the fish. This loss, if there were no compensation from the tissues, would lower the blood chlorides only from ca. 440 mg% to 427 mg%—relatively a very small change, for both of these values lie within the range of normal blood chlorides for the goldfish. It is uncertain whether this constitutes sufficient depletion to activate the chloride-absorbing mechanism, but it is evident that once the absorption was begun, reducing the chloride concentration of the body (by dilution) tends to retard excretion of the ion rather than to accelerate absorption. Conversely, increasing the chloride concentration of the body fluids (by injecting NaCl) retards absorption but does not increase the rate of excretion.

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Transmission by Leaf Hoppers of the Virus Causing Phloem Necrosis of American Elm

W. L. BAKER[1]

Bureau of Entomology and Plant Quarantine, U. S. Department of Agriculture, Columbus, Ohio

Phloem necrosis, a virus disease, is one of the most destructive diseases affecting the American elm. Its origin is unknown, although observations and reports by Garman (*5*) and Forbes (*4*) indicate its presence in the Ohio River Valley as early as 1882. In recent years this disease has become epidemic in many sections and has destroyed thousands of valuable shade trees in such cities as Columbus and Dayton, Ohio; Peoria, Illinois; and St. Louis and Kansas City, Missouri. It has become widespread and is now known to occur in Ohio, Indiana, Illinois, Missouri, Iowa, Nebraska, Kansas, Oklahoma, Arkansas, Mississippi, Tennessee, Kentucky, and West Virginia (*1*).

In 1940 the Bureau of Entomology and Plant Quarantine, in cooperation with the Bureau of Plant Industry, Soils, and Agricultural Engineering, established a laboratory at Columbus, Ohio, for the purpose of studying the disease and determining the possible insect vectors of the virus. This paper reports briefly transmission studies with some of the insect species under investigation.

Late in August and early in September 1940, some adults of the leaf hopper genus *Erythroneura* were collected from elm in the Columbus area and confined for 4 days in cloth sleeves placed over the foliage of diseased elm trees. After this period of feeding the insects were divided into two lots and placed in two cloth-covered cages ($6' \times 6' \times 9'$), each of which contained 4 healthy elm seedlings of approximately $\frac{3}{4}''$ caliper. The insects were not disturbed again and were left to feed until they died. The cages containing the seedlings were kept covered during the active insect seasons through 1942. No disease symptoms having developed meanwhile, the cages were then removed and the trees left to grow unprotected thereafter. Each succeeding year the trees were examined for signs of disease. In August 1945, 3 of the 8 trees showed typical symptoms of phloem necrosis. Two of these died the same summer, and the third died early the following spring. The other 5 have remained healthy to date.

Further extensive tests with this group of leaf hoppers were established after the trees had developed symptoms of disease in 1945 and are now under way, but it will be some time before they will be completed. The results of the tests begun in 1940 are being reported, however, because they indicate that transmission was accomplished by insects in this genus. In addition to the three trees that died, only one tree among several thousand in the immediate vicinity has become diseased since 1940 without previous inoculation by diseased tissue grafting. The exception is a tree that grew in a cage adjoining the one in which two of the three died, the two cages having one cloth wall in common.

The *Erythroneura* specimens used in the tests begun in 1940 could not be recovered for identification, and the species involved remain unknown. More than one species probably was confined in each cage; yet the chances are that most of them were of the species *campora*, which is known to have been common in the collection area in 1940. This species occurs commonly on elm and doubtlessly is distributed throughout the disease region.

In early surveys of elm insects in the disease region several species of the leaf hopper genus *Scaphoideus* were collected. One of these, *S. luteolus* Van D., is a consistent inhabitant of elm. This species, although difficult to separate from closely allied ones in the adult stage, was found to differ markedly from others in the nymphal stages. This difference facilitated collection and permitted the establishment of a large series of transmission tests with this elm-inhabiting species from 1941 through 1943. The species was not used in tests in 1944, and only a few nymphs were used in 1945. In July 1946, however, a small number of nymphs were collected and, after they had fed for various periods on diseased elm foliage, were placed under test on healthy one-year-old American elm seedlings. On July 12, 2 seedlings were exposed to nymphs and adults that had fed the previous 9 days on diseased elm foliage; on July 15, 5 seedlings were exposed to nymphs that had fed the previous 12 days; and on July 26, 2 seedlings were exposed to nymphs and adults that had fed the previous 3 days. In all cases the infective insects were left on the test seedlings until the insects died. When all were dead, the seedlings were placed in a

[1] D. E. Parker supervised these investigations, H. T. Osborn conducted the laboratory tests prior to 1944, and R. U. Swingle, of the Bureau of Plant Industry, Soils, and Agricultural Engineering, gave valuable assistance in early diagnostic work.

cloth-covered cage and sprayed with DDT as an additional precaution against possible contamination by unwanted insects.

