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STUDIES ON THE DWARF DISEASE OF RICE PLANT

By

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I. Introduction

The dwarf disease has long been recognized as one of the most important rice plant diseases in Japan. It is exclusively confined to Japan as far as the writer is aware. About 40 years ago historic severe outbreaks occurred devastating the rice fields in several localities including the Prefectures of Shiga, Kyôto, Hyôgo, Okayama, Hiroshima and Shizuoka. Since then the disease has become more and more widespread throughout the middle and southern part of the Main Island of Japan, Shikoku and Kyûshû and has been responsible for heavy losses in many localities.

Among the earlier investigators of this disease varying opinions were prevalent concerning the nature of this trouble, some workers ascribing it to some unfavourable soil conditions, while others looked upon it as the injury caused by the leaf hopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL.

According to our present knowledge this disease is undoubtedly a virus disease transmissible through the agency of the leafhopper above mentioned, acting as the vector. This is the first virus disease of plants shown to be transmitted by an insect as pointed out by KUNKEL (1926)¹¹¹⁾ based upon the writer's information, and also by HINO (1927).⁷⁶⁾

During recent years no definite progress has been made with the problem of transmission, host range, and the etiology of the disease. Since this deserves more attention, the writer attempted to attack the problem from the virological point of view.

The present work was begun at the suggestions of Dr. YAMADA, Dean of the Tottori Agricultural College and a part of the experiments

herein recorded, was performed in the Phytopathological Laboratory of that College. The work subsequent to 1929 has been carried out under the general supervision of Prof. S. Ito in the Botanical Institute of the Faculty of Agriculture of the Hokkaido Imperial University. The writer wishes to express here his sincere gratitude to Dr. K. MIYABE, Professor emeritus of the Hokkaido Imperial University and Dr. S. ITO, Professor of plant pathology and also to Dr. S. MATSUMURA, Professor of entomology of the Hokkaido Imperial University, Dr. G. YAMADA, Dean of the Tottori Agricultural College, Dr. Y. TOCHINAI, Professor of plant pathology and Mr. T. HAYASHI, Assistant Professor of Zoology of the Hokkaido Imperial University for their valuable suggestions and kind advices. Also to Mr. S. MIZUNO, veteran farmer in the Prefecture of Tottori, Dr. I. HINO, Professor of the Miyazaki College of Agriculture and Forestry, Mr. K. KURIBAYASHI, Plant Pathologist of the Nagano Agricultural Experiment Station, Mr. M. SAKURAI, Plant Pathologist of the Shiga Agricultural Experiment Station and Mr. S. ENDO, Assistant Professor of the Miyazaki College of Agriculture and Forestry to whom he is indebted for the materials which they kindly supplied to the writer.

II. History and geographical distribution of the disease

Although the dwarf disease of rice plant must have been existing for a number of years, it had attracted little attention until towards the end of the last century when serious outbreaks of the disease occurred in several localities including the Prefectures of Shiga, Kyôto, Hyôgo, Okayama, Hiroshima, and Shizuoka.

According to TAKATA (1895-96)²¹⁹⁾ the disease was first definitely noted in 1883 in a certain locality in the Prefecture of Shiga. It had spread through the entire Prefecture by 1892, when an area of over 1100 acres of rice fields was invaded by the disease. The destructive nature of the trouble having been realized, the study of the disease was taken up by the Government of Shiga Prefecture in 1893 and intensive work was done in the experimental fields selected in six counties to investigate the disease and to devise control measures. Consequently TAKATA who took charge of this work and HASHIMOTO in 1894-95 discovered the causal relation of leafhoppers to the dwarf disease of rice plant. The work was continued by the Shiga Agricultural Experiment Station soon after its establishment in 1895 and by a series of experiments carried out in 1897-1906 it was demonstrated that the disease is transmitted by the leafhopper, *Nephotettix apicalis* MOTSCH. var.

cincticeps UHL. and that the disease can be effectively controlled by eliminating the leafhoppers in the rice fields. In recent years the loss due to this disease has been remarkably reduced in this Prefecture by the control of the leafhoppers.

In the Prefecture of Kyôto the disease was first noted in 1886 and became prevalent during the following years. There were great outbreaks of the disease in 1889 and 1900 in this Prefecture. During recent years, however, the trouble has been reduced to a minimum by means of the control of the leafhoppers.

In "Rice of the Prefecture of Okayama"¹⁶⁰⁾ issued from the Government of the Prefecture of Okayama in 1910 it is mentioned that the dwarf disease of rice plant seems to have been known for many years in this Prefecture but there is no definite record of the occurrence of the disease until the summer of 1880. During the following years the disease became more widespread and in 1891 an area of over 3200 acres of rice field was invaded by it causing a reduction in yield amounting to 50000 bushels. During recent years, the disease has declined due to the control of the leafhoppers. It was shown by a survey in 1921 that an area of 70 acres of rice field was threatened by this disease.

In the Prefecture of Hyôgo, there is no authentic record of the occurrence of the disease until 1891, when an area of 7000 acres of rice field was devastated by it.

In the Prefecture of Hiroshima the serious nature of this disease was first realized in 1898-99. It was very severe in 1902-3. In these Prefectures the disease has been effectively controlled in recent years.

The Prefecture of Yamaguchi was invaded by the dwarf disease of rice plant a little later, it being first observed in 1910. Since then the occurrence of the disease has been noted every year though the damage was not very serious.

In the Prefecture of Shizuoka it is recorded that the disease was very severe in 1892 and 1893, although its origin is uncertain. During recent years considerable loss has been caused in the rice producing regions along the Tenryû river.

In the Prefecture of Yamanashi the disease was noted in 1888-89 devastating a rice-field area of 350 acres. Later the damage due to this disease was lessened for some unknown reason, but since the great outbreaks in 1915-16 it has again become a menace to the rice crop.

In the Prefecture of Nagano the disease was not noted with certainty until 1892, when an area of 750 acres was infested by it. Since

then the disease seems to have been of general occurrence in this Prefecture and it was shown by a recent survey that an area of 5000 acres of rice field was suffering from this disease.

Beside these, the disease seems to have been known for many years in the Prefectures of Kagoshima, Kumamoto, Nagasaki, Ehime, Saitama, Tochigi and Ibaragi, although its origin is not definitely known.

On the other hand this disease has been first noted during recent years in the Prefectures of Miyazaki, Shimane and Tottori.

In the Prefecture of Miyazaki it was first noted in 1912-13 but was confined to certain localities until 1918. Since then it became more prevalent year after year inducing at length the great outbreak of 1930, which infested an area of 137,500 acres or 95 percent of the rice fields in this Prefecture and caused a total loss of 395,000 bushels.

In the Prefecture of Shimane the disease first appeared in 1914 and in the same season or a year later it was noted to occur in the Prefecture of Tottori.

It is worthy of note that dwarf disease of rice plant has declined during recent years in the Prefectures of Shiga, Kyôto, Hyôgo, Okayama and Hiroshima where the rice crop seriously suffered from the disease for a number of years. The disease has in recent years been destructive in the Prefectures of Miyazaki, Ehime, Yamanashi, Nagano, Saitama and Ibaragi, while less severe in Tochigi, Shizuoka and also in Gumma.

The distribution of the disease will be shown in the following table and map based upon a recent survey conducted by the Department of Agriculture and Forestry of Japan (1929)⁹⁰⁾ as well as by the writer.

As shown in Table 1 the extreme northern limits of the known distribution of the disease lie in the Prefecture of Miyagi. It does not coincide with the northern limits of the distribution of the leafhopper, *Nephotettix apicalis* var. *cincticeps* which transmits the disease, this insect having been found over a very wide area in Japan, extending into the Prefectures of Akita and Iwate for some distance, in the north. The leafhopper has been reported from practically every region in the southern prefectures.

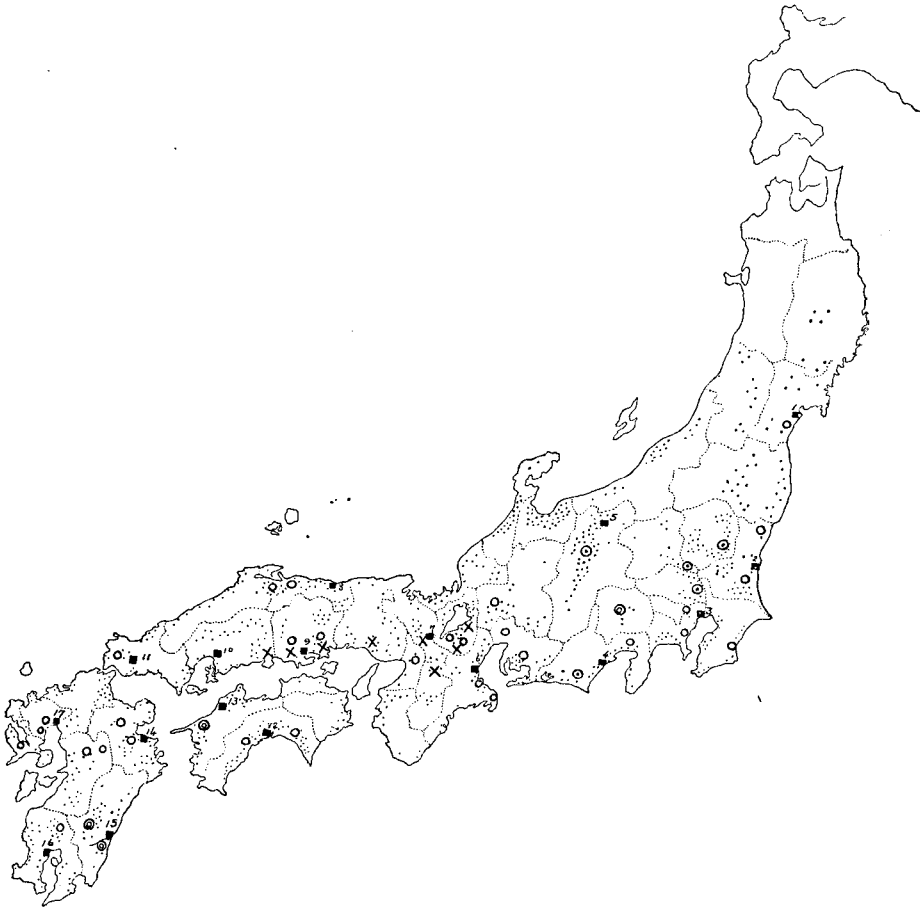
The leafhopper overwinters in its larval stage, usually feeding on *Astragalus sinicus* L. which is grown on rice fields through the winter and spring until it is ploughed in as a green manure in the early summer. In general it may be said that the dwarf disease of rice plant occurs in the regions where *Astragalus sinicus* is raised in abundance as will be seen in the following map.

TABLE 1. Geographical distribution of dwarf disease of rice plant

Locality	Amount of damage	Areas infested (in acres)	Historical remarks
Miyagi	slight	435	
Ibaragi	"	3750	*Severe in 1927
Tochigi	moderate	1250	
Gumma	"	—	Increasing since 1928, severe in 1929
Saitama	"	4100	
Chiba	slight	—	
Tôkyô	"	—	
Kanagawa	"	180	
Yamanashi	great	2500	Severe outbreaks in 1888-1889 and in 1915-1916
Nagano	moderate	5200	Severe in 1892
Shizuoka	"	500	*Severe in 1892 and 1893
Aichi	slight	—	
Gifu	"	—	
Mie	"	10	
Shiga	"	35	*Great outbreak in 1892
Kyôto	nearly none	—	*Great outbreaks in 1889 and 1900
Ôsaka	slight	350	
Nara	nearly none	—	
Wakayama	"	—	
Hyôgo	"	—	Great outbreak in 1891
Okayama	slight	70	*Great outbreak in 1891
Hiroshima	nearly none	—	*Severe in 1902-1903
Yamaguchi	slight	70	First noted in 1910
Tottori	"	90	First noted in 1914 or 1915
Shimane	"	—	First noted in 1914
Ehime	great	3000	
Kôchi	slight	—	
Saga	"	—	
Nagasaki	"	—	
Ôita	"	—	
Miyazaki	great	137500	First noted in 1912 or 1913, great outbreak in 1930
Kumamoto	slight	500	*
Kagoshima	"	—	

(* The disease seems to have been known for a number of years though its origin is not definitely known)

Geographical distribution of dwarf disease of rice plant



Remarks

- Localities where *Astragalus sinicus* L. is raised in abundance.
- Localities where dwarf disease of rice plant occurs.
- ⊙ Localities where dwarf disease of rice plant is severe.
- ⊗ Localities where dwarf disease of rice plant is very severe.
- × Localities where dwarf disease of rice plant was very severe.

1: Sendai, 2: Mito, 3: Tôkyo, 4: Shizuoka, 5: Nagano, 6: Tsu, 7: Kyôto,
 8: Tottori, 9: Okayama, 10: Hiroshima, 11: Yamaguchi, 12: Kôchi, 13:
 Matsuyama, 14: Ôita, 15: Miyazaki, 16: Kagoshima, 17: Saga.

III. Review of literature

The earliest account of the dwarf disease of rice plant was given under the title "Dwarfed rice plant, its cause and control measures."⁹²⁾ in the Official Gazette of Oct. 18, 1890, by the Department of Education replying to the inquiry from the Government of the Prefecture of Kyôto. This article seems, as DAIKUHARA (1900)³⁹⁾ cited it later, to have been written by KERNER, KOZAI and MORI of the Faculty of Agriculture of the Tôkyo Imperial University. It is mentioned in this report that the dwarf disease of rice plant is caused by the products of reduction induced in the soil. On account of the deficiency of oxygen in the soil, the sulphates contained in the soil and manure as well as in the irrigation-water are brought to reduction producing iron sulphide which inhibits the growth of rice plants. As the control measures it was recommended to oxidize the iron sulphide deposited in the soil by means of withdrawing stagnant water, drying of the soil, application of unslaked lime and the surface firing of the soil.

In 1893 YAMADA* reported the results of analyses of the rice plants affected by the dwarf disease as compared with healthy plants, stating that the diseased plant just before the flowering time contains more crude fibre, iron oxide, calcium, sodium and sulphuric acid, and less crude fat, crude protein, non-nitrogenous extract, total nitrogen, albuminous nitrogen, non-albuminous nitrogen, magnesium, potassium, phosphoric acid, chlorine and silica than the healthy plant.

TAKATA²¹⁹⁾ stated in 1895-96 that iron sulphide deposited in the soil, the amount of stagnant water, low temperatures of the irrigation water, climatic conditions or cultural practices do not account for the incidence of the disease but a certain species of leafhopper which he named "mon-yokobai" is responsible for the disease. He pointed out that the progeny grown from seed of diseased plants is not always affected with the disease and also that not all rice varieties are equally susceptible. The leafhopper which he called "mon-yokobai" is apparently *Deltocephalus dorsalis* MOTSCH. as shown by his description accompanied by illustrations. This leafhopper, however, has no connection with the disease as reported later by the Shiga Agr. Experiment Station. According to ISHIKAWA (1928),⁸⁶⁾ HASHIMOTO in 1894-95 was the first to prove the causal relation of the leafhopper to the dwarf disease of rice plant. However, it is uncertain with what species of

* See (39)

leafhopper he worked.

The Shiga Agricultural Experiment Station undertook studies on the disease soon after its establishment in 1895 and continued the work during a period of 20 years. The results of earlier experiments have been reported in "Results of experiments with insect pests, Reports 1-8".¹⁹²⁾ Some noteworthy points will be given below. In Report 1 (1897) it is stated that the dwarf disease of rice plant was caused by the leafhoppers, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL., *Deltocephalus dorsalis* MOTSCH., *Thamnotettix tobae* MATS., *Cicadula fasciifrons* STAL., and likewise by *Zygina limbata* MATS.? while none of the leafhoppers, *Delphacodes striatellus* FALL., *Tettigonia viridis* L. and *Nisia atrovonosa* LETH. was responsible for the incidence of the disease. It is also mentioned that the damage was markedly lessened by eliminating the leafhoppers by means of kerosene. It is evident that the disease was at this time entirely attributed to the leafhoppers. In Report 2 (1900) it is reported that only one species of all leafhoppers tested viz., *Nephotettix apicalis* var. *cincticeps* is capable of producing the true dwarf disease of rice plant. It is also stated that new shoots from the affected plants always contracted the disease even though they had been kept free from the leafhoppers. In Report 6 (1906) it is stated that the leafhoppers collected in the Prefecture of Shiga were capable of producing the disease while those taken from Tôkyo were uninfective. This finding was confirmed in Reports 7 and 8 (1908) showing that the leafhoppers taken from the Prefecture of Okayama produced the disease as well as those from the Prefecture of Shiga but that the leafhoppers from Tôkyo and the northern districts of Honshû were entirely uninfective. Furthermore it is stated that uninfective leafhoppers taken from Tôkyo or the northern districts of Honshû became able to induce the disease if they fed on the dwarf diseased rice plant for about 5 days. Thus it became evident that the leafhopper itself does not cause the disease but transmits it acting as the vector. This was first shown by the experiments performed in 1906 and confirmed during the following years. Accordingly it is in 1908 that the leafhopper, *Nephotettix apicalis* var. *cincticeps* was first authentically recorded to transmit the dwarf disease of rice plant, although the causal relation of the leafhopper to the disease had been proved 8 years before. The results of the experiments carried out by the Shiga Agricultural Experiment Station from 1895 to 1908 have been summarized in its Special Bulletins No. 1 and 2,¹⁹³⁾ issued in 1906 and 1910 respectively.

TAKAMI (1901)²¹⁸⁾ also summarized the results of the earlier experiments of this exp. station, ascribing the disease to the infestation of the leafhopper. The results of the later experiments have been recorded in the Annual Reports for 1907 to 1915,¹⁹⁴⁾ the following points being worthy of note. Besides the rice plant, it is stated, *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA was affected by the disease but that neither *Setaria italica* BEAUV. nor *Panicum miliaceum* L. was subject to the disease. It is also mentioned that a variety of rice plant, "Ishuku-shirazu" was immune from the disease and that a single infective leafhopper was capable of transmitting the disease to a healthy plant when fed on it for only one day.

Either before or after the work of the Shiga Agricultural Experiment Station, the problem of the dwarf disease of rice plant was also attacked by the Agricultural Experiment Stations of Hyôgo (1895-9),⁸⁴⁾ Nara (1897),¹⁵³⁾ Okayama (1902-9),¹⁵⁹⁾ Hiroshima (1903)⁷⁷⁾ and Kyôto (1905, 1910).^{234, 117)} They reached the same conclusion as regards the causal relation of the leafhopper to the disease.

Meanwhile, DAIKUHARA (1902, 1904)³⁹⁾ reported the results of biochemical studies on the dwarf disease of rice plant. According to him the affected leaves contain more crude proteins and ashes, and less crude fibre, crude fat and non-nitrogenous extract than the healthy leaves. They contain much more phosphoric acid, potassium and magnesium, and relatively less calcium than the normal leaves do. The rate of transpiration as well as the translocation of starch is remarkably decreased in the leaves of affected plants. The amount of oxidizing enzymes, particularly peroxidase, and diastase is greater in the affected leaves as compared to that in the healthy leaves while catalase is less active in the former. Mechanical injuries given to the leaves and roots, sorts of manure, and cultural practices do not account for the incidence of the disease. Transmission was not effected by injection of juice from the diseased plant into healthy seedlings. Not all rice varieties are equally susceptible to the disease, "Ishuku-shirazu" being the most resistant. After all DAIKUHARA considered the dwarf disease of rice plant as a disease allied to the sereh of sugar cane, the mosaic disease of tobacco and likewise to the stigmonose of carnations.

In 1905 YONEMARU and ASO²³⁴⁾ reported the results of their experiments in connection with the disease in a Special Bulletin of the Kyôto Agricultural Experiment Station. They also stated that the diseased leaves contained more nitrogen, particularly amides, and much more

oxidizing enzymes than the healthy leaves, and that the seedlings derived from the seed of diseased rice plants did not necessarily contract the dwarf disease. They mentioned that the injury due to leafhoppers could not be considered as the main cause of the dwarf disease but *Nephotettix apicalis* var. *cincticeps* tended to induce the disease. The outline of this report has been published in the Extra Bulletin No. 12¹¹⁷⁾ of the same experiment station, which was issued in 1910.

In 1910 ANDÔ⁷⁾ briefly reported the results of transmission experiments which had been carried out in the Imperial Agricultural Experiment Station. In 1905, according to him, the leafhoppers from Tôkyo were allowed to feed upon dwarf diseased rice plants taken from Shiga Prefecture and then the insects of the second or third generation proved to be capable of producing infections in healthy plants. He further stated that the uninfected leafhoppers from Tôkyo became infective when allowed to feed upon diseased rice plants for 15 days and that the infective power was not transmitted to the progeny, when the infective parents were transferred to healthy rice plants at 5-7 days' intervals in order to keep them from sucking the juice of diseased plants. He emphasized that the disease is not caused by the leafhopper but by certain unknown infective principle which is carried by the leafhopper.

During the Taisho Era (1912-1926) the disease has been investigated by the Agricultural Experiment Stations of Shiga,¹⁹⁴⁾ Kago-shima,⁹⁷⁾ Miyazaki,^{144, 145)} Nagano¹⁵²⁾ and Yamanashi,²³³⁾ establishing the causal relation of the leafhopper, *Nephotettix apicalis* var. *cincticeps* to the disease.

Prior to this, the Imperial Agricultural Experiment Station in Nishigahara near Tôkyo and its Kyûshû Branch Station in Kumamoto had taken up studies on the disease since the turn of the century as cited by BOKURA (1930)²¹⁾ and also by MURATA (1931).¹⁴⁹⁾ The results of investigations have been reported giving only brief notes in the Annual Reports of the Experiment Station⁸⁵⁾ during the Taisho Era. In the Annual Report for 1913-1915 issued in 1916 it is stated that the presence of certain protozoa was noticed in the tissues of the diseased rice plants as well as in the blood of the leafhoppers. However, no detailed account has been given of these protozoa.

KASAI⁹⁹⁾ reported in 1924, the finding of NELSON'S bodies in association with the dwarf disease of rice plant and several other virus diseases of plants, stating that these bodies represent no protozoa but the degenerated or even normal nuclei of the host plant. However, as shown

by his descriptions and illustrations these bodies are apparently different from those described and figured by NELSON (1922).¹⁵⁶⁾

Recently the writer (1931)⁶⁸⁾ reported that certain intracellular bodies are invariably present in the chlorotic tissue of the leaf of the rice plant affected by the dwarf disease.

