have smooth, glistening, round colonies which show strong lemon yellow to orange pigment. Their color is similar to that of *B. phlei* or of the bacillus of rat leprosy. The consistency is soft creamy to viscous. The individual organisms are acid- and alcohol-fast rods, somewhat larger than tubercle bacilli; they are frequently beaded and granular. On subcutaneous injection they produce lesions in guinea pigs, which in gross appearance and distribution are not unlike those in Group II. They cause frequently perisplenitis, perihepatitis and intestinal adhesions. The lesions are self-healing. Histologically, the foci consist of a non-specific, non-caseating granulation tissue, composed of strikingly polymorphous cells. At times, the center of the focus is an abscess. Acidfast rods are found in great abundance in the lesions before regression sets in. Following reinfection with the homologous strain—2 to 4 weeks after the primary infection—visceral

lesions are much more abundant than after a single infection. Although the infected animals become skin sensitive to tuberculin and to the homologous strain, reinfection does not produce a local allergic reaction. The local nodule which develops promptly on primary infection, frequently fails to develop at the site of reinfection. The pathogenic action of these strains has been compared with that of known non-pathogenic acidfast bacilli, namely, one pigmented acidfast rod isolated from tap water and *B. phlei.* Both these strains produce a similar yellow pigment, but their colonies are rough and dry. Massive doses of these 2 strains produced abscesses at the site of injection, but never any visceral lesions.

The 11 strains of this group are apparently not identical; they vary in growth intensity, in pigment, in their growth on liquid media and in their pathogenic action. Whether they have any significance in human pathology remains to be studied.

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Observations on Various Insulin Mixtures Administered Per Os.

ELMER S. GAIS. (Introduced by L. Gross.)

From the Laboratories of the Mt. Sinai Hospital, New York.

At the time of publication of Stephan's paper[1] on the use of "cholosulin", a desoxycholic acid-insulin mixture, we were engaged in the preparation and study of a similar compound based on identical theoretical considerations. Mixtures of desoxycholic acid and insulin were prepared and administered by stomach tube to fasting rabbits, the experiments controlled by subcutaneous injection. The results in 4 experiments with 4 rabbits each, were inconclusive. The blood sugar curves closely paralleled the controls. The recent reports of Bronkhorst, Freud and Laquer,[2] and Wahncau and Bertram,[3] seem to show some slight effect on carbohydrate metabolism by this compound, but they ascribe it to the bile acid and not the insulin in the mixture. The clinical use of Stephan's preparation by Umber and Rosenberg,[4] and others has not proven successful. Our mixture also had no effect on human diabetes.

[1] Stephan, R., *Munch. Med. Woch.*, 1929, **76**, 1579; *Med. Klin.*, 1930, **26**, 228.

[2] Bronkhorst, A. J., Freud, J., de Jongh, S. W., and Laequer, E., *Nederl. Tijdschr. v. Geneesk*, 1930, **74**, 2185; abs. *Endokrinol.*, 1931, **8**, 43.

[3] Wahncau, E., and Bertram, F., *Klin. Woch.*, 1931, **10**, 486.

[4] Umber, F., and Rosenberg, M., *Deut. Med. Woch.*, 1930, **56**, 169.