Symptoms of phloem necrosis are seldom visible prior to mid-June. In late June 1947, therefore, all test trees were checked routinely for the possible appearance of disease. At this time one of the two seedlings placed under test on July 12, 1946, showed typical symptoms of phloem necrosis, and by August 1 the foliage was dead. The seedling was then removed and examined. Near the ground line, in certain portions of the phloem still alive, the symptoms were pronounced. Before this seedling was discarded, three patches of bark were removed and grafted into three healthy seedlings to determine whether the virus could be transmitted from the test seedling. In October one of these seedlings showed early symptoms of phloem necrosis. This seedling was then removed to a propagation room inside the laboratory, where it continued to grow until early in January 1947, when its foliage suddenly died. At this time the seedling showed typical late-stage symptoms of phloem necrosis.

On July 2, 1947, one of the seedlings placed under test on July 26, 1946, was found to have died so suddenly that its leaves had failed to abscise and still hung on—a phenomenon by no means uncommon among naturally infected trees growing in the open. Examination of the inner phloem of this seedling revealed typical phloem necrosis discoloration. These test trees developed symptoms, therefore, in less than a year after being exposed to infective insects. This period contrasts strikingly with a possible inoculation period of 5 years where species of *Erythroneura* were used.

The evidence favors transmission of the virus by this species of *Scaphoideus*, whereas transmission by species of *Erythroneura* is more doubtful. Among several hundred other test trees in the same and adjoining cages and among several thousand trees in a nearby nursery, there exists as yet no other evidence of insect transmission of the virus. As a further check on the significance of the results obtained with *Scaphoideus*, however, an extensive series of tests was established late in the 1947 season, but it is too early to report on these tests.

The individuals of *Scaphoideus* used in the 1946 tests were recovered and, after the development of disease symptoms in the test trees in 1947, were forwarded to the Division of Insect Identification. According to P. W. Oman, all were found to belong to the single species, *Scaphoideus luteolus* Van D. Oman's determination of these specimens was based on specimens compared with the type of *luteolus*. They therefore are not the same, according to him, as the species that DeLong (*2*) described as *luteolus*, but are the same as the species DeLong and Mohr (*3*) described as *baculus*. *Scaphoideus luteolus* is widespread throughout the region where phloem necrosis occurs, having been taken in surveys from Ohio on the east, to Kansas on the west, and to Jackson, Mississippi, on the south. That this species occurs in regions not yet known to harbor the virus is demonstrated by Oman's statement accompanying the determination, in which he reports having seen specimens from the following states outside the disease area: New Jersey, New York, Pennsylvania, Maryland, Virginia, Georgia, and Alabama.

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The Mechanism of Cysteine and Glutathione Protection Against Alloxan Diabetes

Arnold Lazarow and John W. Patterson

Department of Anatomy, Western Reserve University

Stanley Levey

Department of Physiological Chemistry, Wayne University College of Medicine

The suggestion that alloxan may produce diabetes because of inactivation of essential sulfhydryl enzymes of the beta cells of the pancreas was made in an earlier paper by Lazarow (*5*). Inasmuch as large doses of alloxan also destroy other cells (liver, kidney, etc.), it also was postulated that the selectivity of this compound for the beta cells might be due to an especially low glutathione content which rendered the beta cells more susceptible to alloxan (*5*).

The injection of large doses of glutathione, cysteine, thioglycolic acid, and BAL protected rats against a diabetogenic dose of alloxan (*5*, *6*). By contrast, large doses of alanine, methionine, thiourea, and other compounds did not modify its diabetogenic effect. Protection against diabetes by these sulfhydryl compounds occurred only when they were given prior to the diabetogenic dose of alloxan. When glutathione or BAL was given 5 min after a diabetogenic dose of alloxan, no protection occurred. Since protection failed to occur when the sulfhydryl compound was given after the alloxan, it was suggested that, if the diabetogenic action of alloxan were due to a combination with essential sulfhydryl groups of enzymes, this reaction would not be as readily reversed as would be the case if the SH groups of the enzymes were simply oxidized to SS groups. However, a combination of alloxan with the SH groups would explain the failure to protect against diabetes when the sulfhydryl compound was given following the diabetogenic dose of alloxan.

Inasmuch as alloxan, dialuric acid (reduction product of alloxan), and derivatives of these compounds are unstable, the ultraviolet absorption spectra method was considered suitable for studying the possible reactions of alloxan with sulfhydryl molecules. This necessitated the study of the absorption spectra of alloxan and dia-