IV. Symptoms of the disease

The dwarf disease of rice plant usually appears in late June when the rice plants have been transplanted into the rice field and grown 6-8 inches high, although it is not unusual to find the disease in the seed bed.

The first visible symptom of the disease manifests itself as yellowish white specks along the veins of newly unfolded leaves. These specks which develop before the leaves unfold, are yellowish green to yellow when viewed by diffused light. By holding up to the light these specks become distinct, being yellowish white to white in color. The specks elongate and spread out along the leaf parallel to the midrib, forming fine interrupted streaks. (Pl. II, Fig. 4). These range from mere dots to an area several millimetres in length and from 0.2 to 1 mm. in width. The succeeding leaves invariably show the white specks while the lower, previously formed leaves exhibit no signs of the disease. On the leaf which shows the first visible symptom of the disease the specks may be confined to the lower part of the leaf blade or to only one side of the midrib near the base of the leaf. On the succeeding leaves more conspicuous specks develop in abundance and connect with each others forming almost continuous streaks along the veins. Thus the symptom of the disease becomes rather conspicuous in the middle of July.

The growth subsequent to infection being much arrested, the diseased plant becomes remarkably stunted, and the internodes shortened, while numerous diminutive tillers develop, producing a rosette appearance. Affected plants tend to develop a foliage dark green in color.

The roots of the affected plants are much arrested in their growth, and only small roots develop extending horizontally. This condition becomes especially evident when such a plant is transplanted.

The plants infected in their very early stage of growth, become severely stunted being at most 10 inches high. (Pl. I, Fig. 1). They produce none or only a few worthless panicles. The symptoms of the plants affected at a later stage of their development are less prominent.

Upon one or two of the younger shoots of such a plant, the streak may appear but the other shoots remain entirely healthy.

V. Intracellular bodies associated with the disease

1. Occurrence of intracellular bodies in the affected rice plants

As stated in the writer's previous paper,⁶⁸⁾ certain intracellular bodies were revealed both in fresh and embedded material of leaves of rice plant affected with dwarf disease. Similar bodies were found also in the crown and root of the diseased plant.

For fixation of the material several fixatives were used including medium chromo-acetic solution, FLEMMING's fluid, MERKEL's fluid, ALLEN's modification of BOUIN's fluid, GILSON's fluid, ZENKER's fluid, acetic alcohol, formol-acetic alcohol and SCHAUDINN's fluid. Among these, chromo-acetic solution and formol-acetic alcohol proved to be most suitable for general purposes. The fixed material was embedded in paraffin and sectioned by means of a microtome in the usual way. Transverse and longitudinal sections were cut 5 microns thick and stained with HEIDENHAIN's iron-alum haematoxylin or safranin and gentian violet. Longitudinal sections were preferable to demonstrate the intracellular bodies. Some of the materials which had been fixed in SCHAUDINN's fluid, GILSON's fluid, ZENKER's fluid and formol-acetic alcohol were stained with GIEMSA's stains. The GILSON-GIEMSA combination gave very satisfactory results. Attempts were also made to demonstrate chondriosomes, if any, in the intracellular bodies, by fixing the material in REGAUD's fluid, BENDA's fluid, CHAMPY's fluid and SAPÉHIN's fluid and staining the sections with iron alum haematoxylin or with ALTMANN's acid fuchsin.

Microscopic studies of free hand sections of the diseased leaf show chlorotic modifications in the mesophyll-cells adjacent to some of the vascular bundles. In sections mounted in water, the chlorotic tissues are lighter in color or nearly colorless, the chloroplasts in these cells being light colored and smaller in size and number, due to their disintegration. In the cell where most of the chloroplasts have been disintegrated, a certain foreign body is found in close proximity to the nucleus. Such foreign bodies are usually round in shape, more or less larger than the host nucleus and they contain many vacuoles of varying size. These bodies stain yellow in dilute iodine potassium iodide solution, brick red in aceto-carmin, Grenadine pink (after RIDGWAY¹⁷³⁾) in

the alcoholic solution of Sudan III, and pinkish cinnamon in MILLON'S reagent.

However, more comprehensive studies of the intracellular bodies can be done in the microtomed sections properly stained. In the preparations stained with HEIDENHAIN'S haematoxylin, the chlorotic areas are easily recognized since the chloroplasts in these tissues show less affinity for the stain, and they are, according to the degree of chlorosis, smaller and less regular in shape and fewer in number than those in the apparently healthy tissues of the same leaf (Pl. III, Fig. 11). Haematoxylin brings out the intracellular bodies very sharply, since they take the stain as intensely as the host nuclei. When sections are stained with safranin and gentian violet, the bodies show a special affinity for safranin while gentian violet stains the host nuclei violet in color. In the sections stained with GIEMSA'S stains the intracellular bodies show deep blue ("sailor blue" of RIDGWAY) whereas the host nuclei stain purple to violet ("mallow purple" to "nigrosin violet") and the chloroplasts light lilac. The foreign bodies under consideration occur singly in the host cells but occasionally two of them are found in the same cell. They are more abundant in the mesophyll cells but it is not unusual to find them in the epidermal cells (Pl. III, Fig. 8, 9, 12, 13). The bodies are located near or in close contact with the host nucleus which frequently becomes much enlarged. They vary considerably in shape and size. Round to oval are the most usual forms but it is not uncommon to find bodies amoeboid or very irregular in shape. In size they range from 3 to 10 microns in length and 2.5 to 8.5 microns in width, frequently much larger than the host nuclei which are 2.5 to 3.5 microns in diameter. The contents of the intracellular bodies appear to consist of a somewhat homogeneous structure, surrounded by an indistinct membrane, and contain many vacuoles of various size (Pl. IV, Fig. 14, 15, 16). The vacuoles seemed more conspicuous in the bodies from the material fixed in formol-acetic alcohol or ZENKER'S fluid. All attempts to demonstrate a nucleus or chromatin granules in these bodies were unsuccessful but minute granules resembling the so-called chlamydozoa were revealed, although their exact nature was not definitely ascertained (Pl. III, Fig. 12, 13). A search for chondriosomes in these bodies by use of recommended fixatives and stains revealed no bodies of this type. Although the intracellular bodies occur invariably in the chlorotic areas of the leaf, as yet they have not been found in the intact tissue of the diseased leaf or in plants free from the disease. Quite similar bodies have been demon-

strated in the phloem and its adjacent tissues in the root (Pl. III, Fig. 10) and crown of the diseased plant. They are found abundantly in the parenchyma-cells, near or in close contact with the host nuclei.

The intracellular bodies in question are not artifacts, since they can be found in the cells of fresh material as well as in material fixed by various methods and stained with various stains. It is also evident that these bodies are neither deformed or degenerating nuclei, nor hypertrophied chloroplasts of the host cells, although their exact nature remains still unknown. Beyond doubt they are similar to the intracellular bodies which have been found in association with certain virus diseases of plants and animals.

2. Occurrence of the intracellular bodies in the other susceptible plants

As will be described more fully later, *Panicum miliaceum* L., *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA and *Alopecurus fulvus* L. are readily affected by dwarf disease of rice plant whereas wheat, rye and oats are more or less susceptible to the disease. It seemed advisable, therefore, to examine the chlorotic tissues of the affected leaves of these plants in the hope of finding the intracellular bodies which have been demonstrated in diseased rice plants. The affected leaves were fixed in GILSON's fluid and embedded in paraffin in the usual way. Longitudinal and transverse sections were cut 7 microns thick and stained with GIEMSA's stains. Certain foreign bodies have been found to occur invariably in the chlorotic tissues of leaves of the above mentioned plants with the exception of oats which were not studied in this connection owing to the deficiency of suitable materials. These bodies agree in every respect with those found in the leaves of diseased rice plants (Pl. IV, Fig. 17, 18, 19). They are localized usually in the tissues adjacent to the vascular bundles and near or in close contact with the host nuclei which are more or less smaller than them. The shape, structure and staining reaction of these bodies very closely resemble those described in respect of the intracellular bodies found in rice plants and repetition here would be superfluous.

3. Previous investigations of the intracellular bodies associated with virus diseases of plants

IWANOWSKI(1903)⁸⁹⁾ was the first to discover the intracellular bodies in association with virus diseases of plants. Working with the mosaic

disease of tobacco, he found in the cells of chlorotic leaf tissues certain amoeboid plasmic aggregates which occur near or in close contact with the nuclei. He considered that these amoeboid bodies were reaction products of the cells resulting from the irritation by the parasite and that they could not be a causal organism of the disease because bodies as large as these could not possibly pass through a bacterial filter. IWANOWSKI found also the presence of granular cell-inclusions which he thought to be zoogloea of bacteria, the causative agent of the mosaic disease. However he could not successfully culture the bacteria which would cause the mosaic disease. The findings of IWANOWSKI were confirmed by PALM (1922),¹⁶¹⁾ who agreed with the former that the amoeboid bodies in the diseased cells were the result of reaction of the cell to the irritation of the causative agent. PALM, however, considered that the minute granules which IWANOWSKI had assumed to be bacteria "agreed in every respect with the Strongyloplasmen of LIPSCHÜTZ" or the chlamydozoa of PROWAZEK, being homologous with the GUARNIERI bodies. He believed that these small granules might be the causal agency of the mosaic disease of tobacco and named them *Strongyloplasma iwanowskii*.

The intracellular bodies associated with mosaic disease of tobacco were further studied by DICKSON (1925),⁴⁴⁾ GOLDSTEIN (1924, 1926),^{72, 73)} KUNKEL (1924),¹¹⁰⁾ RAWLINS and JOHNSON (1925),¹⁷⁰⁾ F. F. SMITH (1926),¹⁹⁶⁾ HOGGAN (1927),⁷⁸⁾ KLEBAHN (1928)¹⁰⁴⁾ and LIKHITÉ (1929).¹¹⁹⁾ Notably GOLDSTEIN studied the vacuolate amoeboid bodies which she called X-bodies, in the living condition and noticed an apparent change in form and their "distinct amoeboid movement", coming to believe that these bodies are the causal organisms of mosaic disease. LIKHITÉ also regarded the X-bodies as representing a living parasitic organism for which he proposed the name *Vacuolarium iwanowski*. HOGGAN, on the other hand, presented some evidence to show that the X-bodies are products of diseased cells but not of the nature of a causal organism.

The occurrence of similar vacuolated bodies has been shown in mosaic diseases of tomato (HOGGAN 1927),⁷⁸⁾ pepper (HOGGAN 1927),⁷⁸⁾ potato (K. M. SMITH 1924,²⁰⁰⁾ HOGGAN 1927,⁷⁸⁾ CLINCH 1932³⁶⁾), petunia (F. F. SMITH 1926,¹⁹⁶⁾ HOGGAN 1927⁷⁸⁾), *Datura* (F. F. SMITH 1926)¹⁹⁶⁾ and some other solanaceous plants (HOGGAN 1927,⁷⁸⁾ J. H. SMITH 1930,¹⁹⁸⁾ SHEFFIELD 1931¹⁰⁰⁾); likewise in corn mosaic (KUNKEL 1921),¹⁰⁵⁾ sugar cane mosaic (KUNKEL 1924,¹¹⁰⁾ COOK 1925³⁸⁾), wheat mosaic (McKINNEY et al. 1923),¹³⁷⁾ Chinese cabbage mosaic (KUNKEL 1924),¹¹⁰⁾ *Hippeastrum* mosaic (KUNKEL 1922,¹⁰⁶⁾ 1924,¹¹⁰⁾ McKINNEY et al. 1923,¹³⁸⁾ HOLMES

1928),⁸²⁾ Phytolacca mosaic (F. F. SMITH 1926),¹⁹⁶⁾ Dahlia mosaic and dwarf (GOLDSTEIN 1927),⁷⁴⁾ "infectious chlorosis" of *Euonymus japonicus* (F. F. SMITH 1926),¹⁹⁶⁾ Fiji-disease of sugar cane (LYON 1910,¹²²⁾ 1921,¹²³⁾ REINKING 1921,¹⁷²⁾ MCWHORSTER 1922,¹⁴²⁾ KUNKEL 1924¹⁰⁹⁾), spike disease of sandal and *Vinca rosea* L. (NARASIMHAN 1928,¹⁵⁴⁾ 1933¹⁵⁵⁾), lily mosaic (GOLDSTEIN 1928)²³⁷⁾ and in curl of the vine (PETRI 1931).¹⁶²⁾

As to the nature of these bodies there exists a remarkable difference in opinion. LYON (1921) who first reported the presence of such bodies in the galls produced on the sugar cane leaves affected with Fiji disease looked upon them as parasitic organisms and proposed the name *Northiella sacchari* although its systematic position was not definitely known. KUNKEL (1924) also considered these bodies to belong to a parasitic organism, being capable of division and appearing to grow. MCWHORSTER (1922) stated that the foreign body present in the cells of Fiji galls was an amoeba which could be cultured in cane juice and he named the organism *Phytamoeba sacchari*. KUNKEL (1921, 1924), giving a detailed account of similar intracellular bodies associated with the mosaic diseases of corn, *Hippeastrum equestre*, *Brassica pekinensis* and sugar cane, stated that these bodies might represent "one stage in the life of a causal organism". HOLMES (1928) working with the intracellular bodies in *Hippeastrum* mosaic came to the conclusion that these bodies consist of living cytoplasm, chondriosomes being found within the bodies. He states: "Whether the body represents a stage in a foreign organism, a mass of plant cell cytoplasm containing virus, or a mass of the cell cytoplasm not immediately in contact with virus but stimulated by the diseased condition, is not known." K. M. SMITH (1924) found similar amoeboid bodies in the chlorotic leaf tissues of mosaic diseased potato plants, which he considered to be some degeneration product of the cell induced by the mosaic, being effects rather than causes of the disease. HOGGAN (1927) found X-bodies consistently in different solanaceous plants affected with tobacco mosaic or yellow tobacco mosaic etc., but no such bodies could be demonstrated in the same plants when affected with cucumber mosaic or other virus diseases. She also found "vacuolate protoplasm-like material" in the cells of potato in association with several different virus diseases, appearing in various forms, either as definite bodies or as irregular masses whose appearance suggested intermediate stages between the bodies and the host cytoplasm. Accordingly she came to the conclusion that the X-bodies were not of

the nature of a causal organism. CLINCH (1932) also found similar vacuolate bodies in association with simple mosaic, interveinal mosaic, crinkle, and streak of potato. According to him mitochondria are present in these bodies and numerous fatty globules are associated with the X-bodies at certain stages. He suggested that the X-bodies arise as a result of viscosity changes in the cytoplasm of the diseased cells. BREHMER and BÄRNER (1930)³¹⁾ considered the amoeba-like bodies associated with virus diseases of potato to be the causal fungus which belongs to the genus *Plasmodiophora*.

GOLDSTEIN (1927) claims that she was able to demonstrate nucleus-like structures within the X-bodies in the cells of dahlia plants affected by mosaic and dwarf disease. She wrote that the X-bodies could not be considered "simple degeneration products of the host cells, or cell nucleus, because of their very protoplasmic appearance and structure, and the presence within them of internal vacuoles and nucleus-like structures, the presence of peripheral vacuoles, and their apparent division by constriction".

The other investigators did not attempt to interpret the significance of intracellular bodies which they found.

According to MCKINNEY (1925),¹³⁴⁾ there are two types of intracellular bodies in association with virus diseases of plants. The first type is, according to him, represented by the intracellular bodies associated with tobacco and potato mosaics, the Fiji disease of sugar cane and mosaic disease of wheat, which resemble each other in form and structure. Most of the bodies possess a matrix which is homologous in nature and contains vacuoles varying in number and size. All of these bodies appear to be surrounded by a membrane and to consist of protoplasm. The intracellular bodies associated with the mosaic diseases of corn, *Hippeastrum*, and sugar cane represent the second type, presenting a somewhat different appearance from the above mentioned bodies. Most of these bodies seem, MCKINNEY writes, to be made up of compactly arranged rather large, independent granules and not to be surrounded by a membrane, and they are not so suggestive of protoplasm as the bodies of the first type. HOLMES (1928), however, claims that the intracellular bodies associated with *Hippeastrum* mosaic consist of living protoplasm as mentioned before.

It appears that the intracellular bodies which the writer found in association with dwarf disease of rice plant belong to the first type, if there really do exist two types of intracellular bodies as pointed out by

McKINNEY.

Several other types of cell inclusions have been described as associated with certain virus diseases of plants. Among these are plasmodium-like substance in the mosaic diseased cane leaf and stem tissues found by MATZ (1919¹²⁶); "flagellated organisms" associated with mosaic of tomato, dahlia, squash, and Hippeastrum, which were described by ECKERSON (1926⁵⁶); the minute "hyaline bodies" reported by SOROKIN (1927)²⁰⁴ to occur in the transparent spheres resulting from the destroyed chloroplasts in the mosaic diseased tomato leaf; KLEBAHN's scolecosome (1926,¹⁰³ 1928¹⁰⁴) and other cell inclusions in association with "Alloiophyllie" of *Anemone nemorosa*, "infectious chlorosis" of *Abutilon thompsoni*, and mosaic diseases of tobacco and potato etc.; similar bodies in dahlia and lettuce mosaic (BRANDENBURG 1928);²³⁶ SCHAFFNIT-WEBER's elytrosoma associated with mosaic diseases of beet and broad bean (1927¹⁷⁸); "amorphous bodies" reported by PLAKIDAS (1927,¹⁶⁴ 1928¹⁶⁵) to occur in the root tissues of straw-berry affected with yellows or dwarf, and similar structures demonstrated by RAWLINS (1926)¹⁶⁹ in the root tips of sugar beets affected with curly top.

No attempt will be made to discuss in detail these inclusion bodies because it seems to the writer that such bodies do not fall in the same category with the intracellular bodies associated with dwarf disease of rice plant and moreover, some of these bodies have been found to occur in healthy plants.

4. *Absence of inclusion bodies in the viruliferous leafhopper*

Because of the occurrence of the so-called chlamydozoa-like bodies in the intracellular bodies associated with dwarf disease of rice plant, it seemed advisable to look for similar bodies in the viruliferous leafhopper. Such a study might result in the discovery of the causative agent of the disease or some indication of the presence of the virus.

The infective leafhoppers, both nymphs and adults, were fixed in GILSON's fluid and embedded in paraffin in the usual way. Transverse and longitudinal sections were cut 5 or 7 microns thick and stained with GIEMSA's or MALLORY's stains. Similar virus free leafhoppers served as checks.

In certain insect borne diseases of milkweeds (HOLMES 1925)⁸¹ and Euphorbias (FRANÇA 1914)⁶³ caused by protozoans, the causal organisms have been found in the salivary glands and in the alimentary canal of

their insect carriers. Accordingly it seemed desirable to inspect particularly these organs of the leafhopper in the hope of finding the causative agents or some indication of the presence of the virus. As will be more fully stated later, the virus may be transmitted from the viruliferous females to their offspring through the eggs which are in all probability infected by the virus at an early stage of development in the ovaries of the maternal insects. The ovaries, therefore, must not be left out of consideration.

The alimentary canal consists of pharynx, oesophagus, ventriculus, intestines and rectum. The pharynx is represented by a flattened aperture, at the proximal end of which the oesophagus commences, passing through the cephalic ganglion. The oesophagus then extends into the thorax where it opens into the ventriculus. The oesophagus wall is composed of flat epithelial cells and is lined with a cuticular intima. The ventriculus is narrow in front and widest in the posterior region of the thorax where it passes into the abdomen to be connected with the midintestine. The digestive epithelium of the ventriculus consists of large cells which project into the lumen while the walls of the midintestine are lined with an epithelium of large columnar cells. Malpighian tubules arise at the point of junction of the mid- and distal-intestines. The tubules are four in number, very long and convoluted. They have a moniliform appearance and are of uniform width throughout. Both the intestines and Malpighian tubules are intimately bound up with the diffuse fat bodies (Pl. V, Fig. 26).

The salivary glands lie in the prothorax at both sides of the oesophagus. They are composed of a number of glandular cells which are somewhat triangular in section and possess considerably large nuclei (Pl. V, Fig. 22 & 24).

The ovaries of an adult leafhopper occupy the dorsal and lateral regions of the abdominal cavity. The ovarian wall is very thin and each ovarian tubule is filled with numerous egg follicles in various stages of development. The egg cell is round in shape and enveloped by the follicle epithelium which is uniform in thickness and composed of cuboidal cells (Pl. VI, Fig. 27).

The mycetome or pseudovitellus is an organ present in most *Homoptera*. It is a large paired organ located in the first three abdominal segments and has no direct connection with any other organs. The cells composing it harbour microorganisms of a fungal nature and accordingly are termed mycetocytes. It is generally believed that these

microorganisms are symbiotic fungi and this subject, along with their origin and incidence in different insects is discussed by BUCHNER (1930).³²⁾ In all the *Homoptera* the symbionts are transmitted from one generation to the next through the eggs. The mycetome itself has no direct connection with dwarf disease of rice plant because it is present in virus-free as well as in viruliferous leafhoppers. But since both the mycetome and virus have been known to be transmitted from one generation to the next through the eggs, it seemed desirable to look for some indication of the presence of the virus in the mycetome. The mycetome of the leafhopper under consideration is composed of three distinct zones, comprising different types of cells (Pl. VI, Fig. 28). The central portion is occupied by numerous compact angular cells which are provided with large nuclei and packed with mycelium-like strands. Sometimes the inner cells appear to have collapsed, leaving the nuclei among clumps of cytoplasm (Pl. VI, Fig. 29). The next zone is composed of cells, whose outlines are obscure owing to the presence of thick mycelium-like strands which fill up the cell lumens. The nuclei, however, can be found among these mycelium like strands. The third zone is very thin, being one cell thick, and borders the mycetome. These outer cells are small and flat.

In spite of a careful examination of thousands of both transverse and longitudinal sections of viruliferous leafhoppers, the writer was unable to find any inclusion bodies or microorganisms of etiological significance, nor visual evidence of the presence of the virus in the salivary glands and the alimentary canal, in the egg follicles in the ovarian tubules, nor likewise in the mycetome and other organs.

DOBROSKY (1929),⁴⁶⁾ working with aster yellows, also could not find any bacterium, Rickettsia, fungus, protozoan, or X-body, of etiological significance, or visual evidence of the presence of the virus in the leafhopper, *Cicadula sexnotata* FALL. According to SWEZY and SEVERIN (1930),²¹⁷⁾ the beet leafhopper, *Eutettix tenella* BAKER, the carrier of curly top of sugar beets, harbors in its alimentary tract and blood two different bacilliform organisms, which "possess some of the characteristics of Rickettsia". However, the causal relations of these organisms to curly top of beets has not been established.

