several days. Type A, B, and AB cells, strongly sensitized in a saline eluate, react in the usual manner with α and β isoagglutinins as specifically and intensely as prior to their being sensitized. Rh+ cells similarly sensitized likewise are not affected in the reactivity with Rh agglutinins. This is interpreted as showing that the A. G. antibody does not block the Rh and major blood-group receptors. This antibody is not reduced in potency at exposure to 70°C. for 10 minutes, but cannot be demonstrated by the indirect developing test after exposure of the serum to 80°C. for 10 minutes. As shown in the above tables, (in distinction to the Rh antibody) this antibody reacts with red cells irrespective of Rh type, is also an autoantibody and a panantibody, but does not react with rhesus cells.

Hematological, histological, and clinical data, as well as further immunological studies concerning these three patients will be submitted in greater detail in a future publication.

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Further Observations on Leptospirosis in Micronesia

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Murine leptospirosis has given rise to a public health problem in many parts of the world (*2*) because the etiologic agent, *Leptospira icterohemorrhagiae*, also causes Weil's disease in man. Since the United States now has control of many islands in the Pacific, it seemed advisable to start work of a survey nature on these islands to find out whether leptospirosis constitutes a menace to the native peoples and our own personnel. Such investigation was started during the summer of 1946, when the University of Hawaii, in cooperation with the Navy, sent a number of biologists into the field.

In a recent report by Alicata (*1*), covering a part of the first summer's work, it was pointed out that 5 out of 40 rats trapped on Moen and Ponape, in the eastern Carolines, were infected with leptospirae. It is the purpose of this communication to note survey findings on the island of Yap, in the western Carolines. Yap, it will be remembered, lies south and west of Guam, about 9 degrees north of the equator. Prior to World War II it was one of the territories mandated to Japan.

A number of traps were put out near the site of the military government unit at Yap Town. The 28 rats which were taken alive were members of 3 species, *Rattus alexandrinus*, *R. norvegicus*, and a third, tentatively identified by Harvey Fisher, of the University of Hawaii, as *R. exulans micronesiensis*. Each animal was killed by drowning the morning after being caught, and a piece of kidney was removed at autopsy immediately after death. The tissue was preserved in 10 per cent formalin for several months before being sectioned and stained by a modification of the Warthin-Starry silver precipitation technic.

Careful microscopic examination of several sections from each rat kidney failed to reveal any spirochetes in the urinary tubules or elsewhere. Although the number of animals is small, it would seem that murine leptospirosis is not present in the area studied. It may be that the geographic isolation of Yap and its lack of shipping facilities have prevented the introduction of *L. icterohemorrhagiae*. Once introduced into this region, which offers much rain and a large rat population, the organism might gain a foothold quickly.

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Triphenyltetrazolium Chloride as a Dye for Vital Tissues

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The use of triphenyltetrazolium chloride as a test reagent for seed germinability was brought to the attention of one of us (R. A. D.) while on a tour of duty in Germany in 1945 as scientific consultant for the Technical Industrial Intelligence Branch of the Joint Intelligence Objectives Agency. The use of this compound for predicting seed germination was based on its ability to stain only those parts of seed embryos which were capable of growth. This fact suggested that the tetrazolium salt might have a wider application as a test reagent for the vitality of tissues other than seeds.

Tetrazolium salts, including 2,3,5-triphenyltetrazolium chloride, were first prepared by Pechman and Runge (*5*) in 1894. In 1941 Kuhn and Jerchel (*2*) synthesized a number of tetrazolium salts by an improved procedure and called attention to the fact that dilute solutions of 5-methyl- and 5-hendecyl-2,3-diphenyl salts stained yeast, garden cress, and bacteria (*3*). These workers believed that the reduction of the colorless salt solutions to a red compound which dyed the plant tissues was not due to the presence of glutathione, ascorbic acid, or cysteine, for the latter substances did not reduce these salts below a pH of 9.0, whereas the characteristic reductions observed on yeast, garden cress, and bacteria took place in neutral solutions.

As a result of these studies Lakon (*4*) substituted triphenyltetrazolium chloride for the toxic sodium selenite in his "topographic method" for testing the germinating ability of seeds. By a comprehensive series of comparative staining and germination tests he was able to show that it is possible to predict the germinability of corn, oats, rye, wheat, and barley by observing the embryo parts which are stained by the red, insoluble formazan deposited in viable tissues. The unstained portions of the embryo were shown to be incapable of growth.

Porter, Durrell, and Romm (*6*) used Lakon's tetrazolium method and found a close agreement between the percentage of stained embryos and the percentage of normal sprouts obtained in standard germination tests with corn, wheat, rice, buckwheat, popcorn, soybeans, and Bahia grass. Less satisfactory agreement was found in a comparison of the two methods when applied to vetch and sorgo and to some lots of oats, peas, and barley.

Since tetrazolium salts were not available in this country, we synthesized the triphenyl compound and the 5-furfuryl-2,3-diphenyl derivative and have used these compounds in preliminary studies on various types of viable and nonviable