# PROCEEDINGS OF LOCAL BRANCHES OF THE SOCIETY OF AMERICAN BACTERIOLOGISTS

## EASTERN PENNSYLVANIA CHAPTER

PHILADELPHIA COUNTY MEDICAL SOCIETY BUILDING, JANUARY 26, 1937

ESSENTIAL IMMUNIZING ANTIGEN OF PNEUMOCOCCI. *Lloyd D. Felton*, The Johns Hopkins University, Department of Pathology and Bacteriology, Baltimore, Md.

The essential immunizing antigen of the pneumococcus was defined as that fraction of the cell which contained as many as, or more, immunizing doses, than the original cell from which the fraction was derived. Ten grams of Type I pneumococci dried from acetone were separated into five successive fractions: (A) acetone-soluble constituent of the cell; (B) HCl-insoluble at pH 3; (C) HCl-soluble, insoluble with 2 volumes of alcohol; (D) acid-alcohol-soluble, insoluble on neutralization; and (E) alcohol-neutral-soluble fraction. (A) showed no active immunizing value for white mice, a confirmation of Wadsworth's work. (B) contained active immunizing antigen, but because it was composed of many undissolved cells, this fraction was eliminated from consideration. (C) when dried (0.607 gram) immunized mice in dilution of 1:50,000,000, had a precipitin titer of 1:5,000,000, contained 12 per cent hydrolyzable sugar, and 8.3 per cent nitrogen. (D) gave a precipitate (0.439 gram) with immunizing titer of 1:50,000 and precipitin titer of 1:80,000. (E) on evaporation gave a slightly positive Molisch reaction, no precipitinogens, and failed to immunize white mice.

The original cells (10 grams) immunized mice in 1:1,000,000 dilution (0.5 cc. dose). Therefore, there was a total of 10,000,000 immunizing units. The acid-alcohol-insoluble fraction (0.607 gram) immunized mice in dilution 1:50,000,000 and thus contained 30,000,000 immunizing doses (0.607 × 50,000,000). Hence, disregarding the slight immunizing activity of the other fractions, there were found at least three times as many immunizing doses as in the original cells. This fraction, polysaccharide in nature, was not a pure substance.

Another 23 grams of 16-hour growth, fractionated in the same way, yielded 2.21 grams of this same C fraction. The total number of units in the cells was 11,500,000 and in the C fraction 110,000,000. The 48-hour growth (17 grams) contained 8500 immunizing doses, and the 0.190 gram isolated contained 1,900,000 immunizing doses. These experiments were conducted with Type I organisms. It has been found repeatedly that Type II cells act in the same manner.

This fraction produces specific immunity in mice, and heterologous immunity in human beings. However, without purification it may give very severe reactions in human beings.

## NEW YORK CITY BRANCH

TEACHERS COLLEGE, COLUMBIA UNIVERSITY, FEBRUARY 9, 1937

THE DIFFERENTIATION AND DIAGNOSTIC CHARACTERISTICS OF FOUR FUNGI CAUSING SYSTEMIC INFECTIONS. *Rhoda W. Benham.*

Under the name blastomycosis have been grouped a number of the deep-seated fungus infections. This has led to a great deal of confusion as many of these conditions are quite distinct both clinically and etiologically. This can be shown by study of the fungi concerned.

1. American blastomycosis: In this condition the causative organism *Blastomyces dermatitidis* appears in the lesion or in pus from the lesion as spherical budding cells with refractile walls. In culture a white, filamentous growth forms, with large round or pear-shaped spores borne singly on the hyphae, either sessile or on short side branches.

2. Cryptococcosis (European blastomycosis or torula infection): In this disease the organism *Cryptococcus hominis* appears in the lesion as budding cells with wide gelatinous capsules. In culture, budding cells likewise surrounded by a capsule are found. The culture is of a moist, mucoid consistency.

3. Coccidiodal granuloma: The organism *Coccidiodes imitis* forms in tissue large spherical, thick-walled cells which reproduce by endospores. There is no budding. In culture a white mycelial growth forms. There are no spores other than chlamydospores.

4. Meningeal moniliasis: The organism, *Monilia albicans* forms in the lesion budding cells, and elongated forms suggesting mycelia. A pasty yeast-like growth occurs. On suitable media the characteristic mycelium with clusters of budding cells and chlamydospores is found.

Each of these conditions can be correctly diagnosed by the characteristic picture of the organism in the diseased tissue. In the case of "1" and "3" animal inoculation is the most reliable means of diagnosis as the cultures are easily confused.

RECENT EXPERIMENTS ON ANTI-RABIC VACCINATION. *L. T. Webster,* Rockefeller Institute, New York City.

THE INTRACELLULAR CRYSTALLIZATION OF STANLEY'S TOBACCO VIRUS PROTEINS. *H. P. Beale,* of Boyce Thompson Institute for Plant Research, Inc., Yonkers, New York.

A microscopic examination of plants systematically infected with ordinary tobacco mosaic virus discloses the presence of large crystalline substances deposited chiefly on the external layers of host tissue. The crystalline material is confined to the chlorotic areas of the leaf and is not present in the green areas of diseased leaves nor in healthy leaves. The crystalline inclusions appear to be specific for tobacco mosaic disease and occur as plates, definitely hexagonal at times, in side view oblong.

Pieces of epidermal tissue are stripped off the back of the midrib of diseased leaves and mounted in water. A few drops of a saturated solution of magnesium sulphate or dilute acid, such as hydrochloric, sulphuric, acetic, or nitric, are run under the cover slip and the crystalline material is gradually transformed into a mass of needle crystals, bearing a striking resemblance to Stanley's needle crystals,

obtained by a similar salting out or acidification of purified, concentrated virus extract. The intracellular crystals are denatured by an acidity greater than about pH 1 or an alkalinity in excess of about pH 11.8. Recrystallization can not be induced by readjusting the reaction of the host cell to neutral. Stanley's crystalline tobacco protein becomes denatured when subjected to the same limits of H-ion concentration (Science, **81**: 644–645. 1935).

It is concluded from these observations that the intracellular crystalline deposits associated with tobacco mosaic disease are the source of Stanley's crystalline proteins obtained from tobacco virus extract.