VI. Results of transmission experiments

A) No transmission through seeds

The rice plants affected with the dwarf disease at a later stage of

their development often produce a few small panicles with some matured grains, even though the plants as a whole become markedly stunted.

In the fall of 1922, about 200 seeds were collected from diseased rice plants in a certain locality in Tottori Prefecture. On May 8, 1923, these seeds were sown in 7 Wagner pots, after they had been soaked in water for a week. On July 4 the resulting seedling plants were transplanted, taking each 3 plants together into a pot. The plants were not protected in any way since the dwarf disease of rice plant has never been noticed in the vicinity of the City of Tottori where the writer's experiments were carried out. The total number of plants grown from seeds of diseased plants was 165, that is, 135 of "Aikoku" and 30 of "Hayakitabu", both varieties being very susceptible to the dwarf disease. None of these plants developed any symptoms of the disease throughout the season.

On May 28, 1929, 420 seeds of "Aikoku"-variety which had been collected in the previous autumn from diseased plants were sown in several pots. Three hundred and twenty eight plants were grown from these seeds. All these plants remained healthy during the 3 months that they were kept under observation.

As shown above, 493 rice plants in all were grown from seeds of diseased plants and in no case did an affected plant appear.

TAKATA (1895-96)²¹⁹⁾ and YONEMARU and ASO (1905)²³⁴⁾ stated that the seedlings grown from the seed of dwarf diseased rice plants do not necessarily contract the dwarf disease. Similar results were also reported in the Report of the Okayama Agricultural Experiment Station published in 1902.

It seems to be conclusive that the dwarf disease of rice plant is not transmitted through seeds.

Most virus diseases of plants are not readily transmitted through seeds. This has been reported true by various investigators regarding these diseases: tobacco mosaic (MAYER 1886,¹²⁴⁾ ALLARD 1912,¹⁾ 1914,²⁾ 1915,³⁾ CLINTON 1915³⁷⁾), tomato mosaic (ALLARD 1916,⁴⁾ GARDNER and KENDRICK 1922,⁷¹⁾ DICKSON 1922⁴²⁾), sugar cane mosaic (WILBRINK and LEDEBOER 1910²²⁹⁾ beet mosaic (LIND 1915,¹²⁰⁾ ROBBINS 1921,¹⁷⁴⁾ BÖNING 1927,²⁵⁾ JONES 1931⁹⁶⁾), spinach blight (McCLINTOCK and SMITH 1918),¹²⁹⁾ potato mosaic (SCHULTZ and FOLSOM 1923),¹⁸²⁾ corn mosaic (BRANDES 1920,²⁸⁾ BRANDES and KLAPHAAK 1923⁸⁰⁾), crucifer mosaic (CLAYTON 1930),³⁵⁾ peach yellows (SMITH 1888,¹⁹⁵⁾ BLAKE, COOK, and CONNORS 1921¹⁹⁾), curly top of beet (SHAW 1910,¹⁸⁹⁾ SEVERIN 1921¹⁸⁶⁾),

aster yellows (KUNKEL 1926),¹¹¹⁾ streak of maize (STOREY 1925),²¹⁰⁾ strawberry yellows (PLAKIDAS 1927),¹⁶⁴⁾ rosette of peanuts (STOREY and BOTTOMLEY 1928),²¹⁵⁾ mosaic disease of *Prunella vulgaris* (LIRO 1930),¹²¹⁾ and leaf curl of cotton (KIRKPATRICK 1931).¹⁰²⁾

On the other hand, mosaic diseases of legumes and certain other virus diseases of plants have been reported to be seed-borne. They are bean mosaic (REDDICK and STEWART 1919,¹⁷¹⁾ ARCHIBALD 1921,⁸⁾ KURI-BAYASHI 1926,¹¹⁴⁾ MEULEN 1928,¹⁴³⁾ PIERCE and HUNGERFORD 1929,¹⁶³⁾ MERKEL 1929,¹⁴²⁾ FAJARDO 1928,⁵⁹⁾ 1930⁶⁰⁾), soy bean mosaic (GARDNER and KENDRICK 1921,⁶⁹⁾ 1924¹⁰⁰⁾), lima bean mosaic (McCLINTOCK 1917),¹²⁷⁾ Adzuki bean mosaic (MATSUMOTO 1922),¹²⁵⁾ lettuce mosaic (NEWHALL 1923),¹⁵⁷⁾ cucumber mosaic (DOOLITTLE and GILBERT 1919,⁵¹⁾ DOOLITTLE 1920,⁴⁹⁾ DOOLITTLE and WALKER 1925,⁵⁴⁾ BEWLEY and CORBETT 1930¹⁸⁾), leaf roll of potato (MURPHY and MCKAY 1924,¹⁵¹⁾ ELZE 1931⁵⁷⁾), tobacco ring spot (VALLEAU 1930,²²²⁾ 1932,^{223, 224)} HENDERSON 1931⁷⁵⁾), and Aucuba mosaic of potato (ELZE 1931).⁵⁷⁾ According to DOOLITTLE and WALKER (1925)⁵⁴⁾ cucumber mosaic is not transmitted through the seeds of cucumber, muskmelon, squash and pumpkin, but is transmitted through the seeds of *Micrampelis lobata*. BEWLEY and CORBETT (1930),¹⁸⁾ however, claim that cucumber mosaic is transmitted through cucumber seeds.

As regards the mosaic diseases of broad beans (BÖNING 1927,²⁴⁾ MEULEN 1928,¹⁴³⁾ MERKEL 1929,¹⁴²⁾ FUKUSHI 1930⁶⁷⁾), peas (DOOLITTLE and JONES 1925,⁵²⁾ MEULEN 1928,¹⁴³⁾ MERKEL 1929¹⁴²⁾) and clovers (MEULEN 1928,¹⁴³⁾ MERKEL 1929¹⁴²⁾) evidence has been offered to indicate that they are not seed-borne, although certain workers (DICKSON 1922,⁴²⁾ 1924,⁴³⁾ DICKSON and McROSTIE 1922⁴⁵⁾) claimed that mosaic diseases of peas and clovers were transmitted through the seed. The seed transmission of the tomato mosaic is also a disputed point. WESTERDIJK (1910)²²⁸⁾ and likewise BEWLEY and CORBETT (1930)¹⁸⁾ claimed that it was seed borne while ALLARD (1916),⁴⁾ GARDNER and KENDRICK (1922)⁷¹⁾ and DICKSON (1922)⁴²⁾ stated that the mosaic was not transmitted through tomato seeds.

B) No Transmission through the Soil

It is not infrequent to find in the rice fields that the rice plants grown in close proximity to the dwarf diseased plants are entirely free from the disease. Moreover it has often been noticed that some of the

potted rice plants remained healthy in spite of their roots being intermingled with those of the other plants in the same pot, which have contracted the disease.

These observations seem to indicate that the causal agency of the disease is not soil-borne. In order to obtain further evidence the following experiments were carried out.

July 19, 1929, 8 diseased plants were planted into as many Wagner pots and in each of them 5 seedling plants were planted around the diseased plant. These seedlings were 3 weeks old at this time. None of these 40 plants developed the symptoms throughout the season, although their roots intermingled with those of the diseased plants.

August 20, 1931, 10 porcelain pots, 6 inches in diameter and 6 inches in depth, were filled with infested soil and remains of diseased plants. In each pot was planted one diseased plant. Five of these pots received each 10 two-week-old rice seedlings, which were transplanted around the diseased plant, while the other five pots likewise received each 10 rice seeds sown around the affected plant. Consequently 100 resulting plants were grown in close proximity to the diseased plants. A month later a small wood slip was thrust to a considerable depth at several points into the soil in the pots in order to injure some portion of the root system, which might furnish the entrance of infection. All of these 100 plants remained healthy coming into flower.

The results of these experiments are considered to be additional evidence against the transmission of the disease through the soil.

Most virus diseases of plants are not transmitted through the soil. With the following diseases, there is no evidence in favor of soil transmission: potato leafroll (WORTLEY 1918,²³²) BOTJES 1920,²⁶) MURPHY 1921,¹⁵⁰) KASAI 1921,⁹⁸) SCHULTZ and FOLSOM 1921¹⁸¹), potato mosaic (SCHULTZ and FOLSOM 1920),¹⁸⁰) potato streak (ATANASOFF 1922),⁹) sugar cane mosaic (BRANDES 1919),²⁷) peach rosette (McCLINTOCK 1923),¹²⁸) curly top of sugar beets (SEVERIN 1924),¹⁸⁷) broad bean mosaic (BÖNING 1927),²⁴) beet mosaic (BÖNING 1927),²⁵) spinach blight (McCLINTOCK and SMITH 1918),¹²⁹) cucumber mosaic (DOOLITTLE 1920,⁴⁹) DOOLITTLE and WALKER 1925⁵⁴), rosette of peanuts (STOREY and BOTTOMLEY 1928),²¹⁵) leaf curl of cotton (KIRKPATRICK 1931)¹⁰²) and others.

Certain virus diseases, however, have been stated to be transmitted through the soil. Mosaic disease of wheat and rye has been proved to be so transmitted. (McKINNEY 1923,¹³²) 1925¹³³); WEBB 1928²²⁷).

Mosaic disease of tobacco also has been reported by certain workers to be transmissible through the soil (MAYER 1888,²³⁸) BEIJERINCK 1898,¹⁷) CLINTON 1915,³⁷) MCKINNEY and WEBB 1926,¹⁴⁰) MCKINNEY 1927,¹³⁶) MOUTIA 1928,¹⁴⁶) JOHNSON and OGDEN 1929⁹⁵). In this case, the soil infection appears to depend upon injury to some portion of the root system which happens to come in contact with the infected material. DOOLITTLE (1928)⁵⁰) claims the soil transmission of tomato mosaic and streak in the green house.

C) Attempted transmission with the juice of diseased plant

DAIKUHARA (1904)³⁹) was the first to attempt the transmission of the dwarf disease of rice plant with the juice from the diseased plant. No transmission was obtained by hypodermic injection of the expressed sap from the diseased plant into the veins of the leaves of seedlings. In the Annual Report of the Imperial Agricultural Experiment Station for the year 1912,⁸⁵) it is stated that all attempts to transmit the disease with the juice from the diseased leaves or the ground up bodies of leaf-hoppers have failed.

The results of the writer's experiments along the same line also tend to substantiate the view that the dwarf disease of rice plant cannot be transmitted by mechanical inoculations.

Experiment 1. On August 15, 1923, 19 three-week-old seedlings of rice plant ("Aikoku"-variety) were artificially inoculated by hypodermic injection of the expressed juice from the diseased plant. Each seedling received 0.5 cc. of the inoculum which was injected into the stem near the ground. All the inoculated plants remained healthy throughout the season.

Experiment 2. October 4, 1923, 24 seedlings of the "Aikoku"-variety were inoculated in the same manner as above. These seedlings were also about three weeks old. Each seedling received 1 cc. of the inoculum which had been prepared grinding fresh diseased leaves in a mortar, adding a small quantity of distilled water, until the leaf tissue was thoroughly crushed. No symptoms of the disease appeared on any of the inoculated plants.

Experiment 3. August 8, 1927, 15 rice plants were inoculated by means of the needle puncture method. For the preparation of the inoculum, three diseased plants were cut into pieces and ground in a mortar, adding a small quantity of water until the tissue was thoroughly

crushed. Three inoculations were made on each plant, one near the growing tip, another at the node and the other at the base of the stem. A large drop of the inoculum (about 0.5 cc.) was placed with a sterile scalpel on the desired spot of the stem, and with a sterile needle about 30 pricks were made through the fluid, thus working the latter into the tissue. In no case did the disease appear on the inoculated plant.

Experiment 4. October 8, 1927, 15 seedlings were inoculated by rubbing the leaves with fragments of diseased leaves and slightly crushing them altogether between the fingers. None of these plants showed any signs of the disease.

Experiment 5. October 8, 1927, 18 seedlings were inoculated by means of needles. The inoculum was prepared in the same manner as shown in Experiment 3. Six inoculations were made on each seedling, 3 on the stem as in Experiment 3, and the other 3 on the leaves. No signs of the disease appeared on any of these plants.

Experiment 6. June 29, 1928, 49 seedlings were inoculated by needle punctures as mentioned above or by the use of capillary glass tubes containing the inoculum thrusting into the young part of the stem. All the inoculated plants remained free from the disease.

Experiment 7. July 2, 1929, 30 seedlings were inoculated by needle punctures and scratches. Five grams of the roots, stem and leaves of the diseased plant were ground in a mortar and the fluid was pressed out. A drop of the fluid with macerated tissue was placed on the stem near the ground and scratches were made with a needle through the fluid. In addition, 3 leaves of each seedling were inoculated by needle punctures in the same manner as mentioned before. None of these plants developed the slightest trace of any symptoms.

Experiment 8. July 12, 1929, 22 seedlings were inoculated by rubbing the leaves with the fragments of roots and stem of the diseased plant. None of these plants showed any signs of the disease.

Experiment 9. July 22, 1929, 15 seedlings were inoculated by needle punctures in the same manner as in Experiment 7. None of them developed any symptoms of the disease.

Experiment 10. August 8, 1930, 20 seedlings were inoculated by means of the needle puncture method devised by SEIN (1930) for the artificial transmission of the sugar cane mosaic. A slip about one inch long was cut off from a diseased leaf and placed on the leaf to be inoculated which was bent down and held tightly on a wooden label with the thumb and forefinger of the left hand. The fine insect pin

held in the right hand was thrust rapidly in and out repeatedly through different parts of the diseased leaf into the leaf to be inoculated. Four inoculations were made on each plant, one on the stem and three on the leaves, the pricking being repeated about 50 times on each point. No signs of the disease appeared on any of the inoculated plants.

As shown above, 227 rice plants were inoculated with the juice from the diseased plant by several different mechanical means. But none of these plants showed any symptoms of the disease.

It has been shown that certain virus diseases of plants are not readily transmitted by mechanical means. These are peach yellows (BLAKE, COOK and CONNORS 1921),¹⁹⁾ peach rosette (McCLINTOCK 1923),¹²⁸⁾ aster yellows (KUNKEL 1926),¹¹¹⁾ streak of sugar cane and maize (STOREY 1925),^{208, 209)} soy bean mosaic (GARDNER and KENDRICK 1921),⁶⁹⁾ broad bean mosaic (BÖNING 1927),²⁴⁾ lily mosaic (OGILVIE and GUTERMAN 1929),¹⁵⁸⁾ rosette of peanuts (STOREY and BOTTOMLEY 1928),²¹⁵⁾ virus diseases of brambles (ZELLER 1927,²³⁵⁾ RANKIN 1922¹⁶⁸⁾), strawberry yellows (PLAKIDAS 1927),¹⁶⁴⁾ leaf curl of cotton (KIRKPATRICK 1930,¹⁰¹⁾ 1931¹⁰²⁾) and others.

A number of virus diseases of plants have been reported to be transmissible by mechanical means. In some cases, however, such claims have been disproved or remain unconfirmed. For instance, SCHULTZ and FOLSOM (1923)¹⁸²⁾ claimed that the streak of potato was communicable through the juice from the diseased plant but ATANASOFF (1922)⁹⁾ has been unable to confirm their conclusions. KASAI (1921)⁹⁸⁾ reported that he obtained evidence of the transmission of the potato leaf roll through the juice. SCHULTZ and FOLSOM (1923)¹⁸²⁾ and SCHAFFNIT and JÖHNSEN (1933),¹⁷⁷⁾ however, have failed to transmit it by mechanical inoculations. ROBBINS (1921)¹⁷⁴⁾ and JONES (1931)⁹⁶⁾ were unable to transmit the mosaic disease of sugar beet through the juice while BÖNING (1927),²⁵⁾ working with the same disease, obtained positive results only in a few cases. TOWNSEND (1908)²²¹⁾ and SMITH and BONQUET (1915)²⁰²⁾ failed to produce the curly top of sugar beet by artificial inoculation with the juice from the diseased plant but SEVERIN (1924)¹⁸⁷⁾ claims that he succeeded in transmitting the same disease through the juice.

In the other cases various mosaic diseases of plants have been known to be transmissible through the juice of diseased plant or by leaf mutilation. These diseases are tobacco mosaic (MAYER 1886,¹²⁴⁾ IWANOWSKI 1892⁸⁷⁾ and others), yellow mosaic of tobacco (McKINNEY 1926,¹³⁵⁾ JOHNSON 1927,⁹⁴⁾ SMITH 1928¹⁹⁷⁾), ring spot of tobacco (JOHN-

SON 1925,⁹³⁾ 1927,⁹⁴⁾ FROMME, WINGARD and PRIODE 1927,⁶⁵⁾ WINGARD 1928,²³⁰⁾ PRIODE 1928¹⁶⁶⁾, cucumber mosaic (DOOLITTEL 1920,⁴⁹⁾ DOOLITTLE and WALKER 1923,⁵³⁾ WALKER 1926²²⁵⁾, potato mosaic (SCHULTZ, FOLSOM, HILDEBRANDT and HAWKINS 1919,¹⁸⁴⁾ SCHULTZ and FOLSOM 1920,¹⁸⁰⁾ 1923,¹⁸²⁾ 1925¹⁸³⁾, sweet potato mosaic (ROSEN 1926),¹⁷⁵⁾ sugar cane mosaic (EARLE 1919,⁵⁵⁾ MATZ 1919,¹²⁶⁾ BRANDES 1920²⁹⁾, wheat mosaic (MCKINNEY 1925),¹³³⁾ turnip mosaic (SCHULTZ 1921,¹⁷⁹⁾ GARDNER and KENDRICK 1921⁷⁰⁾, sweet pea mosaic (TAUBENHAUS 1914,²²⁰⁾ DOOLITTLE and JONES 1925⁵²⁾, bean mosaic (FAJARDO 1930),^{60, 61)} spinach blight (McCLINTOCK and SMITH 1918),¹²⁹⁾ mosaic of *Convallaria majalis* (BLATNY 1929),²⁰⁾ tulip mosaic (MCKAY, BRIERLEY, and DYKSTRA 1929,¹³⁰⁾ ATANASOFF 1928¹⁰⁾, hyacinth mosaic (ATANASOFF 1928),¹⁰⁾ narcissus mosaic (ATANASOFF 1928),¹⁰⁾ mosaic disease of *Prunella vulgaris* (LIRO 1930)¹²¹⁾ and others.

D) Transmission by the leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL.

All evidence obtained to date indicates that dwarf disease of rice plant is transmitted only by the agency of the leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL. This apparent obligate relationship between the virus and the insect suggests that the studies along this line may furnish a clue which will throw some light on the nature of the virus of the disease.

Since the symptoms of the disease are so distinct as to allow of no doubt in their recognition, dwarf disease of rice plant is favorable for such an experimental study.

1. *On the leafhopper, Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL., which acts as the insect vector of the disease

Before discussing the transmission of the dwarf disease of rice plant by the agency of the leafhopper, it seems desirable to give a brief account of the morphological characters and life history of the leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL.

The adult is a small yellowish green insect with a length of 4.5 to 6 mm. and an average width of 1.6 mm. across the broadest part of the head, the male being smaller than the female. On the dorsal side: vertex yellowish green with a black transverse marking on the anterior margin; pronotum and scutellum green; elytra yellowish green, with black distal ends in the male and with those slightly tinged with gray

in the female; abdomen black. On the ventral side: thorax and abdomen light brown in the female and black in the male.

The egg is about 1 mm. long, yellowish white, elongate, slightly curved and tapering at one end. The eggs are deposited in the tissues of leafsheaths, usually 15 to 25 in a place and are thrust transversely into the tissues lying one under the other. At first the eggs are scarcely visible since they are pushed in so deeply that they are entirely imbedded in the tissues.

The larvae are at first creamy white and hairy with somewhat indistinct black stripes along both sides of the body. As they grow various minute black spots and an irregular pattern of dark color develop on the back of the thorax and abdomen. The dark-coloured nymphs grow into the male while the light-colored ones into the female. They are very active and leap with surprising agility.

This leafhopper is a 4-, or 5-brooded insect and oviposition extends through a long period of time, which makes the seasonal history of this insect rather complicated. In general this leafhopper overwinters as a nymph, feeding on *Astragalus sinicus* L. and grasses as stated before and matures in April or May, according to the weather conditions. Later the adults begin to appear on the seed beds of rice plant. The adults commence to feed on the rice seedlings at once and if the season is sufficiently advanced, they will soon begin to lay eggs and continue to do so for a long period, each female depositing over one hundred eggs. The eggs hatch in 7-13 days and the larvae mature in about 20 days or more. The leafhoppers multiply abundantly on the rice fields after the rice seedlings are transplanted and 3 or 4 generations of the insect are produced there, the first adults of each brood developing in June to July, August, and September respectively. The eggs of the final brood of the season hatch in September to October and the nymphs hibernate among *Astragalus sinicus* L. or grasses growing on the rice fields.

2. *Viruliferous and non-viruliferous leafhoppers reared on diseased plants*

a) Results of experiments carried out in 1928*

On March 30, fifty individuals of virus-free leafhoppers, the progeny

* This series of experiments were carried out in the green house of the Tottori College of Agriculture and the work subsequent to 1930 was performed in the green house of the Faculty of Agriculture, Hokkaido Imperial University in Sapporo.



An insect cage and glass tubes employed in the writer's experiments

of leafhoppers collected the previous fall on rice plants in the vicinity of the city of Tottori where dwarf disease of rice plant has never been found to occur and retained virus free feeding on healthy rice plants in insect proof cages, were introduced into two insect proof cages enclosing diseased rice plants in a green house. The diseased plants mentioned above were experimentally infected ones by the agency of viruliferous leafhoppers the previous fall. The offspring of the leafhoppers thus reared on diseased rice plants were tested individually for their ability to transmit the disease. Single leafhoppers were placed in glass tubes, about 30–40 cm. long by 3 cm. in diameter, closed at the upper end with a thin cotton cloth, and put on young rice seedlings 10–30 days after germination. The leafhoppers were introduced into the tubes by means of the pipette method originally used by KUNKEL (1926),¹¹¹⁾ and allowed

to remain there for a period of 1-23 days feeding on the rice plants. Insect transfers were made before the window in a small room. Since the leafhopper has a tendency to fly towards a light-source, any leafhopper that escaped when cage was opened went to the window and could be easily recaptured. The leafhoppers were often successfully arrested from escape by means of a black cloth under which the cage was opened.

