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A STUDY OF THE VIRULENCE OF THE BRUCELLA VACCINE STRAIN
FOLLOWING ITS PRESENCE IN AN IRRADIATED ORGANISM

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A STUDY OF THE VIRULENCE OF THE BRUCELLA VACCINE STRAIN
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Following is the translation of an article by Z. V. Shevtsova, Gamaleyn Institute of Epidemiology and Microbiology, AMN, USSR, published in the Russian-language periodical Zhurnal Mikrobiologii, Epidemiologii i Immunobiologii (Journal of Microbiology, Epidemiology and Immunobiology), No 3, 1965, pages 56--58. It was submitted on 30 Apr 1964. Translation performed by Sp/7 Charles T. Ostertag Jr.

Up until the present time the question concerning the feasibility of an increase of virulence of microbes in an irradiated organism is controversial. Klemparskaya and co-authors (1958) consider that under the influence of ionizing radiation conditions are created in an organism which further an increase of the virulent properties of microorganisms. Chekatilo (1958) observed changes in the biological properties of typhoid microbes following residence in the organism of irradiated guinea pigs (200--300 r). On the basis of data obtained the author makes the conclusion that the increase in virulence is directly proportional to the severity of radiation sickness and the period of residence in the irradiated organism. Other authors (Bond et al., 1952, Silverman, et al., 1954, Sviridova, 1958) were not able to detect changes in the virulent properties of microbes isolated from irradiated animals. The resolution of this problem acquires great significance when utilizing live vaccine strains with a reduced virulence for the immunization of an irradiated organism.

In connection with this, when studying the effectiveness of immunization with live brucella vaccine from the 19-BA *Brucella abortus* strain under conditions of a radiation influence, we considered it necessary to clear up whether or not the virulent properties of a brucella vaccine strain changed as a result of residence in the organism of irradiated animals.

Guinea pigs (weighing 280--300 grams) were irradiated on a RUM-3 duplex unit with a uniform field of 30 x 40 cm at a voltage of 180 kilovolts, power of current 15 milliamperes, filters of 0.5 mm Cu and 1 mm Al; the dosage rate in the air over the entire irradiated field equalled 42 r/min; an irradiation dose of 200 r caused a lethality of 20--30% of the animals taken in the test (30 days of observation). The guinea pigs were immunized subcutaneously (1 billion microbial cells) on the 3rd day following irradiation and destroyed for the purpose of investigating for the presence of the culture on the 3, 10, 20, and 30th days following the administration of the vaccine, that is, on the 6, 13, 23, and 33rd days following irradiation. In this manner the first 10 cultures were obtained from animals which were found in an acute stage of

radiation sickness (8th and 13th day), and the other 15 cultures -- during the period of fading away (23rd and 33rd day). It must be stressed that the last 15 cultures were found in the organism of guinea pigs throughout the entire acute period of radiation sickness. In selecting these periods we were guided by the data of Klemparskaya and Chekatilo, in whose tests the virulence of typhoid bacteria increased along with the increase in the time of their residence in the organism of the irradiated animals. Some cultures were obtained from animals which died in the corresponding periods.

Since even the most virulent cultures of brucella, as a rule, do not cause the death of animals, a determination of the lethal dose cannot be the criterium of their virulence. At the present time it has been accepted to study the virulence of brucella by means of determining the minimum infecting dose which causes a generalized process. Considered as virulent are those strains of the bovine type of brucella which, following the administration of 10--100 microbial cells, cause a generalized infection in experimental animals (guinea pigs, white mice). The vaccine strain used by us, even in a dose of 100,000 microbial cells, caused a weakly expressed and rapidly passing (by the 30th day) generalization and belonged to the cultures with a lowered virulence. For infection we used two doses -- 100 and 1,000 microbial cells, which did not cause a generalized process, but in the event of increased virulence would cause generalization. Each dose was administered to 3 fresh guinea pigs. For purposes of control, the initial vaccine strain was administered to nonirradiated guinea pigs, in whose organisms it was found over the same periods of time as the experimental cultures. All told 29 cultures were checked (on 174 guinea pigs). Of these, 25 were obtained from irradiated and 4 from control animals. The cultures isolated from the animals preserved the initial morphological, cultural and antigenic properties.

The capacity for spreading in the 5 cultures obtained after a 3-day residence in an irradiated organism did not differ essentially from the analogous properties of the control strain, passaged through the nonirradiated organism of a guinea pig; in a dose of 100 microbial cells these cultures caused a variable regional process, and in a dose of 1,000 microbial cells the brucella were found in the regional or distant lymph nodes.

Of the 5 cultures investigated following a 10-day residence in an irradiated organism, 2 were obtained from guinea pigs which had died (No 13 and No 14). In a dose of 1,000 microbial cells these cultures caused a generalized process in one out of three guinea pigs. However, the generalization was weak (the culture was isolated only from the spleen). Based on the type of process caused, the other three cultures did not differ from the control.

Of the five cultures isolated from the irradiated organism after 20 days following administration, two were also obtained from animals which had died (No 15 and 16), and based on the capacity for spreading in the organism were no different from the initial strain. The remaining 3 cultures also did not essentially differ from the control strain; weak generalization was noted (growth in broth with a seeding from the spleen) in one out of three pigs

following the administration of one culture (No 19) in a dose of 1,000 microbial cells. In checking the 10 cultures obtained after 30 days following infection, two caused a weak generalized process (growth in broth with a seeding from the spleen) in two pigs out of 18.

In this manner, based on the capacity for spreading in the organism of 20 cultures, checked after 3, 10, 20 and 30 days residence in an irradiated organism, they essentially do not differ from the original strain.

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