loss of fish became apparent, numbers of dead fish being observed around the edge of the affected areas.

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PATHOGENICITY AND VIRULENCE OF CERTAIN BACTERIA

INCIDENTAL to studies on the egg-propagation of certain filtrable viruses as previously reported[1, 2] opportunity was afforded for observing the effect of introducing several species of bacteria into developing eggs of the chicken and other domesticated fowl. At the outset the sensitivity of the embryo and its membranes to various filtrable viruses and to numerous toxic influences[3, 4, 5, 6] was reflected in the response to the injection of different concentrations of *Salmonella pullorum*. With given strains of freshly isolated *S. pullorum* the extent and severity of the lesion produced as well as the survival time of the embryo were quite definitely and uniformly correlated with the quantity of the inoculum and the virulence of the culture for baby chicks.

These results suggested the possible adaptability of the method of egg inoculation for determining the pathogenicity and for ascertaining the virulence of various strains and species of bacteria. In investigating this hypothesis the preliminary observations here recorded were confined to inoculations upon the chorio-allantoic membrane of eggs incubated 10 to 15 days prior to treatment. The cultures used represented 4 strains of *Brucella abortus,* var. *bovis* and *suis,* 3 strains of diplococci of equine origin, 3 strains of hemophilic bacteria isolated from the upper respiratory tract of young chickens, 8 species of Salmonella and 3 species of Pasteurella. Eggs employed as controls were injected with the sterile suspending medium or with suspensions of the various bacteria killed by heat. Proof of infection of the embryo and/or its membranes was established by the production of gross lesions and direct pure culture isolation of the organism inoculated.

The cultures of diplococci and hemophiles appeared virtually devoid of pathogenicity for the developing egg even in the relatively large quantities employed (as much as 0.2 cc of the undiluted 15 to 24-hour broth cultures). These 2 groups of organisms in other trials were not proved to possess specific pathogenic properties for the homologous host. Brucella, Pasteurella and Salmonella cultures were lethal to the embryo in very dilute concentrations, while the control suspensions of dead organisms produced no more than slight local injury to the extra-embryonic tissues and were seldom associated with the death of the embryo.

Marked differences in virulence for developing eggs were manifested between smooth stock and freshly isolated cultures of Salmonella and Pasteurella. Simultaneous comparative titrations with Pasteurella cultures on 2 to 10-day old chicks revealed a correlation in results, although much less definite and uniform than in the egg-inoculation method. The *P. avicida* culture, when inoculated subcutaneously, killed chicks in dosages 10^7 times smaller than were required with the *P. equiseptica,* while *P. cuniculicida* required larger doses than *P. equiseptica.* For eggs the *P. equiseptica* and *P. cuniculicida* required dosages approximately 10^4 times greater than did *P. avicida* to kill chicken embryos within 48 hours. Intracranial inoculations of the *P. avicida* and *P. equiseptia* strains into a group of 9 horses gave results which could be interpreted as validating the titrations upon eggs and chicks. However, the more uniform and accurate measurements of virulence obtained by egg inoculation as compared to animal inoculation emphasizes the superiority of the new method.

The delicacy with which differences of pathogenicity and/or of virulence among strains of certain bacteria may be determined by inoculating the developing avian egg suggest that this method may also be utilized to detect alterations in these characters among variants of a certain strain.

The potential value and adaptability of the developing avian egg for other phases of purely bacteriological investigation and experimentation is suggested by the findings here reported and in consequence of the simplicity of application and economy of the method.

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DOWNWARD SHIFT OF pH CAUSED BY ADDITION OF GLUCOSE TO BORIC ACID BUFFER SOLUTIONS

THE accompanying table was prepared for use in a study of O_2 consumption by yeast. It may be useful in other studies. In each test, to 20 ml of boric acid buffer solution (prepared according to Clark: "Determination of H ions," 2d ed., 1928; table 35) was added a known weight of glucose, as shown, and the resultant pH value (at 25°) was measured potentiometrically (quinhydrone electrode).

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