TABLE 2. Results of transmission experiments with the leafhoppers reared on diseased rice plants in 1928

(Series 1. Experiments with a single leafhopper on each rice plant)

Date of begin. experiment	No. of insect tested	Sex of insect	Duration of feeding (in days)	Result
June 11	No. b 1	male	3	-
"	2	"	"	-
"	3	"	"	-
"	4	"	8	+ (June 30)
"	5	"	"	+ (June 30)
"	6	"	"	-
"	7	"	10	+ (July 3)
"	8	"	"	-
"	9	"	"	-
"	10	"	20	+ (July 1)
"	11	"	"	+ (July 9)
"	12	"	"	+ (July 9)
"	13	"	"	-
"	14	"	"	-
June 23	15	"	3	+ (July 9)
"	16	"	"	-
"	17	"	"	-
June 26	18	"	1	-
"	19	"	"	-
"	20	"	"	-
June 27	21	"	7	+ (July 9)
"	22	"	"	-
"	23	"	"	-
July 4	24	"	5	-
"	25	"	"	-
"	26	"	"	-
July 7	27	"	3	-

Date of begin. experiment	No. of insect tested	Sex of insect	Duration of feeding (in days)	Result
July 7	No. b 28	male	3	-
"	29	"	"	-
"	30	"	"	-
"	31	"	"	-
"	32	"	"	-
"	33	"	"	-
July 11	34	"	1	+ (July 23)
"	35	"	"	-
"	36	"	"	-
"	37	"	"	-
"	38	"	"	-
"	39	"	"	+ (July 30)
"	40	"	"	-
"	41	"	"	-
"	42	"	"	-
"	43	"	"	-
Aug. 7	44	"	"	-
"	45	"	"	-
"	46	"	"	-
"	47	"	"	-
"	48	"	"	-
"	49	"	"	-
Aug. 28	50	"	7	-
"	51	"	23	-
"	52	"	11	-
"	53	"	23	-
"	54	"	"	-
"	55	"	13	-
"	56	"	23	-
"	57	"	"	-
"	58	"	"	-
"	59	"	"	-
"	60	"	"	-
"	61	"	"	-
"	62	"	10	-
"	63	"	23	-
"	64	"	"	-
"	65	"	10	-

Date of begin. experiment	No. of insect tested	Sex of insect	Duration of feeding (in days)	Result
Aug. 28	No. b 66	male	23	—
"	67	"	"	—
"	68	famale	"	—
"	69	"	11	—
"	70	"	7	—
"	71	"	13	—
"	72	"	23	—
"	73	"	7	—
"	74	"	23	—
"	75	"	"	—
"	76	"	10	+ (Oct. 1)
"	77	"	23	—
"	78	"	11	—
"	79	"	23	—
"	80	"	"	—
"	81	"	"	—
"	82	"	"	—
"	83	"	"	—
"	84	"	"	—
"	85	"	"	—
"	86	"	"	—
"	87	"	"	—

(Series 2. Experiments with more than one leafhopper on each plant)

Date of begin. experiment	No. of insects per plant	Duration of feeding (in days)	Result
June 26	3 (male)	1	+ (July 10)
"	"	"	+ (July 11)
"	"	"	—
June 23	"	3	+ (July 3)
"	"	"	+ (July 7)
"	"	"	—
June 27	"	7	+ (July 12)
"	"	"	+ (July 12)
"	"	"	—
May 31	"	10	+ (June 21)
"	"	"	—

Date of begin. experiment	No. of insects per plant	Duration of feeding (in days)	Result
May 31	3 (male)	10	—
"	"	"	—
"	"	"	—
"	" (female)	"	—
"	" (nymph)	"	+ (July 6)
"	"	"	—
"	"	"	—
"	"	"	—
"	"	"	—
June 26	5 (male)	1	—
"	" (female)	"	—
"	"	"	+ (July 12)
June 23	5 (male)	3	—
"	" (female)	"	—
"	"	"	+ (July 9)
June 27	5 (male)	7	+ (July 9)
June 9	" (nymph)	10	+ (July 6)
"	"	"	—
"	"	"	—
"	"	12	—
"	"	"	—
"	"	"	—
June 26	10 (female)	1	—
"	"	"	—
June 23	" (male)	3	+ (July 9)
"	" (female)	"	—
"	"	"	—
June 27	" (male)	7	+ (July 16)

b) Results of experiments carried out in 1930-1931

On April 14, eighty or ninety leafhoppers were collected on *Astragalus sinicus* L. growing on the rice fields in the vicinity of Utsunomiya and two days later about half of these insects were placed in insect proof cages containing dwarf diseased rice plants in the green house. Some of these insects were females, and each produced a numerous progeny. The first nymphs hatched on May 5 and the first adult was bred on June 7 requiring about one month to complete the nymphal

instars. The adults of the second, third and fourth generation began to appear on August 25, December 10 and March 10, 1931, respectively. The adult leafhoppers thus reared on diseased rice plants were tested individually for their capacity to transmit the disease in the same way as shown before. The results of the experiments will be shown in the following table.

TABLE 3. Results of the transmission experiments with the leafhoppers reared on diseased rice plants in 1930-1931

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
June 18	No. c 1	male	68	18-34°C	-
"	2	"	"	"	-
"	3	"	13	14-30	-
July 2	4	"	19	15-33	+ (July 20)
Aug. 2	5	"	21	21-36	-
"	6	"	"	"	-
"	7	"	"	"	-
July 11	8	"	45	20-36	-
"	9	"	28	19-36	+ (Aug. 8)
"	10	"	45	20-36	-
"	11	"	"	"	-
"	12	"	25	19-36	-
"	13	"	"	"	-
"	14	"	"	"	-
"	15	female	"	"	-
July 21	16	"	14	21-36	-
"	17	"	45	20-34	+ (Aug. 14)
Aug. 11	18	male	55	17-35	-
"	19	"	"	"	-
"	20	"	"	"	-
"	21	"	86	15-35	-
"	22	"	55	19-35	-
Sept. 1	23	"	54	14-36	-
"	24	"	30	15-36	+ (Oct. 7)
"	25	"	17	16-36	-
"	26	"	"	"	+ (Sept. 18)
"	27	"	16	"	+ (Sept. 17)
Sept. 15	28	"	25	14-36	+ (Oct. 10)

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Sept. 15	No. c 29	male	52	14-35°C	-
"	30	"	22	15-36	+ (Oct. 7)
"	31	"	35	14-36	-
"	32	"	23	"	+ (Oct. 8)
"	33	"	35	"	-
"	34	"	22	15-36	-
"	35	"	30	14-36	+ (Oct. 15)
"	36	"	25	"	+ (Oct. 10)
Dec. 27	37	"	37	17-31	-
"	38	"	"	"	-
"	39	"	32	"	-
"	40	"	"	"	-
"	41	"	37	"	-
"	42	"	48	"	-
"	43	"	25	"	-
Jan. 7	44	"	26	"	-
"	45	"	"	"	-
"	46	"	34	"	-
"	47	"	26	"	-
"	48	"	34	"	-
"	49	"	26	"	-
"	50	"	"	"	-
"	51	"	34	"	-
"	52	"	66	18-34	-
"	53	"	26	17-31	-
"	54	"	34	"	-
"	55	"	44	"	-
"	56	"	"	"	-
"	57	"	"	"	-
"	58	"	34	"	-
"	59	"	44	"	-
Mar. 24	60	"	"	19-34	-
"	61	"	27	18-33	-
"	62	"	7	18-35	-
"	63	female	46	19-34	-
"	64	"	57	19-35	-
Mar. 30	65	male	21	18-33	-
"	66	female	26	"	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Mar. 30	No. c 67	female	26	18-33°C	—
Apr. 4	68	male	29	19-33	—
"	69	"	24	"	—
Apr. 15	70	"	10	20-35	—
"	71	"	35	20-36	—
"	72	"	8	19-35	—
Apr. 25	73	"	34	19-34	—
"	74	"	"	"	—
"	75	"	25	21-36	—

c) Results of experiments carried out in 1931-1932

On August 14, about 30 non-viruliferous leafhoppers, the progeny of the insects maintained virus free since they had been collected on *Astragalus sinicus* L. the previous year, were placed in an insect proof cage containing dwarf diseased rice plants. The offspring of these leafhoppers reared on diseased rice plants were tested individually for their ability to transmit the disease. The results of the experiments will be shown in the following table. Since almost all the leafhoppers failed to produce infections in healthy plants, the experimental results will be presented in an abridged manner.

TABLE 4. Results of the transmission experiments with the leafhoppers reared on diseased rice plants in 1931-1932

Date of begin. experiment	No. of insects tested	Sex	Duration of feeding (in days)	Temperature	No. of viruliferous insects
Nov. 28	7	male	11-20	19-30°C	0
"	2	"	31-40	20-31	0
"	1	"	51-60	19-30	0
"	1	female	1-10	23-25	0
"	3	"	11-20	19-30	0
"	3	"	21-30	20-31	0
"	2	"	31-40	"	0
"	1	"	51-60	19-30	0
Dec, 1	6	male	11-20	20-31	0
"	5	"	21-30	21-32	0

Date of begin. experiment	No. of insects tested	Sex	Duration of feeding (in days)	Temperature	No. of viruliferous insects
Dec. 1	2	male	31-40	20-31°C	0
"	2	"	41-50	19-31	1 (Jan. 11)
Dec. 7	1	"	1-10	21-31	0
"	3	"	11-20	"	0
"	1	"	21-30	"	0
"	5	"	31-40	20-31	0
Dec. 13	3	"	1-10	21-32	0
"	20	"	11-20	"	0
"	2	"	21-30	20-32	0
"	1	"	31-40	19-30	0
"	6	"	41-50	"	0
"	4	"	51-60	19-31	0
"	2	"	71-80	"	0
Dec. 16	1	"	1-10	20-31	0
"	4	"	11-20	"	0
"	11	"	21-30	19-31	0
"	5	"	31-40	19-30	0
"	3	"	41-50	19-31	0
Dec. 26	1	"	1-10	20-31	0
"	1	"	11-20	19-29	0
"	1	"	21-30	19-30	0
"	1	"	31-40	"	0
"	3	"	41-50	"	0
"	3	"	51-60	"	0
"	1	nymph	21-30	"	0
"	1	"	41-50	"	0
"	4	"	51-60	"	0
"	6	"	61-70	18-30	0
"	7	"	81-90	19-31	0
Jan. 26	2	male	1-10	18-31	0
"	7	"	11-20	19-31	0
"	7	"	21-30	18-31	0
"	4	"	31-40	"	0
"	6	"	41-50	"	0
"	1	"	51-60	18-32	0

d) Results of experiments carried out in 1932-1933

On April 13, about 200 leafhoppers were collected on *Astragalus sinicus* L. growing on the rice fields in the vicinity of Shiraoka near Ômiya and two days later about half their number were introduced into insect proof cages enclosing dwarf diseased rice plants in the green house. The progeny of these insects thus reared on diseased rice plants were tested individually for their ability to transmit the disease in the same way as stated before: viz., each rice seedling was exposed to a single leafhopper in a glass tube throughout its life. The results of the experiments will be shown in the following table.

TABLE 5. Results of transmission experiments with the leafhoppers reared on diseased rice plants in 1932-1933

Date of begin. experiment.	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
June 14	No. e 1	male	42	18-33°C	—
"	2	"	40	"	—
"	3	"	"	"	—
"	4	"	33	17-33	—
"	5	"	38	"	—
"	6	"	20	16-33	—
"	7	"	40	18-33	—
"	8	"	33	17-33	—
"	9	"	40	18-33	—
"	10	"	35	17-33	—
"	11	"	60	18-34	—
June 20	12	"	54	19-34	—
"	13	"	"	"	—
"	14	"	"	"	—
"	15	"	62	18-34	—
"	16	"	37	18-33	—
"	17	"	"	"	—
"	18	"	18	17-33	—
"	19	"	28	"	—
"	20	"	20	"	—
"	21	"	17	"	—
"	22	"	12	"	—

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
June 20	No. e 23	male	40	18-33°C	-
"	24	"	54	19-34	-
"	25	"	36	18-33	-
"	26	"	40	"	-
"	27	"	33	"	-
"	28	"	25	17-32	-
"	29	"	32	18-33	-
"	30	"	"	"	-
"	31	"	33	"	-
June 24	32	"	30	"	-
"	33	"	24	17-33	-
"	34	"	33	18-33	-
"	35	"	25	"	+ (Aug. 3)
"	36	"	30	"	-
"	37	"	44	"	-
"	38	"	24	17-33	-
"	39	"	30	18-33	-
"	40	"	32	"	-
"	41	"	47	18-34	-
"	42	"	39	18-33	-
"	43	"	8	16-33	-
"	44	"	24	17-33	-
"	45	"	58	18-34	-
"	46	"	33	18-33	-
"	47	"	9	16-33	-
"	48	"	14	17-33	-
July 7	49	"	12	18-32	-
"	50	"	11	"	-
"	51	"	7	16-31	-
"	52	"	12	18-32	+ (July 20)
"	53	"	"	"	-
"	54	"	11	"	+ (July 20)
"	55	"	12	"	-
"	56	"	"	"	+ (July 20)
"	57	"	13	"	-
"	58	"	11	"	-
"	59	"	"	"	-
"	60	"	12	"	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
July 7	No. e 61	female	12	18-32°C	—
"	62	"	"	"	—
"	63	"	11	"	+ (July 24)
"	64	"	"	"	—
"	65	"	12	"	—
"	66	"	"	"	—
"	67	"	20	18-34	—
"	68	"	12	18-32	—
"	69	"	"	"	—
"	70	"	11	"	—
"	71	male	12	"	—
"	72	"	11	"	—
"	73	"	7	16-31	—
"	74	"	12	18-32	—
"	75	"	"	"	—
"	76	"	"	"	—
"	77	"	"	"	—
"	78	"	"	"	—
"	79	"	"	"	+ (July 20)
"	80	"	"	"	—
"	81	"	"	"	+ (July 21)
July 20	82	"	20	20-36	—
"	83	"	"	"	—
"	84	"	21	"	—
"	85	"	"	"	—
"	86	"	18	"	—
"	87	"	"	"	—
"	88	"	24	"	—
"	89	"	5	20-37	—
"	90	"	14	20-36	—
"	91	"	"	"	—
"	92	"	"	"	—
"	93	"	"	"	—
"	94	"	7	21-36	—
"	95	"	20	20-36	—
"	96	"	24	"	—
"	97	"	"	"	—
"	98	"	28	"	—

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
July 20	No. e 99	female	11	21-36°C	-
"	100	"	26	20-36	-
"	101	"	16	"	+ (Aug. 5)
"	102	"	"	"	-
"	103	"	11	21-36	+ (Aug. 5)
"	104	"	62	18-34	-
"	105	"	24	20-36	-
"	106	"	16	"	+ (Aug. 5)
"	107	"	32	"	-
"	108	"	11	21-36	+ (Aug. 5)
"	109	"	24	20-36	-
July 30	110	male	5	18-36	-
"	111	"	11	"	-
"	112	"	22	19-36	-
"	113	"	"	"	-
"	114	"	16	20-36	+ (Aug. 15)
"	115	"	17	"	-
"	116	"	37	19-36	-
"	117	"	5	18-36	-
"	118	"	11	"	-
"	119	"	14	20-36	-
"	120	"	21	"	-
"	121	"	43	18-35	-
"	122	"	10	18-36	-
"	123	"	11	"	-
"	124	"	22	19-36	-
"	125	"	38	"	-
"	126	"	18	20-36	-
"	127	female	17	"	-
"	128	"	14	"	+ (Aug. 24)
"	129	"	17	"	+ (Aug. 19)
"	130	"	5	18-36	-
"	131	"	14	20-36	+ (Aug. 18)
"	132	"	17	"	-
Aug. 15	133	male	31	18-34	-
"	134	"	21	18-35	-
"	135	"	27	18-34	-
"	136	"	10	18-36	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Aug. 15	No. e 137	male	12	18-36°C	-
"	138	"	31	18-34	-
"	139	"	4	17-37	+ (Aug. 23)
"	140	"	16	18-36	-
"	141	"	8	18-37	-
"	142	"	22	18-35	-
"	143	"	31	18-34	-
"	144	"	9	18-37	+ (Aug. 24)
"	145	"	6	18-36	-
"	146	"	"	"	-
"	147	female	"	"	+ (Aug. 26)
"	148	"	"	"	-
"	149	"	"	"	-
"	150	"	"	"	-
"	151	"	14	18-37	+ (Sept. 15)
"	152	"	10	18-36	-
"	153	"	"	"	-
"	154	"	"	"	-
"	155	"	9	18-37	-
"	156	"	"	"	-
"	157	"	22	18-35	-
"	158	"	9	18-37	-
"	159	"	15	"	-
"	160	"	"	"	-
"	161	"	"	"	-
"	162	"	10	18-36	-
Sept. 6	163	male	39	13-33	-
"	164	"	54	14-33	-
"	165	"	"	"	-
"	166	"	46	"	-
"	167	"	37	13-33	-
"	168	"	23	13-32	-
"	169	"	"	"	+ (Sept. 29)
"	170	"	10	16-34	-
"	171	"	"	"	-
"	172	"	43	13-33	-
"	173	"	"	"	-
"	174	"	52	14-33	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Sept. 6	No. e 175	male	39	13-33°C	-
"	176	"	23	13-32	-
"	177	"	39	13-33	-
"	178	"	54	14-33	-
"	179	"	59	15-34	-
"	180	"	54	14-33	-
"	181	"	10	16-34	+ (Sept. 29)
"	182	"	52	14-33	-
"	183	"	59	15-34	-
"	184	"	42	13-33	-
"	185	"	23	13-32	-
"	186	"	33	13-33	-
"	187	"	42	"	-
"	188	"	23	13-32	-
"	189	"	42	13-33	-
"	190	"	23	13-32	-
"	191	female	"	"	-
"	192	"	42	13-33	-
"	193	"	"	"	-
"	194	"	"	"	-
"	195	"	26	13-32	-
"	196	"	33	13-33	-
Sept. 7	197	male	9	16-33	-
"	198	"	36	12-33	-
"	199	"	"	"	+ (Oct. 22)
"	200	"	40	13-33	-
"	201	"	9	16-33	-
"	202	"	34	13-33	-
"	203	"	22	"	-
"	204	"	"	"	-
"	205	"	"	"	-
"	206	"	41	"	-
"	207	"	22	"	+ (Sept. 29)
"	208	"	"	"	-
"	209	female	"	"	-
"	210	"	45	14-33	-
"	211	"	58	15-34	-
"	212	"	22	13-33	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Sept. 7	No. e 213	female	22	13-33°C	-
"	214	"	25	"	-
"	215	"	41	"	-
"	216	"	25	"	-
"	217	"	22	"	+ (Sept. 29)
"	218	"	"	"	-
"	219	"	"	"	-
"	220	"	9	16-33	-
"	221	"	41	13-33	-
"	222	"	22	"	-
"	223	"	59	15-34	-
"	224	"	22	13-33	-
"	225	"	59	15-34	-
"	226	"	32	13-33	-
"	227	"	45	14-33	-
Sept. 8	228	male	6	16-33	+ (Sept. 25)
"	229	"	27	14-32	-
"	230	female	31	13-33	+ (Oct. 19)
"	231	"	8	16-33	+ (Sept. 29)
"	232	"	40	13-33	-
"	233	"	21	"	-
"	234	"	8	16-33	-
"	235	"	40	13-33	-
Sept. 22	236	male	10	13-31	+ (Oct. 2)
"	237	"	44	14-34	-
"	238	"	"	"	-
"	239	"	"	"	-
"	240	"	7	13-32	+ (Sept. 29)
"	241	"	39	14-34	-
"	242	"	32	13-34	-
"	243	"	44	14-34	-
"	244	"	38	"	-
"	245	"	44	"	-
"	246	"	39	"	-
"	247	"	12	13-33	-
"	248	"	44	14-34	-
"	249	"	"	"	-
"	250	female	10	13-31	-

Date of begin. experiment	No. of insect tested	Sex	Duration of feeding (in days)	Temperature	Result
Sept. 22	No. e 251	female	26	12-33°C	-
"	252	"	"	"	-
"	253	"	10	13-31	-
"	254	"	44	14-34	-
"	255	"	27	13-33	-
"	256	"	13	13-32	+ (Oct. 5)
"	257	"	44	14-34	-
Sept. 30	258	male	22	"	-
"	259	"	11	12-33	-
"	260	"	22	14-34	-
"	261	"	"	"	-
"	262	"	2	15-31	-
"	263	female	22	14-34	-
"	264	"	"	"	-
"	265	"	31	17-34	-
"	266	"	36	"	-
"	267	"	22	14-34	-
"	268	"	"	"	-
"	269	"	19	12-34	-
"	270	"	36	17-34	-
"	271	"	"	"	-
"	272	"	11	12-33	+ (Oct. 11)
"	273	"	36	17-34	-
"	274	"	8	12-34	-
Oct. 19	275	"	12	19-35	+ (Oct. 31)
"	276	"	17	20-34	-
"	277	"	"	"	-
"	278	"	11	19-34	-
"	279	"	7	"	+ (Oct. 26)
"	280	"	17	20-34	-
"	281	"	15	"	-
"	282	"	3	"	-

Subsequent to November 15, almost all leafhoppers tested failed to produce infection in healthy plants and the results will be presented in an abridged manner.

Date of begin. experiment	No. of insects tested	Sex	Duration of feeding (in days)	Temperature	No. of viruliferous insects
Nov. 15	1	male	21-30	20-30°C	0
"	1	"	31-40	"	0
"	1	female	1-10	19-30	0
"	3	"	11-20	"	0
Dec. 1	1	male	1-10	20-31	0
"	1	"	11-20	"	0
"	2	female	"	"	0
Mar. 3	6	male	20	20-33	0
4	4	"	"	"	0
"	1	female	"	"	0
6	14	nymph	"	"	0
15	18	male	"	19-35	0
17	5	"	"	"	0
"	1	female	"	"	0
19	6	male	"	19-36	0
Apr. 24	5	"	"	20-38	0
May 12	7	"	"	20-35	0
June 1	8	"	"	13-35	0
22	10	"	"	18-38	0

The results shown in the above tables will be summarized as follows.

