

for brucellosis. Although it is well known that proven cases of the disease may at times present little or no agglutinin response, the diagnosis of brucellosis is frequently made on the basis of low-grade titers without adequate supporting data. Many clinicians consider brucella agglutinin titers in dilutions of 1:500 as almost certainly diagnostic of active infection. The development of titers as high as this following vaccination for cholera may well be expected to introduce further confusion in the clinical interpretation of this test. Cholera vaccination has been infrequently used in this country prior to the war; but large numbers of veterans are returning from the Pacific and Asiatic

theaters who have been subjected to this immunization.

Studies are underway to determine the duration of the persistence of brucella antibodies following vaccination for cholera, and the possible reappearance of antibodies in the circulating blood as an anamnestic phenomenon. The possible effect of other types of immunizations on brucella antibody stimulation must also be considered.

Our preliminary data presented here indicate that there is an H antigen in *Vibrio comma* which is present also in brucella. Studies are in progress to elucidate further the antigenic interrelationships of these organisms.

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Excitability of Muscle to Direct and Indirect Stimulation During Prolonged Direct Stimulation.*

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Although electrical stimulation is used widely in diagnosis and in physical therapy and is the only practical procedure to test muscle function in animals, the effects of prolonged direct stimulation have not been studied extensively.

Alternating 60-cycle current was used in 3 independent but *in phase* circuits for stimulation through 3 pairs of electrodes, the first pair being inserted into the knee and tendon of the gastrocnemius muscle *in situ* of anesthetized rats (nembutal) and cats (dial), (D_1 electrodes); the second pair (D_2) inserted into the muscle itself at its upper and lower ends; the third pair (I) being employed for stimulation of the peripheral end of the cut sciatic nerve. The strength of the stimulus was adjusted with a rheostat and measured by a volt meter; in addition, the resistance in the muscle was measured by means of an oscilloscope in a shunt parallel to the muscle. Stimulation was continued either with D_1 or with D_2

until the muscle length reached the pre-stimulus resting value. During and after the continued stimulation, brief tetanic stimuli were applied at regular intervals through the 2 other pairs of electrodes. In all cases maximum stimuli were applied through a 10,000-ohm resistance placed in series with muscle and nerve. The muscle was loaded with 200 g and contraction was isotonic. Before continued stimulation the effect of summation was tested for all combinations of D_1 , D_2 , and I. There was no summation between D_1 and I, or D_2 and I, and a tendency to slight summation of $D_1 + D_2$.

Steel, platinum, silver, and nonpolarizable ($\text{Cu-CuSO}_{41.0M} - \text{K}_2\text{SO}_{4\text{sat.}} - \text{NaCl}_{\text{sat.}}$ $\text{NaCl}_{0.9\%}$ -Tissue) electrodes were employed in various experiments. There was no essential difference in the results to be reported. Forty-seven experiments were performed in rats and 3 experiments in cats.

With continued stimulation through D_1 , there is no response to D_2 or I at the very beginning, but after a few minutes a marked

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response to D_2 appears, while response to I is absent. The absence of response to I cannot be due to the absence of muscle fibers available for stimulation, since response to D_2 is present. After 10 to 20 minutes of stimulation with D_1 , response to I begins to reappear and to increase gradually in size, but usually remains somewhat smaller than the D_2 response. After "fatigue" (*i.e.*, approach to the original resting length) the responses to both I and D_2 are very marked and may approach the height of initial contraction. If at this stage continued stimulation through D_2 or I is superimposed on the uninterrupted D_1 stimulation, a large tetanic contraction occurs, which however returns to the baseline much faster than it would have had the muscle been stimulated solely through these electrodes. If the stimulation of D_2 or I is interrupted at this moment, while D_1 stimulation continues, the response to D_2 or I stimulation recovers.

When continued stimulation is applied through D_2 , the responses both to D_1 and I stimulation appear within a short time and are present throughout the development of fatigue.

The continued stimulation through D_1 is

accompanied by an increase in muscle resistance, as indicated by the oscilloscope studies. This increased resistance is related in time of appearance to the return of response to indirect stimulation. The effect of D_2 stimulation was less marked. Investigations by means of crossed D_1 and D_2 electrodes revealed that the difference between continued D_1 and D_2 stimulation is due to the lower D_1 electrode inserted into the tendon.

The results described above could be reproduced in all experiments with only one exception. Experiments on cats were confirmatory. It appears doubtful whether the diminishing response to continued direct stimulation may be regarded as a simple fatigue phenomenon. Rather it appears more likely to be due to some kind of rectification or polarization in the tissues, however, not to polarization of the electrodes themselves.

The more intimate mechanism of these effects is being investigated. The effects themselves appear to have some practical significance in connection with the therapeutic use of alternating currents in muscle stimulation. They may also be of importance in relation to fundamental aspects of excitability and fatigue in striated muscle.

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"Folic Acid" Active Compounds in the Nutrition of the Monkey.*

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Two years ago we reported¹ that the norite eluate concentrate of Hutchings *et al.*,² a rich source of "folic acid," was active in producing normal growth and preventing leucopenia in monkeys given a synthetic ration containing all the B vitamins available in pure form. Hutchings *et al.*³ had shown previously that this concentrate contained factors necessary

for the normal development of chicks. More recently Briggs *et al.*⁴ concluded that the norite eluate contains at least 3 factors: folic acid necessary for the growth of *Lactobacillus casei*; vitamin B₁₀, necessary for normal feathering; and vitamin B₁₁, necessary for normal growth in chicks. They⁵ further demonstrated that fractions high in vitamin B₁₀ and

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