TABLE 6. Summarized results of transmission experiments with the leafhoppers reared on diseased rice plants

Year	Duration of feeding (in days)	No. of insects tested	No. of infective insects	Percent. of infective insects
1928	1-10	50 (♀ 3)	8 (♀ 1)	16
	11-20	10 (♀ 3)	3	30
	21-30	27 (♀ 14)	0	0
		87	11	13
1930	1-10	3	0	0
	11-20	6 (♀ 1)	4 (♀ 1)	67
	21-30	69 (♀ 3)	7	24
	31-40	15	0	0
	41-50	11 (♀ 2)	0	0
	51-60	7 (♀ 1)	0	0
	61-70	3	0	0
	81-90	1	0	0
	75	11	15	

Year	Duration of feeding (in days)	No. of insects tested	No. of infective insects	Percent. of infective insects
1931	1-10	9 (♀ 1)	0	0
	11-20	52 (♀ 3)	0	0
	21-30	31 (♀ 3, n* 1)	0	0
	31-40	22 (♀ 2)	0	0
	41-50	20 (n 1)	1	5
	51-60	14 (♀ 1, n 4)	0	0
	61-70	6 (n 6)	0	0
	71-80	2	0	0
	81-90	7 (n 7)	0	0
			163	1
1932	1-10	46 (♀ 24)	9 (♀ 3)	20
	11-20	173 (♀ 52, n 14)	19 (♀ 15)	11
	21-30	61 (♀ 24)	4 (♀ 1)	7
	31-40	51 (♀ 12)	2 (♀ 1)	4
	41-50	27 (♀ 9)	0	0
	51-60	17 (♀ 4)	0	0
	61-60	2 (♀ 1)	0	0
			377	34

* n stands for nymph.

As shown above, 702 leafhoppers which had been reared upon dwarf diseased rice plants were tested singly for their capability of transmitting the disease, and only 57 (or 8 per cent) of them proved to be infective. Of the leafhoppers tested 506 were male, 163 female and the others nymphs. Among them 35 males and 22 females proved to be infective, the percentage of infective males and females amounting to 7 and 13, respectively. These infective leafhoppers were able to produce infections in healthy rice plants when allowed to feed upon them for a comparatively short period. The results shown above appear to justify the conclusion that certain individuals of the leafhoppers reared on dwarf-diseased rice plants are incapable of transmitting the disease when single leafhoppers are confined to each healthy rice plant.

In his studies on aster yellows, KUNKEL (1926)¹¹¹⁾ found that "most, if not all, individuals of *Cicadula sexnotata* FALL. were capable of taking up the aster yellow virus" when they were allowed to feed on yellow diseased aster plants. STOREY (1925,²¹⁰⁾ 1928²¹²⁾, on the other

hand, asserted that certain individuals of the leafhopper, *Cicadulina mbila* (*Balclutha mbila* NAUDE) remained uninfective, even when they fed upon streak diseased maize plant through the whole course of development of the insect from the first instar to the adult stage.

3. *Length of time infective leafhoppers must remain on rice plants to transmit the disease*

A series of experiments were carried out to ascertain the minimum length of time infective leafhoppers must remain on healthy rice plants to transmit the disease. Single leafhoppers, experimentally proved to

TABLE 7. Results of experiments on the relation between the length of feeding period and the number of infection obtained

Date	Insect No.	Sex	Length of time insects remained on plants				
			5	10	20	30	60 (min.)
Sept. 22'30	e 26	♂					—
Sept. 10'30	e 30	„					—
„	e 32	„					+
Aug. 19'32	e 114	„				+	(21)
Oct. 4'32	e 169	„	—	—	—	+	(18)
„	e 207	„	—	—	—	+	(19)
„	e 236	„	—	—	+	+	(25)
„	e 240	„	—	—	—	—	—
Oct. 5'32	e 256	♀	—	—	—	+	(16)
Oct. 12'32	e 272	„	—	—	—	+	(21)
Oct. 25'32	e 151-4	„	—	—	—	+	(14)
Oct. 27'32	e 151-1	„	—	+	—	+	(25)
Jan. 2'33	e 151-4-13	„	—	—	—	+	(15)
Jan. 7'33	e 151-1-26	„	+	—	+	—	(12)
„	e 151-1-6	„	+	+	+	+	(10)
Jan. 9'33	e 151-1-13	♂	—	—	—	+	(9)
			—	—	—	+	(9)
			+	—	+	—	(10)
			+	+	+	+	(10)
			—	—	—	+	(16)
			—	—	—	+	(16)
			—	—	—	+	(14)
			—	—	—	+	(14)
			2/12	2/12	4/12	9/13	11/16
Length of incubation period (in days)			10-24	9-12	16-25	9-25	9-25

The numbers in parentheses indicate incubation periods in days.

be infective, were confined to each young rice plant in a glass tube. When the leafhopper alighted on the leaf of rice plant and appeared to begin sucking the juice, the time was recorded and the insect was allowed to remain there during a definite length of time. It was not definitely known whether the leafhopper was actually sucking the juice all the time. The results of the experiments will be shown in table 7.

As shown in the table, most of the viruliferous leafhoppers were capable of transmitting the disease to healthy plants when they were allowed to feed on the plants for 30 to 60 minutes, and as short a period as 5 minutes was sufficient to transmit the disease. This indicates that only a small amount of virus may produce a typical case of dwarf disease of rice plant. It presents a striking contrast that certain individuals of the leafhoppers reared on dwarf diseased rice plants are capable of transmitting the disease to healthy plants after only 30-60 minutes' feeding whereas some leafhoppers from the same culture on diseased plants cannot produce infections in healthy plants even though allowed to feed on them as long as they live. SMITH and BONCQUET (1915),²⁰²⁾ working with the curly top of beets, reported that exposure of beets for 5 minutes to feeding by infective leafhoppers was sufficient to cause the disease. In their studies on spinach blight, McCLINTOCK and SMITH (1918)¹²⁹⁾ also found that virus bearing aphids (*Macrosiphum solanifolii* and *Rhopalosiphum persicae*) produced infections in healthy plants when allowed to feed on them for 5 minutes. It seems that these investigators worked with a lot of insects, instead of single aphids. According to KURIBAYASHI (1931),¹¹⁶⁾ stripe disease of rice plant was transmitted to healthy plants when single infective leafhoppers (*Delphacodes striatellus* FALL.) were allowed to feed on them for a period of 5 hours.

Incubation periods varied, as shown in the table, from 9 to 25 days, nearly as long as those when the feeding time was prolonged to one day, the incubation periods in the latter case ranging from 6 to 37 days, generally from 8 to 24 days.

4. Retention of infective power by the leafhopper

In 1915 Murata stated in his manual entitled "Insect pests of rice, barley and wheat and their control measures", that infective leafhoppers would partly or entirely lose their ability to induce the dwarf disease of rice plant when they were repeatedly transferred to new healthy plants at 2 or 3 day intervals. He has published, however, no papers presenting his experimental data. Since this aspect of the

problem seems to have significant bearing upon the nature of the virus, it has been further investigated by the present writer by transferring single infective leafhoppers daily or at several day intervals to new healthy rice plants. Single leafhoppers, experimentally proved to be infective were confined to each young rice plant of "Aikoku"-variety, 10 to 30 days after germination, in a glass tube, allowed to remain there during a definite length of time and then transferred to new healthy plants. The results of experiments will be presented in the tables, 8 and 9.

TABLE 9. Results of transmission experiments with infective leafhoppers transferring the insects to new healthy plants at several day intervals (1930 and 1932)

e 4 (♂)	July 21-22 22-23 23-30 30-31 31-2 2-5 5-8 8-9 9-11 11-14 14-21. + + + + + + + + + + + +
e 30 (♂)	Oct. 10-13 13-16 16-23 23-27 27-30 30-2 2-5 5-8 8-11. - - - + - - - - +
e 32 (♂)	Oct. 10-13 13-16 16-20 20-23 23-27 27-30 30-2 2-5 5-8 8-22. + + + + + + + + - +
e 114 (♂)	Aug. 16-19 19-22 22-25 25-28 28-31 31-3 3-6 6-9. + + + + + + + -
e 169 (♂)	Sept. 29-2 2-5 5-8 8-11 11-13. + + + + +
e 236 (♂)	Oct. 2-5 5-8 8-11 11-14 14-17. + + + + +
e 240 (♂)	Sept. 29-2 2-5 5-8 8-11 11-14 14-17 17-20. + + + + + + +

As shown in these tables, there is a marked variability as to the infectivity of each viruliferous individual of leafhoppers. Most leafhoppers retained their infective power as long as they lived or during a period varying from 12 to 33 days, and certain leafhoppers produced infections in every healthy plant on which it was confined for 24 hours on consecutive days. In comparatively few leafhoppers, however, their infective power was apparently reduced or lost even under favorable conditions, when each individual was supplied with a healthy rice plant at one to several day intervals. The leafhopper, e279-1 which lived for a period over 50 days retained the virus for 33 days and then remained free from virus throughout its subsequent life. BONCQUET and STAHL

(1917)²³⁾ reported that the ability of the beet leafhopper, *Eutettix tenella* BAKER, to transmit curly top was lost in from 15 to 35 days if the insects were transferred daily to healthy beets. SEVERIN (1924), on the other hand, stated that infective beet leafhoppers retained their infectivity during all of the nymphal stages, after each molt, and during the entire adult life. CARSNER (1919)³³⁾ kept infective beet leafhoppers on apparently non-susceptible plants for over one hundred days without loss of infectivity. In his studies on aster yellows, KUNKEL (1926)¹¹¹⁾ found that while many individuals of the leafhopper, *Cicadula sexnotata* FALL. retained the virus through life, some individuals seemed to lose it after a short time. In the case of *Cicadulina (Balclutha) mbila* NAUDE which acts as the vector of maize streak disease, STOREY (1928)²¹²⁾ reports that "while the infective power is usually not lost by the leafhopper, such loss may occasionally occur."

Incubation periods varied from 6 to 37 days, generally 8 to 24 days, when single leafhoppers were confined on each seedling plant for one day. It seems that the length of incubation period depends not only upon the infective power of the leafhopper and the duration of its feeding period but also upon the growing state of the infected plants as well as the environmental conditions which affect plant growth. Especially, low temperatures retard the development of the disease.

It is worthy of note that not all the infested plants contracted the dwarf disease. Occasionally plants in the progressive series of infections remained healthy whereas the other plants in the same series developed the disease, as shown in the cases of leafhoppers, e256, e151-4-9, e279-1 etc. Similar cases have been recorded in the curly top of sugar beet and other virus diseases of plants. In their studies on the curly top of beets, CARSNER and STAHL (1924)³⁴⁾ state that "a viruliferous leafhopper may not produce the disease each time it feeds on a healthy plant, even though the periods of feeding be 24 or 48 hours or even longer." STOREY (1928)²¹²⁾ in his studies of maize streak disease finds that when viruliferous single leafhoppers were transferred daily to new healthy plants, frequent failures of infection occurred on some plants upon which they fed. SAMUEL et al. (1930)¹⁷⁶⁾ working with "spotted wilt" of tomatoes and its transmission by the thrips, *Frankliniella insularis*, find that infective individuals fed for successive days on healthy tomato plants did not infect every plant on which they fed. SMITH (1931)²⁰¹⁾ in his studies on the insect transmission of potato leafroll writes that occasionally plants in the pro-

gressive series of infections with viruliferous aphids failed to develop leafroll, although plants before and after in the series became infected.

Under the writer's experimental conditions the failures of infection on plants, upon which viruliferous leafhoppers fed, may be partly explained on the assumption that the leafhoppers had not actually fed on those plants, because some leafhoppers could survive 3 or 4 days without feeding, as shown by the following experiment.

Each leafhopper was kept in a glass tube as mentioned before, containing no food plant and standing on the moistened soil in a pot. The number of insects dead was recorded daily until they all died out.

The results are shown in the following table.

TABLE 10. Results of starvation experiments with the leafhopper

Series 1. With insects reared on dwarf diseased rice plants
(Jan. 22-Mar. 9 1933, temp: 18-32°C)

Duration of starvation (in hours)	No. of insects dead	No. of insects survived
18-24	9 (♀ 9, ♂ 0)	21 (♀ 18 ♂ 3)
24-48	6 (♀ 5, ♂ 1)	15 (♀ 13 ♂ 2)
48-72	12 (♀ 10, ♂ 2)	3 (♀ 3 ♂ 0)
72-96	1 (♀ 1, ♂ 0)	2 (♀ 2 ♂ 0)
96-120	2 (♀ 2, ♂ 0)	0
	30 (♀ 27, ♂ 3)	

Series 2. With insects reared on healthy rice plants
(Jan. 5-14 1933, temp: 20-28°C)

Duration of starvation (in hours)	No. of insects dead	No. of insects survived
18-24	12 (♀ 8, ♂ 4)	18 (♀ 11 ♂ 7)
24-48	5 (♀ 3, ♂ 2)	13 (♀ 8 ♂ 5)
48-72	8 (♀ 6, ♂ 2)	5 (♀ 2 ♂ 3)
72-96	4 (♀ 1, ♂ 3)	1 (♀ 1 ♂ 0)
96-120	1 (♀ 1, ♂ 0)	0
	30 (♀ 19, ♂ 11)	

Only a small portion of the leafhoppers could survive more than 3 days without food as shown in the table. And moreover it can be hardly explained why the leafhoppers would not take food for as long

as 5 days, as in the cases of e151-4-9 and e151-1-23 in table 7, in spite of the presence of food plants. As a matter of fact, no leafhopper was observed remaining apart from the food plant longer than a day. At least in such cases, one inevitably supposes that certain viruliferous leafhoppers occasionally fail to produce infections in healthy plants, probably owing to the temporary exhaustion of the virus in the salivary glands and the anterior portion of the alimentary tract.

As will be stated later, the virus of dwarf disease of rice plant may be transmitted through eggs from viruliferous female leafhoppers to their offspring and certain nymphs emerging from these viruliferous eggs are capable of producing infections in healthy rice plants. Accordingly it seemed desirable to determine how long the virus is retained in the insect body when these viruliferous nymphs are transferred daily to new healthy rice plants. Single nymphs which had emerged from the eggs of viruliferous females were confined on each young rice plant of "Bôzu No. 5" variety in a glass tube, allowed to remain there one day and transferred to new healthy plants. The tiny nymphs which had just emerged from eggs could be readily transferred to rice plants by means of a small camel's hair brush as will be described later. However, some difficulties were experienced to manipulate the nymphs 2-5 days of age, since they are very active and leap with surprising agility. The transfer could be best effected by means of the pipette method as stated before, putting the nymph on the soil near the rice plant on to which it later crawled up. When the insects grow older they can be transferred more readily to new healthy plants. The results of experiments will be presented in table 11.

As shown in the table, all the infective nymphs tested retained their infectivity during all of the nymphal stages, the virus being retained during the process of moulting, and through the entire adult life or for a period of 50-90 days, without access of the virus. In this series of experiment also, most of the infective leafhoppers occasionally failed to produce infections in healthy rice plants. It appears that a period from 1 to 14 days must elapse before most of the infective nymphs become capable of producing infections in healthy plants although certain individuals may be infective immediately after the emergence from eggs. This admits several interpretations: either (1) the tiny newly emerged nymphs may be able only to transfer a dose of the virus insufficient to produce infection in a healthy rice plant, or (2) some developmental change or multiplication of the virus may occur within

the insect body before it is fully infective, or (3) the virus may migrate from the other parts of the body to the salivary glands and the anterior portion of alimentary tract before it is injected into the plant tissue through the proboscis.

5. *Virulence of the progeny of the infective leafhoppers*

As stated before, only a small portion of the leafhoppers reared on dwarf diseased rice plants were capable of transmitting the disease and most of the infective leafhoppers could produce infections in healthy plants when they were allowed to feed upon the plants less than one hour. This appeared to suggest the presence of "active" and "inactive" races of leafhoppers in the transmission of the disease as in the case of the leafhopper, *Cicadulina mbila* which acts as vector of the virus of maize streak.²¹⁴⁾

A few infective leafhoppers were introduced in an insect proof cage enclosing dwarf diseased rice plants. Each female leafhopper soon produced a numerous progeny. The younger leafhoppers, adult or nymphs just before the final moulting, thus reared on diseased rice plants were tested singly for their capability of transmitting the disease by means of the "single hopper culture method" described before. The results obtained will be shown in the following table.

TABLE 12. Results of transmission experiments with the progeny of infective leafhoppers, reared on diseased plants

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 1	male	Feb. 2, 1933	Feb. 25
2	"	"	Feb. 14
3	"	"	Feb. 17
4	"	"	Feb. 12
5	"	"	Feb. 15
6	nymph (subsequently ♀)	Mar. 6	Mar. 16
7	" (♀)	"	no infection
8	"	"	Mar. 18
9	" (♀)	"	Mar. 15
10	" (♀)	"	Mar. 13
11	"	"	"

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 12	nymph (subsequently ♀)	Mar. 6	Mar. 13
13	„ (♀)	„	Mar. 14
14	„ (♂)	„	Mar. 25
15	„	„	Mar. 18
16	„	„	Mar. 13
17	„	„	Mar. 12
18	„ (♂)	„	Mar. 17
19	„	„	Mar. 14
20	„ (♂)	„	Mar. 16
21	„ (♂)	„	„
22	„ (♂)	„	Mar. 19
23	„	Mar. 21	Apr. 2
24	„	„	Apr. 1
25	„	„	Apr. 2
26	„	„	Mar. 30
27	„	„	Apr. 5
28	„	„	Apr. 3
29	„ (♂)	„	„
30	„	„	Apr. 6
31	„	„	Apr. 4
32	„	„	Apr. 1
33	„	„	„
34	„ (♀)	„	„
35	„	„	„
36	„ (♂)	„	Apr. 2
37	„	„	Apr. 6
38	„	„	Mar. 30
39	„ (♀)	„	„
40	„	„	Apr. 2
41	„	„	Apr. 1
42	„	„	Apr. 3
43	„	„	Apr. 4
44	„	„	Apr. 6
45	„	„	Mar. 31
46	„	„	Apr. 1
47	„ (♂)	„	Apr. 2
48	„	May 19	May 28

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 49	nymph (subsequently ♀)	May 19	June 5
50	„ (♀)	„	May 28
51	„ (♀)	„	May 27
52	„ (♂)	„	May 26
53	„ (♀)	„	June 3
54	„ (♂)	„	June 6
55	„ (♀)	„	June 5
56	„ (♀)	„	May 30
57	„ (♂)	May 26	June 10
58	„ (♂)	„	June 13
59	„ (♂)	„	„
60	„ (♂)	„	June 6
61	„ (♀)	„	June 24
62	„ (♀)	„	June 6
63	„ (♂)	„	June 16
64	„ (♂)	„	June 6
65	„ (♀)	„	June 8
66	„ (♀)	„	„
67	„ (♂)	„	June 6
68	„ (♂)	„	June 11
69	„ (♀)	„	June 24
70	„ (♀)	„	June 6
71	„ (♂)	„	June 8
72	„ (♂)	„	June 13
73	female	July 13	July 20
74	male	„	„ 19
75	„	„	„ 21
76	nymph	„	„ 24
77	„	„	„ 25
78	female	„	„ 18
79	„	Aug. 26	Sept. 2
80	„	„	„ 5
81	„	„	no infection
82	„	„	Sept. 2
83	„	„	„ 3
84	„	„	no infection
85	„	„	Sept. 9

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 86	female	Aug. 26	no infection
87	"	"	Sept. 2
88	"	"	" 20
89	male	"	" 10
90	"	"	" 4
91	"	"	" 2
92	"	"	" 2
93	"	"	no infection
94	"	"	Sept. 4
95	"	"	" 1
96	"	"	" 6
97	"	"	" 4
98	"	"	" 4
99	"	"	" 2
100	"	"	" 4
101	"	"	" 4
102	"	"	" 20
103	"	"	" 4
104	"	"	no infection
105	"	"	Sept. 3
106	"	"	" 4
107	"	"	" 1
108	nymph	"	" 1
109	" (♀)	"	" 2
110	" (♀)	"	no infection
111	"	"	Sept. 4
112	"	"	" 2
113	"	"	" 6
114	male	"	" 2
115	female	Nov. 2	Nov. 16
116	"	"	" 11
117	"	"	Dec. 14
118	"	"	no infection
119	male	"	"
120	female	"	Nov. 11
121	"	"	" 11
122	nymph (♀)	Nov. 9	" 24
123	"	"	" 23

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 124	nymph	Nov. 9	Nov. 18
125	" (♀)	"	no infection
126	"	"	Nov. 24
127	"	"	" 21
128	" (♀)	"	" 21
129	" (♂)	"	" 21
130	" (♂)	"	" 22
131	" (♀)	"	" 24
132	"	"	" 24
133	" (♀)	"	" 18
134	" (♀)	"	" 26
135	" (♂)	"	" 24
136	" (♀)	"	" 27
137	"	"	" 23
138	" (♀)	"	" 21
139	" (♂)	"	Dec. 1
140	"	"	Nov. 19
141	" (♀)	"	" 21
142	"	"	" 23
143	" (♀)	"	" 23
144	" (♀)	"	Dec. 14
145	"	"	Nov. 28
146	" (♀)	"	" 21
147	" (♀)	"	" 21
148	" (♀)	"	Dec. 14
149	" (♂)	"	Nov. 23
150	" (♀)	Nov. 25	Dec. 30
151	" (♂)	"	" 18
152	" (♀)	"	" 26
153	"	"	no infection
154	"	"	Dec. 13
155	" (♀)	"	no infection
156	"	"	"
157	"	"	"
158	"	"	"
159	" (♀)	"	Dec. 14
160	" (♀)	"	" 14
161	"	"	" 15

Insect No.	Sex	The date of transfer of the insect to healthy plant	Appearance of the symptom
e' 162	nymph	Nov. 25	Dec. 27
163	"	"	" 20
164	"	"	" 11
165	" (♀)	"	" 14
166	"	"	no infection
167	" (♂)	"	Dec. 11
168	" (♀)	"	" 11
169	"	"	" 15

As shown above, 153 individuals out of 169 leafhoppers tested produced infections in healthy rice plants, which began to show the symptom 6–29 days, mostly 9–13 days after the insects were transferred to these plants. In other words almost all the progeny of infective leafhoppers were viruliferous when they had been reared upon diseased plants whereas a large proportion of leafhoppers from the other colonies bred on diseased plants were free from virus. This admits two interpretations: either the offspring of infective leafhoppers may have “inherited” the virus from their parents or they may have the predisposition to carry the virus readily after feeding upon diseased rice plants.

In parallel to the experiments above mentioned, a series of experiments were started to determine whether the infective power is “inheritable” to the offspring by breeding successive generations from infective parents or from crosses between infective females and uninfected males and vice versa.

A pair of male and female leafhoppers, either one or both infective, were confined on a young healthy rice plant in a glass tube and allowed to remain there for one day. Subsequently they were transferred to new healthy rice plants every day during which period oviposition occurred. The precaution was taken to confine single nymphs on rice plants in separate glass tubes and to allow them to remain there until they grew into adults in order to obtain female leafhoppers which had never copulated. The eggs are laid in the leafsheath at a few spots near the ground and thrust transversely into the tissue lying one under the other. A few days after oviposition the spot where the eggs are deposited becomes easily discernible by its slightly discolored and more

or less raised surface. The eggs hatch in about 10 days. The tiny nymphs just emerged from eggs were transferred to healthy rice plants taking the necessary precaution so that they had no opportunity to feed upon the plants on which the eggs had been deposited and which might have been affected with the disease in consequence of the infestation of the infective parents. Daily observations were continued as the hatching of the eggs approached and the nymphs which had just emerged from the eggs were transferred singly to a young healthy rice plant in a glass tube by means of a small camel's hair brush. The eggs usually hatch in the early morning and in a similar manner to that described by STAHL and CARNSNER (1918)²⁰⁶ and also by SEVERIN (1921)¹⁸⁶ in respect to the beet leafhopper, *Eutettix tenella*. Keeping the plants which harbour the eggs in a dark and cool place the hatching of eggs could be retarded to some extent. When such plants were placed in the sunlight, some of these eggs immediately began to hatch out. Another method of transfer of the nymphs is as follows: the leafsheath with eggs is cut off 4-5 cm. long and leaned against healthy rice plants in a glass tube. When the eggs hatch the tiny nymphs immediately migrate to the healthy plants crawling up the stems and they later can be easily transferred singly to a young healthy rice plant in a glass tube. The results of experiments will be shown in the following tables.

TABLE 13. Virulence of the progeny of infective leafhoppers

Infective female × uninfected male			
No. e 279 (♀) × a (♂)			
Insect No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e 279-1	Oct. 29' 32	Nov. 21	female
e 279-2	"	24	"
e 279-3	"	30	"
e 279-4	"	22	"
e 279-5	"	24	"
e 279-6	"	"	"
e 279-7	"	"	male
e 279-8	"	16	female
e 279-9	"	18	"

Insect No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e 279-10	Oct. 30	Nov. 25	prematurely died
e 279-11	"	"	male
e 279-12	"	21	prematurely died
e 279-13	31	28	female

No. e151-4 (♀) × b (♂)

e 151-4-1	Nov. 4	Dec. 23	prematurely died
e 151-4-2	"	25	"
e 151-4-3	"	no infection	female
e 151-4-4	"	Dec. 14	"
e 151-4-5	5	15	prematurely died
e 151-4-6	"	2	female
e 151-4-7	6	12	"
e 151-4-8	"	25	"
e 151-4-9	"	5	male
e 151-4-10	Nov. 7	Dec. 15	prematurely died
e 151-4-11	"	5	"
e 151-4-12	8	Nov. 26	female
e 151-4-13	"	Dec. 29	"
e 151-4-14	9	7	male
e 151-4-15	10	14	female
e 151-4-16	"	20	"

No. e151-1 (♀) × c (♂)

e 151-1-1	Nov. 5	no infection	female
e 151-1-2	"	Dec. 2	"
e 151-1-3	"	no infection	male
e 151-1-4	6	Dec. 2	female
e 151-1-5	7	no infection	"
e 151-1-6	"	Jan. 2	"
e 151-1-7	9	Dec. 8	male
e 151-1-8	"	no infection	female
e 151-1-9	12	"	male
e 151-1-10	"	Dec. 23	female
e 151-1-11	13	30	male

Insect. No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e 151-1-12	15	no infection	female
e 151-1-13	"	Dec. 29	male
e 151-1-14	16	no infection	female
c 151-1-15	"	"	male
e 151-1-16	"	Dec. 17	female
e 151-1-17	18	no infection	"
e 151-1-18	"	Jan. 7	prematurely died
e 151-1-19	20	Dec. 17	male
e 151-1-20	"	13	female
e 151-1-21	"	"	"
e 151-1-22	"	19	male
e 151-1-23	21	10	"
e 151-1-24	"	20	female
e 151-1-25	22	Jan. 9	"
e 151-1-26	"	Dec. 14	"

No. e 151-4-4 (♀) × d (♂)

e 151-4-4-1	Jan. 16	no infection	male
e 151-4-4-2	"	"	prematurely died
c 151-4-4-3	"	"	"
e 151-4-4-4	"	"	female
e 151-4-4-5	"	"	male
e 151-4-4-6	"	"	female

No. e 279-4 (♀) × e (♂)

e 279-4-1	Jan. 31	Feb. 20	prematurely died
e 279-4-2	"	15	"
e 279-4-3	"	23	"
e 279-4-4	Feb. 1	20	male
e 279-4-5	"	17	female
e 279-4-6	"	no infection	prematurely died
e 279-4-7	"	"	"
e 279-4-8	2	Feb. 12	"
e 279-4-9	"	20	"
c 279-4-10	"	23	"

Insect. No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e 279-4-11	Feb. 2	Feb. 15	female
e 279-4-12	3	Mar. 1	prematurely died
e 279-4-13	"	Feb. 24	female
e 279-4-14	"	19	male
e 279-4-15	"	15	"
e 279-4-16	4	23	female
e 279-4-17	"	Mar. 1	prematurely died
e 279-4-18	"	Feb. 24	female
e 279-4-19	"	23	male
e 279-4-20	"	21	prematurely died
e 279-4-21	"	Mar. 7	"
e 279-4-22	5	Feb. 21	"
e 279-4-23	"	"	"
e 279-4-24	"	22	male
e 279-4-25	7	27	prematurely died
e 279-4-26	8	Mar. 10	female
e 279-4-27	"	1	prematurely died
e 279-4-28	"	3	female
e 279-4-29	10	16	prematurely died
e 279-4-30	"	1	female
e 279-4-31	12	5	prematurely died
e 279-4-32	"	3	male
e 279-4-33	14	1	prematurely died
e 279-4-34	"	2	"
e 279-4-35	"	Feb. 27	female

No. e 279-1 (♀) × f (♂)

e 279-1-1	Feb. 11	no infection	
e 279-1-2	12	"	
e 279-1-3	"	"	
e 279-1-4	"	"	
e 279-1-5	14	"	
e 279-1-6	"	"	

Insect. No.	Date of emergence from egg	Date of appearance of symptom	
e 279-1-7	Feb. 14	no infection	
e 279-1-8	"	"	
e 279-1-9	16	"	
e 279-1-10	18	"	
e 279-1-11	20	"	
e 279-1-12	"	"	
e 279-1-13	"	"	
e 279-1-14	"	"	
e 279-1-15	"	"	
e 279-1-16	"	"	
e 279-1-17	"	"	
e 279-1-18	"	"	
e 279-1-19	"	"	
e 279-1-20	"	"	
e 279-1-21	"	"	
e 279-1-22	"	"	
e 279-1-23	24	"	
e 279-1-24	"	"	
e 279-1-25	"	"	
e 279-1-26	"	"	
e 279-1-27	"	"	

No. e 151-1-16 (♀) × g (♂)

Insect. No	Date of emergence from egg	Length of feeding period	Result
e 151-1-16-1	Feb. 5	30 days	no infection
e 151-1-16-2	"	10	"
e 151-1-16-3	"	30	"
e 151-1-16-4	"	"	"
e 151-1-16-5	"	10	"
e 151-1-16-6	"	20	"
e 151-1-16-7	6	30	"
e 151-1-16-8	"	"	"
e 151-1-16-9	"	10	"
e 151-1-16-10	"	30	"
e 151-1-16-11	"	"	"
e 151-1-16-12	"	10	"
e 151-1-16-13	7	30	"
e 151-1-16-14	"	"	"

Insect. No.	Emergence from egg	Length of feeding period	Result
e 151-1-16-15	Feb. 7	30 days	no infection
e 151-1-16-16	8	5	"
e 151-1-16-17	"	30	"
e 151-1-16-18	"	20	"
e 151-1-16-19	"	"	"
e 151-1-16-20	"	"	"
e 151-1-16-21	"	"	"
e 151-1-16-22	"	"	"
e 151-1-16-23	9	10	"
e 151-1-16-24	"	"	"
e 151-1-16-25	10	30	"
e 151-1-16-26	"	10	"
e 151-1-16-27	"	"	"
e 151-1-16-28	11	30	"
e 151-1-16-29	"	"	"
e 151-1-16-30	"	5	"

No. e'51 (♀) × h (♂)

Insect No.	Emergence from egg	Appearance of symptom	
e' 51-1	June 30	July 15	
e' 51-2	"	11	
e' 51-3	"	12	
e' 51-4	"	11	
e' 51-5	July 8	25	
e' 51-6	9	22	
e' 51-7	10	25	
e' 51-8	11	24	

No. e 279-4-5-3-3 (♀) × i (♂)

e 279-4-5-3-3-1	July 1	July 15	
e 279-4-5-3-3-2	9	31	
e 279-4-5-3-3-3	"	"	
e 279-4-5-3-3-4	"	29	
e 279-4-5-3-3-5	"	Aug. 16	

No. e' 62 (♀) × j (♂)

e' 62-1	July 7	July 26	
e' 62-2	"	23	

No. e' 69 (♀) × k (♂)

Insect No.	Emergence from egg	Appearance of symptom	
e' 69-1	July 11	no infection	
e' 69-2	"	"	

No. e' 65 (♀) × l (♂)

e' 65-1	July 15	Aug. 2	
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No. e' 61 (♀) × m (♂)

e' 61-1	July 17	Aug. 10	
e' 61-2	"	no infection	
e' 61-3	"	"	
e' 61-4	19	"	
e' 61-5	"	Aug. 4	
e' 61-6	"	no infection	
e' 61-7	"	Aug. 6	
e' 61-8	"	no infection	
e' 61-9	"	Aug. 6	
e' 61-10	"	"	
e' 61-11	20	no infection	
e' 61-12	"	Aug. 5	

No. e' 131 (♀) × n (♂)

e' 131-1	Jan. 1 '34	Jan. 30	
e' 131-2	" 12	" 29	
e' 131-3	" 13	Feb. 7	
e' 131-4	" "	" 14	
e' 131-5	" 14	Jan. 29	

Infective female × infective male

No. e 279-4-5 (♀) × No. e 279-4-4 (♂)

Insect No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e 279-4-5-1	Mar. 26	Apr. 6	male
e 279-4-5-2	27	no infection	"
e 279-4-5-3	"	"	female
e 279-4-5-4	"	Apr. 6	male
e 279-4-5-5	"	10 (♀)	"
e 279-4-5-6	Apr. 2	10 (♀)	"
e 279-4-5-7	4	26	female

No. e' 34 (♀) × No. e' 29 (♂)

Insect No.	Date of emergence from egg	Date of appearance of symptom	Sex of insect (when matured)
e' 34-1	Apr. 30	May 16	female
e' 34-2	„	14	p. d. *
e' 34-3	May 3	22	male
e' 34-4	9	20	„
e' 34-5	22	June 14	female
e' 34-6	25	21	male
e' 34-7	June 8	23	„
e' 34-8	10	25	„
e' 34-9	„	24	„
e' 34-10	„	26	female
e' 34-11	„	no infection	„
e' 34-12	19	July 6	„
e' 34-13	„	7	male
e' 34-14	„	„	female

No. e' 39 (♀) × No. e' 36 (♂)

e' 39-1	May 1	May 13	male
e' 39-2	„	16	p. d.
e' 39-3	„	18	male
e' 39-4	„	15	„
e' 39-5	10	29	„
e' 39-6	21	June 14	female
e' 39-7	„	5	„
e' 39-8	„	10	male

No. e' 49 (♀) × No. e' 68 (♂)

e' 49-1	July 8	July 25	p. d.
e' 49-2	10	27	female
e' 49-3	„	24	p. d.
e' 49-4	„	„	„

No. e' 53 (♀) × No. e' 58 (♂)

e' 53-1	July 8	July 27	male
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* prematurely died

uninfective female \times infective male

a (♀) \times No. e 151-1-22 (♂)			
No. of insects tested	Date of emergence from egg	Length of feeding period	No. of infective leafhoppers
2	Jan. 25'33	31 days	0
2	26	30	0
5	27	29	0
b (♀) \times No. e 151-1-13 (♂)			
10	Feb. 7	33	0
1	8	32	0
2	9	31	0
5	10	30	0
1	12	28	0
c (♀) \times No. e 151-1-13 (♂)			
5	Feb. 14	31	0
d (♀) \times No. e' 47 (♂)			
5	Apr. 30	30	0
3	May. 4	"	0
1	5	"	0
e (♀) \times No. e 279-4-32 (♂)			
5	May 13	30	0
3	15	"	0
6	17	"	0
f (♀) \times No. e' 64 (♂)			
4	July 1	30	0
g (♀) \times No. e' 72 (♂)			
3	July 2	30	0
h (♀) \times No. e' 129 (♂)			
2	Dec. 29	52	0
2	" 30	51	0
7	" 31	50	0
1	Jan. 5'34	45	0
1	" 7	43	0

No. of insects tested	Date of emergence from egg	Length of feeding period	No. of infective leafhoppers
3	Jan. 10 '34	40	0
4	" 15	35	0
2	" 19	31	0
i (♀) × No. e' 129 (♂)			
3	Jan. 8 '34	42	0
1	" 13	37	0
5	" 15	35	0
3	" 21	29	0
j (♀) × No. e' 135 (♂)			
5	Dec. 21	60	0
2	" 28	53	0
3	Jan. 1 '34	49	0
k (♀) × No. e' 151 (♂)			
3	Dec. 27	54	0
4	" 28	53	0
3	" 29	52	0
1	" 31	50	0
3	Jan. 1 '34	49	0
l (♀) × No. e' 151 (♂)			
1	Jan. 3 '34	47	0
2	" 4	46	0
2	" 8	42	0
3	" 9	41	0
1	" 14	36	0
m (♀) × No. e' 167 (♂)			
3	Dec. 28	53	0
4	" 30	51	0
n (♀) × No. e' 167 (♂)			
1	Dec. 29	52	0
1	" 31	50	0
3	Jan. 1 '34	49	0
2	" 4	46	0

The data in the above table will be shown summarized as follows.

TABLE 14. Summarized results of the experiments with the progeny of infective leafhoppers

Parents		No. of progeny tested	No. of progeny infective
female	male		
infective female × infective male			
e 279-4-5	e 279-4-4	7	5
e' 34	e' 29	14	13
e' 39	e' 36	8	8
e' 49	e' 68	4	4
e' 53	e' 58	1	1
infective female × uninfected male			
e 279	a	13	13
e 151-4	b	16	15
e 151-1	c	26	17
e 151-4-4	d	6	0
e 279-4	e	35	33
e 279-1	f	27	0
e 151-1-16	g	30	0
e' 51	h	8	8
e 279-4-5-3-3	i	5	5
e' 62	j	2	2
e' 69	k	2	0
e' 65	l	1	1
e' 61	m	12	6
e' 131	n	5	5
uninfected female × infective male			
a	e 151-1-22	9	0
b	e 151-1-13	19	0
c	"	5	0
d	e' 47	9	0
e	e 279-4-32	14	0
f	e' 64	4	0
g	e' 72	3	0
h	e' 129	22	0
i	"	12	0
j	e' 135	10	0
k	e' 151	14	0
l	"	9	0
m	e' 167	7	0
n	"	7	0

As shown in above table, the majority of the offspring from the infective parents proved to be viruliferous and the progeny from the crosses between infective females and uninfected males were either viruliferous or free from the virus whereas those from the crosses between uninfected females and infective males were entirely free from virus. It appears that the eggs are not affected by the virus after they have been deposited in the leafsheath but probably at an early stage of their development in the ovary of the maternal insect body, because in no case has it been observed that infective progeny emerged out of the eggs from the uninfected females which had been laid in the leafsheaths of diseased plants. It is worthy of note that some individuals of the progeny from the infective females were viruliferous while the others from the same parents were apparently free from the virus. This seems to indicate that all the ova produced in an ovary are not always affected by the virus.

Although a number of virus diseases of plants have been demonstrated to be transmissible by the agency of certain insects, in no case has it been shown that the virus is transmitted through the eggs of an insect carrier. McCLINTOCK and SMITH (1918)¹²⁹⁾ reported that the virus of spinach blight (or spinach mosaic) was transmitted from infective aphids to their vivipariously produced progeny up to the 4th generation. This is the only evidence yet recorded that indicates the possible transmission of virus from insect vectors to their offspring. HOGGAN (1933),⁸⁰⁾ however, has not been able to confirm their conclusion. CARSONER and STAHL (1924)³⁴⁾ state that "the ability to produce the curly top of beet is not inherent" in the leafhopper, *Eutettix tenella* BAKER. Aster yellows is, according to KUNKEL (1926),¹¹¹⁾ not transmitted through the eggs of the insect carrier. STOREY (1928)²¹²⁾ also reports in his work on maize streak disease that "the eggs laid by infective leafhoppers (*Cicadulina mbila*) produced uninfected progeny."

Dwarf disease of rice plant, therefore, occupies a unique position among the virus diseases of plants for the reason of its transmission through the eggs of the insect carrier.

According to MURATA (1931),¹⁴⁹⁾ ONUKI and MURATA, formerly entomologists in the Imperial Agricultural Experiment Station, were probably the first to notice that the offspring of infective leafhoppers were capable of producing dwarf disease in healthy rice plants. In 1902? they found that certain leafhoppers emerged from the eggs which had been taken from Shiga Prefecture where this trouble was prevalent

at that time, produced infections in healthy rice plants. However, they gave no detailed account of it. The idea generally prevailing at that time was that the disease was due to the infestation of leafhoppers and accordingly these entomologists considered that the capability of producing dwarf disease was a characteristic specific to the leafhoppers native to the Prefecture of Shiga because all the leafhoppers of the same species taken in the vicinity of Tôkyo failed to produce infections in healthy rice plants. Later, however, they found that even uninfected leafhoppers from Tôkyo became infective after feeding upon dwarf diseased rice plants as stated before. MURATA (1915)¹⁴⁸⁾ briefly stated that the capability of producing dwarf disease of rice plant was transmitted from infective parents to their progeny through 3 or 4 generations but he presented no experimental procedures nor results which are considered to be sufficient ground for inducing such very significant conclusions.

At any rate these entomologists were evidently in ignorance of the fact that they were dealing with a virus disease in dwarf disease of rice plant and that the virus may be transmitted through the eggs of the leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL.

6. *Inoculation of rice plants with the macerated tissues of infective leafhoppers*

On April 5, 1933, about one hundred infective leafhoppers, a majority of them being nymphs in the 3rd or 4th instars, were ground in a mortar, adding a few drops of tap water. With the macerated tissues of leafhoppers thus prepared, 20 young rice plants were inoculated. Two inoculations were made on each plant, one on the leaf and the other at the base of stem. The inoculum was placed with a sterile scalpel on the desired spot and 30 pricks were made through the macerated tissues and body fluid of infective leafhoppers with a sterile needle, thus working the latter into the tissue. The inoculated plants were 2 weeks old plants of "Aikoku"-variety. All the inoculated plants remained healthy.

May 28, a similar experiment was carried out inoculating 30 young rice plants with the macerated tissues and body fluid of infective leafhoppers. The inoculum was prepared by grinding about 70 leafhoppers in a mortar adding a few drops of 0.85% saline solution. The procedure of inoculation was exactly the same as mentioned above. No symptoms of the disease appeared on any of the inoculated plants.

As a consequence of these inoculation experiments it appears that dwarf disease of rice plant is not transmitted to healthy plants by means of the macerated tissues and body fluid of infective leafhoppers.

7. Acquisition of infective power by the leafhopper

As stated before, the larger portion of the adult leafhoppers which had been reared upon dwarf diseased rice plants were entirely free from the virus. Accordingly the writer was naturally led to the supposition that most leafhoppers would not acquire infective power when the feeding period upon diseased plants was reduced to several days.

Non-viruliferous leafhoppers were allowed to feed upon dwarf diseased rice plants in insect proof cages for a definite length of time and transferred singly to a healthy rice seedling plant in a glass tube as described before, to test their infectivity. The effect was also tested of groups of these leafhoppers upon healthy rice plants in insect proof cages. Generally male leafhoppers were used instead of females in order to avoid egg deposition.

a) Results of experiments carried out in 1927

The non-viruliferous leafhoppers used in this series of experiments were collected on rice plants in the vicinity of Tottori where dwarf disease has never been found to occur.

TABLE 15. Acquisition of infective power by the leafhopper
(glass tube experiments in 1927)

Date	No. of leafhoppers tested	Sex	Length of feeding period		No. of infective insects
			on diseased plant	on healthy plant	
Oct. 12	3	♂	1 day	3 days	0
7	2	„	„	5	0
Sept. 30	3	„	„	7	0
10	3	„	„	14	0
1	6	„	7	1	0
4	6	„	„	„	0
1	6	„	„	3	0

b) Results of experiments carried out in 1928

The leafhoppers used in these experiments were the progeny of healthy leafhoppers which had been maintained on healthy rice plants since the previous season.

TABLE 16. Acquisition of infective power by the leafhopper
(glass tube experiments in 1928)

Date	No. of leafhoppers tested	Sex	Length of feeding period		No. of infective insects
			on diseased plant	on healthy plant	
July 15	6	♂	3 days	16 days	0
Aug. 31	7	„	10	13	0
Apr. 4	3	♀	30	3	0
Mar. 30	9	♂	„	7	0
„	9	♀	„	„	0
Sept. 10	5	♂	10	16	0
11	6	„	20	20	0
„	8	„	30	20	0

c) Results of experiments carried out in 1930-31

On the fourteenth of April, 80 or 90 leafhoppers were collected on *Astragalus sinicus* L. growing on the rice fields in the vicinity of Utsunomiya and two days later about half the number were introduced into insect proof cages enclosing healthy rice plants in the green house. The progeny of these insects were maintained on healthy rice plants to obtain non-viruliferous colonies of leafhoppers at the desired moment during the period of experiment. They were allowed to feed upon diseased plants for a definite length of time ranging from 1 to 70 days and then confined either individually on a single healthy rice plant in a glass tube or in a group on healthy plants in insect proof cages. As a rule, these insects were allowed to remain on the healthy plants until they died. The results of experiments will be shown below.

TABLE 17. Acquisition of infective power by the leafhopper
(glass tube experiments in 1930-31)

Date	No. of insects tested	Sex	Length of feeding period				No. of infective insects
			on diseased plant	Temp.	on healthy plant	Temp.	
May 5	2	♂	1 day	15-25°C	41-50 days	14-33°C	0
8	4	"	"	17-28	31-40	"	0
16	7	"	"	19-27	21-30	13-33	0
20	2	"	"	13-42	"	13-32	0
June 10	5	"	"	13-29	71-80	18-33	0
12	4	"	"	13-33	61-70	21-36	0
18	5	"	"	14-30	71-80	"	0
"	5	"	3	13-31	61-70	22-35	0
Apr. 28	1	"	7	16-32	11-20	16-28	0
June 11	5	"	"	14-32	61-70	22-36	0
Apr. 20	4	"	15	16-32	11-20	17-29	0
June 18	11	"	"	15-30	51-60	21-36	0
"	5	"	20	"	41-50	22-36	0
Aug. 1	4	"	"	22-36	21-30	17-32	0
"	2	"	"	"	41-50	16-36	0
"	1	"	"	"	51-60	"	0
Oct. 10	1	"	"	13-34	21-30	13-30	0
"	1	"	"	"	51-60	16-28	0
Sept. 30	1	"	30	13-36	1-10	15-33	0
"	3	"	"	"	11-20	13-31	0
"	4	"	"	"	21-30	"	0
"	1	"	"	"	41-50	14-30	0
"	1	"	"	"	51-60	16-28	0
Nov. 19	1	"	"	16-29	11-20	16-29	0
"	1	"	"	"	31-40	16-31	1 (Jan. 27)
"	2	"	"	"	41-50	17-31	0
"	1	"	"	"	51-60	"	0
"	1	n	"	"	21-30	16-30	0
"	1	"	"	"	31-40	16-31	0
"	1	"	"	"	41-50	17-31	0
"	2	"	"	"	51-60	"	0
Dec. 7	3	♂	"	"	11-20	"	0
"	1	"	"	"	21-30	17-32	0
"	2	"	"	"	31-40	18-33	0
"	1	"	"	"	41-50	"	0
Nov. 19	7	"	50	"	21-30	17-31	2 (Feb. 18, Feb. 2)
"	2	"	"	"	31-40	"	0
"	1	"	"	"	41-50	17-32	0
Dec. 16	1	"	"	17-30	1-10	17-34	0
"	3	"	"	"	11-20	18-35	1 (Feb. 20)
"	3	"	"	"	21-30	"	1 (Mar. 1)
"	2	"	"	"	31-40	"	0

n stands for nymph.

Date	No. of insects tested	Sex	Length of feeding period				No. of infective insects
			on diseased plant	Temp.	on healthy plant	Temp.	
Dec. 16	1	♂	50 days	17-30°C	51-60 days	18-36°C	0
17	2	"	"	"	1-10	"	0
"	2	"	"	"	11-20	"	0
"	1	"	"	"	21-30	"	0
Nov. 19	4	n	70	16-30	1-10	17-36	0
"	2	"	"	"	11-20	18-35	0
"	1	"	"	"	21-30	"	0
"	1	"	"	"	31-40	"	0
Dec. 16	1	♂	"	17-32	1-10	"	0
"	1	"	"	"	11-20	"	0
"	1	"	"	"	21-30	"	0
"	7	"	"	"	31-40	"	0

TABLE 18. Acquisition of infective power by the leafhopper
(cage experiments in 1930)

Date	No. of insects in each cage	No. of plants in each cage	Length of feeding period				No. of infective insects
			on diseased plant		on healthy plant		
Sept. 25	30 (♂)	2	10 days (15-36°C)		3-62 days (14-32°C)		1*
"	20 "	2	" "		" "		1*
12	10 "	2	15	"	11-34	(13-36)	0
"	6 "	2	"	"	11-13	"	0
25	6 "	2	"	(14-38)	5-7	(8-35)	0
2	5 "	2	20	(16-36)	1-5	(14-35)	0
29	15 "	3	30	(13-36)	2-19	(13-31)	0
"	6 "	2	"	"	2-15	"	0
Dec. 7	10 "	10	"	(16-29)	6-59	(18-34)	0

* On Dec. 6, sixty-two days after the transfer of the insects to healthy rice plants, only one leafhopper was found alive in each of the two cages labelled A and B. These insects were then confined singly in a glass tube enclosing a healthy rice plant. The insect from cage A was found dead on Dec. 10 and the other on Dec. 30. On Dec. 26, distinct symptoms of dwarf disease began to appear on both rice plants which had been subjected to the infestation of these insects in glass tubes. Prior to this, one of the plants in cage A began to show the symptoms of disease on Dec. 21 whereas the other plant in the same cage and the plants in cage B remained healthy until Jan. 16. The evidence seems to indicate that the above mentioned leafhopper in cage B remained uninfected during a period as long as 62 days at least after it had picked up the virus from dwarf diseased rice plants. In other words the incubation period of the virus in this insect body was more than 62 days under the writer's experimental conditions.

d) Results of experiments carried out in 1931

The leafhoppers used in these experiments were the progeny of non-viruliferous leafhoppers which had been cultured since the previous year in insect proof cages enclosing healthy rice plants.

TABLE 19. Acquisition of infective power by the leafhopper
(glass tube experiments in 1931)

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant	Temp.	on healthy plant	Temp.	
Aug. 20	1 (♂)	5 days	20-37°C.	11-20 days	16-35°C.	0
"	2 "	"	"	21-30	"	0
"	1 "	"	"	41-50	"	0
"	2 "	"	"	51-60	16-34	0
Sept. 5	5 "	"	16-35	21-30	14-35	0
"	2 "	"	"	31-40	15-34	0
"	2 "	"	"	41-50	"	0
"	3 "	"	"	51-60	16-34	0
"	3 "	"	"	61-70	17-34	0
"	3 (♀)	"	"	21-30	14-35	0
"	4 "	"	"	31-40	15-34	0
"	6 "	"	"	41-50	"	0
"	2 "	"	"	51-60	16-34	0
"	1 "	"	"	61-70	17-34	0
"	2 "	"	"	71-80	"	0
"	1 (♂)	10	16-36	21-30	14-33	0
"	2 "	"	"	31-40	15-33	0
"	2 "	"	"	61-70	17-33	0
"	1 (♀)	"	"	11-20	14-33	0
"	1 "	"	"	21-30	"	0
"	4 "	"	"	31-40	"	0
"	4 "	"	"	41-50	15-33	0
"	2 "	"	"	71-80	18-32	0
Aug. 24	3 (♂)	30	15-35	11-20	15-33	0
"	7 "	"	"	21-30	14-33	0
"	4 "	"	"	31-40	15-33	0
"	6 "	"	"	41-50	17-34	0
Sept. 17	2 "	"	14-32	1-10	19-34	0
"	1 "	"	"	11-20	18-35	0
"	6 "	"	"	21-30	19-34	0
"	4 "	"	"	31-40	20-34	0

TABLE 20. Acquisition of infective power by the leafhopper
(cage experiments in 1931)

Date	No. of insects in each cage	No. of plants in each cage	Length of feeding period		No. of infective insect
			on diseased plant	on healthy plant	
Aug. 14	20 (♂)	20	1 day (20-35°C)	11-39 days (17-36°C)	0
"	19 "	20	3 (21-39)	9-19 (17-35)	0
17	20 (n)	20	" (22-40)	11-49 (15-35)	0
18	35 "	20	5 (")	8-58 (")	0
31	17 (♂)	17	" (17-37)	3-27 (")	0
May 22	9 "	9	10 (16-31)	2-39 (")	0
June 2	10 "	10	" (10-31)	9-55 (")	0
Aug. 20	20 "	20	" (15-35)	9-51 (16-34)	0
Sept. 5	17 "	17	" (16-36)	10-40 (15-33)	0
Aug. 20	39 (♀)	20	30 (16-35)	2-45 (16-33)	0

e) Results of experiments carried out in 1932-33

On April 13, about two hundred nymphs of *Nephotettix apicalis* var. *cincticeps* were collected from *Astragalus sinicus* L. growing on the rice fields in the vicinity of Shiraoka near Ômiya. About 100 insects were confined in insect proof cages enclosing healthy rice plants. Almost all these leafhoppers proved free from virus and their progeny were kept in culture continuously on healthy rice plants in order to obtain non-viruliferous leaf hoppers at the desired moment.

TABLE 21. Acquisition of infective power by the leafhopper
(glass tube experiments in 1932-33)

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant	Temp.	on healthy plant	Temp.	
June 7	1 (♂)	5 days	14-30°C	1-10 days	15-33°C	0
"	3 "	"	"	11-20	16-33	0
"	9 (n)	"	"	"	"	0
"	1 (♂)	"	"	21-30	17-33	0
"	9 (n)	"	"	"	"	0
"	3 (♂)	"	"	31-40	18-33	0
"	3 (n)	"	"	"	"	0
"	1 (♂)	"	"	41-50	"	0

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant	Temp.	on healthy plant	Temp.	
June 7	6 (n)	5 days	14-30°C	41-50 days	18-33°C	0
"	1 (♂)	"	"	51-60	"	0
"	2 (n)	"	"	"	"	0
"	1 (♂)	"	"	"	"	0
June 23	1 (♀)	"	17-32	1-10	16-33	0
"	2 (♀)	"	"	11-20	17-33	0
"	2 (♀)	"	"	31-40	18-33	0
June 12	2 (♂)	10	16-33	1-10	17-33	0
"	1 (n)	"	"	"	"	0
"	1 (♂)	"	"	11-20	"	0
"	4 (n)	"	"	"	"	0
"	1 (♂)	"	"	21-30	17-32	0
"	4 (n)	"	"	"	"	0
"	5 (♂)	"	"	31-40	18-33	0
"	3 "	"	"	41-50	"	0
"	2 (n)	"	"	"	"	0
"	3 (♂)	"	"	51-60	18-34	0
"	1 (n)	"	"	61-70	18-33	0
"	1 "	"	"	71-80	"	0
"	1 "	"	"	81-90	"	0
June 23	1 (♂)	"	17-33	1-10	18-34	0
"	1 "	"	"	11-20	18-33	0
"	1 "	"	"	41-50	19-35	0
"	2 (♀)	"	"	"	"	0
"	1 "	"	"	51-60	"	0
Aug. 9	4 (♂)	"	21-38	1-10	18-36	1 (Aug. 31)
"	2 "	"	"	11-20	18-35	0
"	1 "	"	"	41-50	16-32	0
Aug. 19	8 "	"	18-37	1-10	19-32	0
"	9 "	"	"	11-20	18-32	1 (Sept. 14)
Sept. 14	1 "	"	11-32	1-10	13-32	0
"	3 "	"	"	11-20	13-33	0
"	5 "	"	"	21-30	13-34	0
"	3 "	"	"	31-40	14-34	0
"	5 "	"	"	41-50	15-34	0
"	1 "	"	"	51-60	18-33	0
Oct. 1	7 "	"	12-33	1-10	14-35	0

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant	Temp.	on healthy plant	Temp.	
Oct. 1	6 (♂)	10 days	12-33°C	11-20 days	18-34°C	0
"	3 "	"	"	21-30	19-34	0
"	2 "	"	"	31-40	"	0
"	4 "	"	"	41-50	"	0
Nov. 6	4 (n)	15	19-30	21-30	20-30	0
"	4 "	"	"	31-40	"	0
"	2 "	"	"	41-50	"	0
"	2 "	"	"	51-60	"	0
"	5 "	"	"	61-70	"	0
"	2 "	"	"	71-80	"	0
June 24	2 (♂)	20	17-32	1-10	18-34	0
"	2 (n)	"	"	"	"	0
"	1 (♂)	"	"	11-20	20-35	0
"	7 (n)	"	"	"	"	0
"	2 "	"	"	21-30	20-36	0
"	5 (♂)	"	"	31-40	"	0
"	3 (n)	"	"	"	"	0
"	1 (♂)	"	"	41-50	19-36	0
"	2 (n)	"	"	"	"	0
"	1 "	"	"	51-60	"	0
"	2 "	"	"	61-70	19-34	0
July 16	1 (♂)	"	20-36	1-10	20-35	0
"	1 "	"	"	11-20	21-35	0
Aug. 5	9 "	"	19-36	1-10	18-33	0
"	7 "	"	"	11-20	"	0
"	1 "	"	"	21-30	"	0
"	2 "	"	"	31-40	16-32	0
"	1 "	"	"	41-50	15-32	0
Aug. 25	3 "	"	18-33	11-20	11-32	0
"	1 "	"	"	21-30	13-32	0
"	1 "	"	"	31-40	12-33	0
July 3	9 "	30	"	11-20	20-36	0
"	2 "	"	"	21-30	19-36	0
"	3 "	"	"	31-40	19-35	0
Oct. 25	1 (♀)	"	19-32	11-20	18-31	0
"	1 (n)	"	"	"	"	0
"	1 (♂)	"	"	21-30	20-31	0

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant	Temp.	on healthy plant	Temp.	
Oct. 25	2 (n)	30 days	19-32°C	21-30 days	20-31°C	0
"	1 (♂)	"	"	31-40	20-30	0
"	2 (♀)	"	"	"	"	0
"	1 (n)	"	"	"	"	0
"	1 (♂)	"	"	41-50	"	0
Aug. 25	3 (♂)	50	20-31	1-10	20-31	1 (Jan. 5)
"	2 "	"	"	"	"	0
"	2 (n)	"	"	"	"	0
"	2 (♀)	"	"	11-20	20-30	0
"	1 "	"	"	"	"	1 (Jan. 10)
"	1 (n)	"	"	"	"	0
"	1 (♂)	"	"	31-40	"	0
Apr. 20'33	7 (♂)	30	20-38	11-20	13-34	0
"	7 "	"	"	21-30	"	0
"	2 "	"	"	31-40	13-35	0
"	1 "	"	"	41-50	15-36	0
May 20	5* "	5	20-36	11-20	12-34	0
"	1* "	"	"	21-30	13-34	0
"	7* "	"	"	31-40	13-35	1 (July 9)
"	5* "	"	"	41-50	15-36	0
"	1* "	"	"	51-60	16-36	1 (July 23)
"	1* "	"	"	61-70	17-36	0
"	5* "	10	18-35	11-20	12-34	0
"	11* "	"	"	21-30	13-35	0
"	1* "	"	"	31-40	14-35	0
"	9* "	"	"	41-50	15-36	3 (July 17, 18, 19)
"	3* "	"	"	61-70	18-37	0
June 6	4* "	50	18-38	11-20	22-37	0
"	2* "	"	"	21-30	22-38	1 (Aug. 17)

* non-viruliferous progeny originated from an infective female leafhopper.

f) Results of experiments carried out in 1933

The leafhoppers used in this series of experiments were obtained from virus free colonies of leafhoppers which had been bred in the following manner. On April 14, thirteen leafhoppers were collected on *Astragalus sinicus* L. growing on rice fields at Shimadachi-mura near Matsumoto

and likewise 133 individuals at Shiraoka near Ômiya and in the vicinity of Utsunomiya the next day. Almost all the leafhoppers were in their nymphal stage but molted to become adults within a week. On April 17 and 18 the leafhoppers were confined singly on young healthy rice plants in separate glass tubes and allowed to remain there for a month. All but one individual proved to be free from virus and accordingly were transferred into insect proof cages enclosing healthy rice plants. The progeny of these insects were maintained on healthy rice plants to secure virus free leafhoppers at the desired moment during the period of experiment. The non-viruliferous leafhoppers were allowed to feed upon diseased plants in an insect proof cage for a definite length of time ranging from 1 to 30 days and then confined singly to each healthy rice plant in a glass tube. They were transferred to new young plants at 10, 20, or 30 days' intervals. The results of experiments will be shown below.

TABLE 22. Acquisition of infective power by the leafhopper
(glass tube experiments in 1933)

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant		on healthy plant		
July 20	1 (n)	1 day	22-38°C	1-10 days 20-38°C		0
"	1 (♂)	"	"	11-20	21-38	0
"	1 (,,)	"	"	21-30	22-38	0
"	1 (,,)	"	"	31-40	21-37	0
"	1 (n)	"	"	"	"	0
"	5 (♂)	"	"	41-50	"	0
"	3 (,,)	"	"	51-60	"	0
"	1 (n)	"	"	"	"	0
"	1 (,,)	"	"	61-70	20-36	0
"	3 (♂)	"	"	71-80	19-36	0
"	2 (n)	"	"	"	"	0
31	6 (,,)	"	22-34	1-10	22-40	0
"	2 (♂)	"	"	11-20	22-39	0
"	9 (n)	"	"	"	"	0
"	7 (,,)	"	"	21-30	22-38	0
"	5 (♂)	"	"	"	"	0
"	2 (,,)	"	"	31-40	"	0
"	6 (n)	"	"	"	"	0
"	1 (♂)	"	"	41-50	20-37	0

Date	No. of insects tested	Length of feeding period		No. of infective insects	
		on diseased plant	on healthy plant		
July 31	5 (n)	1 day	22-34°C	41-50 days 20-37°C	0
"	2 (,,)	"	"	51-60 18-37	0
"	5 (,,)	"	"	61-70 "	0
July 20	2 (♂)	3 days	22-43	1-10 20-37	0
"	7 (n)	"	"	" "	0
"	1 (♂)	"	"	11-20 21-38	0
"	6 (n)	"	"	" "	0
"	4 (♂)	"	"	21-30 "	0
"	2 (,,)	"	"	" "	1 (Aug. 25)
"	1 (,,)	"	"	31-40 21-37	0
"	5 (,,)	"	"	41-50 21-38	0
"	1 (n)	"	"	51-60 20-37	0
"	1 (♂)	"	"	61-70 19-37	0
31	1 (,,)	"	22-40	1-10 23-40	0
"	5 (,,)	"	"	11-20 22-39	0
"	2 (,,)	"	"	31-40 22-38	0
"	3 (,,)	"	"	41-50 20-37	0
"	1 (,,)	"	"	51-60 "	0
"	9 (,,)	"	"	61-70 18-36	0
Aug. 8	1 (n)	"	22-37	1-10 22-39	0
"	3 (♂)	"	"	11-20 21-38	0
"	5 (n)	"	"	" "	0
"	1 (♂)	"	"	21-30 "	0
"	2 (n)	"	"	" "	0
"	1 (,,)	"	"	31-40 18-37	0
"	1 (,,)	"	"	41-50 18-36	0
"	2 (♂)	"	"	51-60 "	0
"	5 (n)	"	"	" "	0
June 16	14 (,,)	5	15-37	21-30 20-38	0
"	13 (,,)	"	"	31-40 "	0
"	5 (,,)	"	"	41-50 21-38	1 (Aug. 3)
"	13 (,,)	"	"	51-60 "	0
"	3 (,,)	"	"	61-70 "	0
"	1 (,,)	"	"	71-80 "	0
"	1 (,,)	"	"	101-110 20-37	0
July 6	19 (,,)	"	21-39	1-10 22-36	0
"	4 (,,)	"	"	11-20 21-36	0
"	1 (,,)	"	"	21-30 "	0

Date	No. of insects tested	Length of feeding period				No. of infective insects
		on diseased plant		on healthy plant		
July 6	9 (n)	5 days	21-39°C	31-40	22-38°C	0
"	3 (,,)	"	"	41-50	"	0
"	4 (,,)	"	"	51-60	"	0
June 16	11 (,,)	10	15-38	11-20	20-38	0
"	13 (,,)	"	"	21-30	21-38	0
"	23 (,,)	"	"	31-40	"	0
"	1 (,,)	"	"	41-50	"	0
"	2 (,,)	"	"	51-60	"	0
"	4 (,,)	20	17-38	1-10	"	1 (July 16)
"	10 (,,)	"	"	11-20	"	2 (July 21, 23)
"	10 (,,)	"	"	21-30	"	0
"	5 (,,)	"	"	31-40	"	0
"	6 (,,)	"	"	41-50	"	0
"	3 (,,)	"	"	51-60	"	0
"	14 (,,)	30	18-38	1-10	22-38	2 (July 24)
"	7 (,,)	"	"	11-20	21-37	1 (July 27)
"	4 (,,)	"	"	21-30	22-38	0
"	5 (,,)	"	"	31-40	"	0

The results of these experiments will be shown summarized in the following table.

TABLE 23. Acquisition of the infective power by the leafhopper

	Duration of feeding period on dwarf diseased rice plants (in days)										Total
	1	3	5	7	10	15	20	30	50	70	
No. of leafhoppers tested	130	122	246	24	313	56	118	230	43	18	1300
No. of infective leafhoppers	0	1	3	0	7	0	3	4	7	0	25
Percent. of infective leafhoppers	0	0.8	1.2	0	2.2	0	2.5	1.7	16.3	0	2.8

As shown above only 25 individuals of 1300 leafhoppers became infective after they had been allowed to feed upon diseased rice plants during a period from 3 to 50 days. The shortest period necessary for a non-viruliferous leafhopper to feed on diseased plants to pick up the virus was 3 days but a larger proportion of leafhoppers acquired the

virus after 10-50 days' feeding. The length of the incubation period of the virus in the insect carrier was not definitely determined, but the data available at present indicate that it was 10-40 days in certain cases where a temperature from 11° to 38°C. predominated as shown below, while it was prolonged to over two months in another case at a temperature ranging from 14° to 32°C. as mentioned before.

Insect No.	Feeding period on diseased plant	Feeding period on healthy plant			
	May 20-25 (20-36°C) (5 days)	May 25-June 4 (11-34°C) (10 days)	June 4-29 (13-36°C) (25 days)		
E' 2	—	—	+ (July 9)		
E' 7	—	—	+ (July 23)		
	May 20-30 (20-36°C) (10 days)	May 30-June 9 (12-35°C) (10 days)	June 9-July 9 (16-38°C) (30 days)	July 9-16 (21-38°C) (7 days)	
E' 35	—	—	—	+ (July 18)	
E' 36	—	—	—	+ (July 19)	
E' 47	—	—	—	+ (July 17)	
	June 16-21 (15-37°C) (5 days)	June 22-July 5 (17-36°C) (13 days)	July 5-Aug. 3 (21-38°C) (29 days)		
F 20	—	—	+ (Aug. 3)		
	July 20-23 (22-43°C) (3 days)	July 23-Aug. 12 (22-37°C) (20 days)	Aug. 12-22 (20-37°C) (10 days)		
F 245	—	—	+ (Aug. 25)		

At any rate it is evident, as a result of these experiments, that the virus is not transmitted mechanically from diseased plants to healthy ones by the agency of leafhoppers.

8. *Host range of dwarf disease of rice plant*

The symptoms of dwarf disease of rice plant closely resemble those of corn stripe disease in Cuba and likewise those of streak disease of maize and sugar cane in South Africa. The latter diseases are transmitted by the agency of the leafhoppers, *Peregrinus maidis* ASHM.²⁰⁵⁾ and *Cicadulina mbila* NAUDE,^{208, 209, 210)} respectively. According to STOREY (1925)^{208, 209)} various species of cultivated and wild grasses are subject to the attack of streak disease of maize. The writer, accordingly, attempted to transmit dwarf disease of rice plant to corn and several other grasses by the agency of the leafhopper, *Nephotettix apicalis* var.

cincticeps. All the inoculated plants were raised in 5-inch pots in insect proof cages and the young plants 2 to 10 days after germination were exposed to the infestation of leafhoppers in cages or glass tubes as described before. The cages employed were 30 cm. square and 60 cm. high. The potted young plants were placed in the cages and a definite number of leafhoppers were introduced therein. When the plants were unfavorable food plants for the leafhopper, certain individual leafhoppers would not migrate to the plants to feed upon them but remained on the wall of their cages eventually to be starved to death. In the later experiments, therefore, glass tubes were used instead of cages, allowing 3 or 4 plants to be grown in a glass tube closed at the upper end by a thin cotton cloth and a definite number of infective leafhoppers were transferred on to these plants. The leafhoppers were allowed to remain there until they all died or the symptom of the disease began to appear. The results of these experiments are shown in the following table.

TABLE 24. Results of attempted transmission of dwarf disease of rice plant to cultivated and wild grasses

Series 1. Cage experiment

Date	Name of plant	No. of insect per plant	Length of feeding period	No. of plants	
				inoculated	affected
July 9'28	<i>Zea mays</i> L. Unnamed variety	2	5-12 days	28	0
June 20'30	Longfellow & Sapporo Hachigyo	3	7-14	10	0
June 30'30	<i>Setaria italica</i> BEAUV. Yûshi	3	„	5	0
June 30'30	<i>Echinochloa Crus-</i> <i>galli</i> BEAUV. subsp. <i>colona</i> var. <i>edulis</i> HONDA Shiro-sangoku & Shiratama	3	20	10	5
July 31'30	Shiro-sangoku	2	21	10	9
June 30'30	<i>Panicum miliaceum</i> L. Wase	3	20	5	5
July 31'30	Wase	2	„	10	7
Jan. 23'33	Wase	1	22	10	6
Feb. 2'33	<i>Hordeum sativum</i> var. <i>vulgare</i> HACK. subvar. <i>coeleste</i> HACK. Marumi	1	5	10	0
Oct. 3'33	<i>Poa pratensis</i> L.	2	30	20	6

Series 2. Glass tube experiment (1933)

Date	Name of plant	No. of insect per plant	Length of feeding period	No. of plants	
				inoculated	affected
	<i>Zea mays</i> L.		days		
Mar. 1	Sapporo Hachigyo	1	7-10	5	0
„	Longfellow	„	„	6	0
3	Longfellow	„	„	5	0
20	Sapporo Hachigyo	3	5-7	10	0
21	Sapporo Hachigyo	10	„	5	0
	<i>Hordeum sativum</i> JESS. var. <i>vulgare</i> HACK. subvar. <i>coeleste</i> HACK.				
June 14	Marumi	2	7-13	10	0
	<i>Hordeum sativum</i> JESS. var. <i>hexastichon</i> HACK.				
Mar. 11	Chevalier	1	7-10	10	0
June 14	Chevalier	2	7-15	5	0
Mar. 11	Golden Melon	1	7-10	10	0
June 14	Golden Melon	2	7-15	10	0
	<i>Avena sativa</i> L.				
Feb. 28	White Belgium	1	7-10	10	0
May 14	White Belgium	1	7-15	10	1 (Jun. 8)
Mar. 13	Race Horse	1	„	10	1 (Apr.17)
May 14	Race Horse	1	5-7	10	2 (Jun.2,7)
	<i>Secale cereale</i> L.				
Mar. 16	Unnamed variety	1	7-10	10	3 (Apr.17)
May 14	Unnamed variety	1	5-8	10	0
	<i>Triticum vulgare</i> VILL.				
Mar. 16	Sapporo Harukomugi	1	7-10	10	2 (Apr.17)
May 25	Sapporo Harukomugi	3	7-20	3	0
June 7	Sapporo Harukomugi	2	„	10	0
Mar. 17	Martin Amber	1	7-10	7	0
May 25	Martin Amber	3	7-20	5	0
June 7	Martin Amber	2	„	10	0
	<i>Setaria italica</i> BEAUV.				
Mar. 16	Yûshi	1	5-10	10	0
Mar. 17	Hôten 20-go	1	5-15	10	0
	<i>Andropogon Sorghum</i> BROF. var. <i>vulgaris</i> HACK. subvar. <i>japonicus</i> HACK.				
Mar. 14	Jagan	1	7-10	10	0
May 15	Jagan	1	5-7	20	0

Date	Name of plant	No. of insect per plant	Length of feeding period	No. of plants	
				inoculated	affected
Apr. 5	<i>Andropogon Sorghum</i> BROT. var. <i>obovatus</i> HACK.	1	7-10	5	0
	Kuromi-baraho				
Mar. 11	<i>Panicum miliaceum</i> L.	1	10	7	7
	Wase				
Mar. 16	Manshû-shiro	1	„	2	2
Apr. 5	Manshû-shiro	1	12	4	4
Apr. 5	<i>Echinochloa Crus-galli</i> BEAUV. subsp. <i>colona</i> var. <i>edulis</i> HONDA	1	„	10	10
	Shiro-sangoku				
Apr. 25	<i>Alopecurus fulvus</i> L.	1	10-15	10	7
May 9	„	1	„	5	1

In the experiments performed in 1928 and 1930, it was not definitely known whether the leafhoppers used were all viruliferous, although they all had been reared on dwarf diseased rice plants. Consequently the results may be regarded as inconclusive when negative. All the leafhoppers used in the experiments carried out in 1930 were progeny from infective female leafhoppers which had been bred on diseased plants so it is highly probable that almost all of them were viruliferous.

As shown in Table 24, it has been conclusively proved that *Panicum miliaceum* L., *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA, *Poa pratensis* L., and *Alopecurus fulvus* L. are subject to attack by the virus causing dwarf disease of rice plant. The symptoms on these plants closely resemble those on rice plant, being characterized by streak and spotting on leaves and stunting of the whole plant. (Pl. I Fig. 2, Pl. II Fig. 5, 6) These plants proved to be favorite food plants for the leafhopper under consideration.

Rye, wheat and oats are slightly susceptible to the disease, infection having been produced in only a small portion of the inoculated plants. This may be partly due to the fact that these plants are uncongenial food plants for the leafhoppers which could not survive on them for longer than two or three weeks at most. The manifestations of the disease on these plants are generally less conspicuous but the severely affected plants are much stunted and the yellowish white spots and streaks on the leaves are rather striking when viewed by transmitted

light. (Pl I Fig. 3, Pl. II Fig. 7)

The plants which gave negative results are corn, Italian millet, barley and sorghum, although some of them may in the future prove to be more or less susceptible, if the work is carried out on a large scale. These plants were apparently unfavorable as food plants for the leafhoppers. They could survive on them for less than 15 days and perished sooner or later.

In "Results of experiments with insect pests. Report 8"¹⁹²⁾ published by the Shiga Agric. Exp. Station in 1908, it is briefly mentioned that dwarf disease of rice plant was successfully transferred to *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA but all the attempts to transmit it to *Panicum miliaceum* L. and *Setaria italica* BEAUV. failed. It appears that similar experiments were repeatedly conducted with invariably the same results during a period from 1904 to 1912 as briefly stated in that station's Special Bulletin No. 2¹⁹³⁾ and likewise in Annual Reports for the years 1908 to 1912.¹⁹⁴⁾

It is worthy of note that *Alopecurus fulvus* L. is susceptible to dwarf disease of rice plant. Since this is a biennial wild grass abundantly growing on the rice fields throughout Japan, it is possible that the virus causing dwarf disease of rice plant may be carried over the winter in this wild grass.

9. Overwintering of the virus of dwarf disease of rice plant

There are two possible ways in which the virus causing dwarf disease of rice plant survives over the winter: either in its insect carrier or in the perennial wild hosts.

On April 13, 1932, as stated before, about two hundred leafhoppers were collected on *Astragalus sinicus* L. growing on the rice fields in the vicinity of Shiraoka near Omiya and two days later about half the number were divided into several groups of 20 to 30 individuals and placed in as many insect proof cages enclosing young healthy rice plants. Twenty-nine days later, in one of the cages where about 30 leafhoppers were confined on as many healthy rice plants, one plant began to show distinct symptoms of dwarf disease. Thus it became evident that some individual leafhoppers, though probably few in number, which had hibernated on *Astragalus sinicus* and likewise on some wild grasses, carried the virus causing the disease.

In 1933 similar experiments were carried on by means of the glass tube method before described. The leafhoppers used were collected on

Astragalus sinicus growing on rice fields at Shimadachi-mura near Matsumoto on April 14 and likewise at Shiraoka near Ômiya and in the vicinity of Utsunomiya on the next day. Nearly all the leafhoppers were in their nymphal stage but molted to become adults within a week. On April 17 and 18 the leafhoppers were confined singly in a glass tube enclosing a young healthy rice plant and allowed to remain there for a month with the exception of a few leafhoppers which died in two or three weeks. The results of the experiment are shown in the following table.

TABLE 25. Infectivity of leafhoppers overwintered on *Astragalus sinicus* L. (1933)

Source of insects	Date of transfer of the insect to rice plant	No. of insects tested	Sex of insect	No. of infective insect
Shimadachi-mura	Apr. 17	13	male 4 female 9	0 1 (May 10)
Shiraoka	Apr. 17-18	83	male 31 female 57	0 0
Utsunomiya	Apr. 18	45	male 14 female 27	0 0
		146		1

As shown above only one individual of 146 leafhoppers tested proved to be infective. This leafhopper was collected on *Astragalus sinicus* growing on rice fields at Shimadachi-mura where dwarf disease of rice plant has been prevalent to some extent year after year.

In 1934 similar experiments were repeated again with the leafhoppers collected at three different localities, i.e., Utsunomiya, Kônosu and Matsumoto. These leafhoppers were supplied to the writer through the courtesy of Messrs. T. WATANABE, S. KATO and K. KURIBAYASHI. Although 300 to 400 leafhoppers were supplied only a third of them were found alive when they were received, a majority of them having died during their transport. The results of the experiments are shown in the following table.

TABLE 26. Infectivity of leafhoppers overwintered on *Astragalus sinicus* L. (1934)

Locality	Date of transfer of the insect to rice plant	No. of insects tested	Sex of insect	No. of infective insect
Utsunomiya	May 4	79	male 37 female 42	0 1 (May 20)
Kônosu	May 17	12	female	0
Matsumoto	May 28	14	female	1 (June 15)
		105		2

Of 105 leafhoppers tested 2 individuals, as shown above, proved to be infective.

The percentage of plants infected in these experiments is not high but the results are considered to be conclusive owing to the conditions under which the experiments were performed. The rice plants for experimental purposes were grown in insect proof cages and each leafhopper was allowed to feed upon one rice plant in a glass tube where there could not be any source of accidental infection at any time throughout the experiment.

The above results, however, do not necessarily indicate that the virus of dwarf disease of rice plant overwinters in the leafhopper, because the virus may survive over winter in its perennial wild hosts and subsequently may be picked up by the leafhoppers which feed upon these plants. The leafhoppers overwinter feeding on *Astragalus sinicus* L., *Alopecurus fulvus* L. and other grasses which are grown on the rice fields during the winter. They are especially abundant on *Astragalus sinicus* which is raised as the winter crop on the rice fields for ploughing in as a green manure. So far as *Astragalus sinicus* is not the symptomless carrier of the virus of dwarf disease of rice plant, it cannot be the source of virus since this plant has never been found to show any manifestation of virus disease. *Alopecurus fulvus* L. was experimentally proved to be subject to dwarf disease of rice plant as mentioned before. It is possible, therefore, that the virus may be carried over the winter in this wild grass. However, there are no records to indicate the occurrence of symptoms similar to those of dwarf disease of rice plant on wild grasses and the writer has never met with even one suspected case. Accordingly it is not definitely known to what extent this grass may

play a rôle as the source of virus causing the dwarf disease of rice plant. Rye, wheat and oats are also susceptible to dwarf disease of rice plant under certain conditions. It appears, however, that these plants have no possible relation to the overwintering of the virus of this disease, because they are not favorable food plants for the leafhopper which transmits the virus and moreover rye and oats are not raised at all in the regions where dwarf disease of rice plant is prevalent.

In short the conclusion seems undoubtedly that the virus causing dwarf disease of rice plant in all probability survives over winter in its insect carrier unless the fact is definitely established that the virus actually overwinters in *Alopecurus fulvus* L. and other biennial or perennial wild grasses under natural conditions or that *Astragalus sinicus* L. harbours the virus during the winter without showing any manifestation of the disease.

VII. Discussion and Conclusion

1. Diseases allied to dwarf disease of rice plant

The symptoms of dwarf disease of rice plant, its infectiousness, the invisibility of the causative agent under the microscope, the intracellular bodies in association with the affected tissues, and the mode of transmission, when taken altogether into consideration, lead one to the belief that this disease undoubtedly belongs to the virus disease group. Generally speaking, the virus diseases of plants can be classified based upon the most striking symptoms, into mosaic, curly leaf, leaf roll, yellows, streak and rosette. Dwarf disease of rice plant is a streak with a rosette appearance.

The virus diseases of plants can be divided into three groups, if the mode of transmission is taken into consideration. The diseases of the first group can be transmitted by the organic union only, such as budding or grafting, peach yellows,* peach rosette, little peach and phony disease of peach being representatives. The second group includes the diseases which are transmitted principally by the agency of insect carriers as represented by dwarf disease of rice plant, stripe disease of rice plant, streak of maize and sugar cane, corn stripe disease,

* Quite recently KUNKEL (1933)¹³³ successfully transmitted peach yellows to young seedling peach trees by the leafhopper, *Macropsis trimaculata*. Accordingly this should be transferred to the second group.

aster yellows and likewise by rosette of peanuts. In this group the transmission may be effected by grafting as already verified by KUNKEL (1926)¹¹¹⁾ and STOREY and BOTTOMLEY (1928)²¹⁵⁾ in aster yellows and rosette of peanuts respectively, but there is no evidence that these diseases are communicated through the juice from the affected plants. To the third group belong the diseases which are transmitted through the juice from the diseased plants, a majority of mosaic diseases and various other virus diseases being included in this group. The transmission by grafting and by the agency of insect vectors are usual in the third group. Certain diseases of this group have been shown to be seed borne while some others are transmitted through the soil.

Taking the symptoms and the mode of transmission into consideration, one comes to the conclusion that dwarf disease of rice plant closely resembles streak disease of maize and sugar cane which is transmitted by the agency of the leafhopper, *Cicadulina mbila*. Streak disease affects sugar cane, maize, teosinte, oats, *Panicum miliaceum* L. and some other grasses in South Africa (STOREY 1925).^{208, 209)} Stripe disease of rice plant which is characterized by yellowish stripes on the leaf and stunting of the whole plant and is transmitted by the leafhopper, *Delphacodes striatellus* FALL., more or less resembles dwarf disease of rice plant but in some respects it differs from the latter as pointed out by KURIBAYASHI (1931).¹¹⁵⁾ Corn stripe disease in Cuba is transmitted, according to STAHL (1927),²⁰⁵⁾ by the leafhopper, *Peregrinus maidis* ASHM. and somewhat resembles the rice disease under consideration in the manifestation of the disease. The corn mosaic investigated by KUNKEL (1921)¹⁰⁵⁾ in Hawaii is also transmitted by the just named leafhopper (KUNKEL 1922,¹⁰⁷⁾ 1927¹¹²⁾) and is more or less similar to the disease in question so far as the striping of the leaves and dwarfing of the plant are concerned. Whether the corn stripe disease in Cuba is identical with the corn mosaic in Hawaii is not fully known. Anyhow these diseases are most closely allied to dwarf disease of rice plant. Aster yellows and curly top of beets are also transmitted by the agency of specific leafhoppers but the manifestations of these diseases are somewhat different from those of dwarf disease of rice plant.

2. *The relationship between the leafhopper and dwarf disease of rice plant*

It appears that the causal relation of leafhoppers to dwarf disease of rice plant has been held under suspicion since the time of the earliest

record of this malady, as obviously shown in a brief article entitled "Dwarfed rice plant, its cause and control measures" which was published by the Department of Education in the Official Gazette of October 18, 1890 as mentioned before. In this article they stated: "Since leafhoppers are extremely abundant on rice plants in the regions under consideration (Prefecture of Kyôto), sucking plant juice badly to suppress the growth of rice plant, some bodies ascribe the dwarfed condition of rice plants to the infestation of leafhoppers."

According to ISHIKAWA (1928),⁸⁶⁾ HASHIMOTO (1894-95) was the first to prove experimentally the causal relation of leafhoppers to dwarf disease of rice plant. It is, however, uncertain with what species of leafhopper he worked. TAKATA (1895-96)²¹⁹⁾ asserts that a certain species of leafhopper named "mon-yokobai" is responsible for the disease. The leafhopper which he called "mon-yokobai" is apparently *Deltocephalus dorsalis* MOTSCH. as shown by his description accompanied by illustrations. This leafhopper, however, has no bearing upon the disease as reported by the Shiga Agricultural Experiment Station (1900).¹⁹²⁾

In "Results of experiments with insect pests. Report I"¹⁹²⁾ published by the Shiga Agricultural Experiment Station in 1899, it is stated that dwarf disease was caused by the leafhoppers, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL., *Deltocephalus dorsalis* MOTSCH., *Thamnotettix tobae* MATS., *Cicadula fasciifrons* STAL., and likewise by *Zygina limbata* MATS. (?) while none of the leafhoppers, *Delphacodes striatellus* FALL., *Tettigonia viridis* L. and *Nisia atrovenosa* LETH. was responsible for the incidence of the disease. It is evident that the disease was at that time entirely attributed to the infestation of leafhoppers. In Report II (1900) it is reported that only one species of all leafhoppers tested, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL. was capable of producing the true dwarf disease of rice plant, which was confirmed subsequently by long continued experiments. In Report VI issued in 1906, it is reported that dwarf disease was induced by the leafhoppers taken from Shiga Prefecture while those from Tôkyo failed to produce infections in healthy plants. This finding was confirmed in Reports VII and VIII published in 1908 showing that the leafhoppers taken from Okayama Prefecture produced the disease as well as those from Shiga Prefecture but those from Tôkyo and the northern part of Honshû (Main Island) were entirely uninfective. Furthermore it is stated that even the uninfective leafhoppers taken from Tôkyo or the

northern districts of Honshû acquired the ability to induce the disease when they were allowed to feed upon the dwarf diseased rice plants for about 5 days. Thus it became evident that the leafhopper itself does not cause the disease but transmits it acting as the vector. This was first shown by the experiments conducted in 1906 and confirmed during the subsequent years. Accordingly, it is in 1908 that the leafhopper, *Nephotettix apicalis* var. *cincticeps* was first authentically recorded to transmit dwarf disease of rice plant, although the causal relation of the leafhopper to the disease was proved 8 years before. Prior to this, according to BOKURA (1930)²¹⁾ and MURATA (1931),¹⁴⁰⁾ Onuki, formerly entomologist of the Imperial Agricultural Experiment Station, succeeded in proving in 1902 (?) that the leafhopper, *Nephotettix apicalis* var. *cincticeps* acts as the carrier to transmit dwarf disease of rice plant. Onuki, however, published no account of it. ANDÔ (1910)⁷⁾ briefly reported the results of transmission experiments with the leafhopper carried out in the Imp. Agr. Exp. Sta. In 1905, according to him, the leafhoppers from Tôkyo were caged on dwarf diseased rice plants taken from Shiga Prefecture and the insects of the 2nd or 3rd generation proved to be capable of producing infections. He further stated that uninfected leafhoppers from Tokyô became infective when allowed to feed on diseased rice plants for 15 days (experiments carried out in 1906 and subsequent years).

As far as the writer is aware, curly top of beets is the first virus disease of plant in the foreign literature, shown to be transmitted by an insect. In 1906¹¹⁾ BALL reported a great outbreak of the beet leafhopper, *Eutettix tenella* BAK. which seriously damaged the fields of Utah in the previous year and stated that "its punctures seem to cause a sort of thickening of the veins of the leaf and an unhealthy condition called 'curly-leaf' or 'blight'." In two succeeding papers (1907,¹²⁾ 1909¹³⁾) BALL emphasized the causal relation between the leafhopper attack and the curly leaf and stated that the curly leaf condition apparently spread from leaf to leaf until finally the whole plant was affected, even though the leafhopper might have disappeared in the meantime. He (1909)¹³⁾ briefly referred to the experimental production of "curly-leaf" in cages arranged jointly by himself and E. G. TITUS. BALL's findings with regard to the causal relation of the beet leafhopper to curly top were confirmed by SHAW (1910)¹⁸⁹⁾ and later by SMITH and BONCQUET (1915).²⁰²⁾ In 1915²²⁾ BONCQUET and HARTUNG reported that the beet leafhoppers taken from wild plants had no power to

produce curly top but they became viruliferous after feeding for a few days upon affected plants. This fact was corroborated by SMITH and BONCQUET²⁰²⁾ in the same year.

A few years earlier than this, ALLARD (1912,¹⁾ 1914²⁾) asserted that the occurrence of mosaic disease of tobacco was associated with aphid infestation. According to him (1914,²⁾ 1917⁶⁾) *Rhopalosiphum* (= *Myzus*) *persicae* SULZ.* may become an active carrier of infection in the green house, after it has been feeding upon mosaic diseased tobacco plants, while *Macrosiphum tabaci* PERG. appears to be an efficient carrier of the virus under field conditions.

Following these pioneer works many investigators have reported on various cases of insect transmission of virus diseases of plants. A relatively small number of them have been shown to be transmitted by leafhoppers; e.g., it has been reported that *Peregrinus maidis* ASHM. acts as the vector of the mosaic disease of corn in Hawaii (KUNKEL 1922)¹⁰⁷⁾ and the corn stripe disease in Cuba (STAHL 1927),²⁰⁵⁾ that *Typhlocyba ulmi* L. spreads leafroll of potato (MURPHY 1923),¹⁵¹⁾ that *Cicadulina mbila* (NAUDE) and *C. zea* transmit the streak disease of sugar cane and corn (STOREY 1925,^{208,209)} 1931²¹³⁾), that *Cicadula sexnotata* FALL. disseminates aster yellows (KUNKEL 1924,¹⁰⁸⁾ 1926¹¹¹⁾), that *Euscelis striatulus* acts as the carrier of the virus of cranberry false blossom disease (DOBROSKY 1929,⁴⁷⁾ 1931,⁴⁸⁾ BECKWIRTH & HULTON 1929¹⁶⁾) and that *Delphacodes striatellus* FALL. transmits the stripe disease of rice plant (KURIBAYASHI 1931).¹¹⁵⁾ Quite recently it has been shown that peach yellows is also transmitted by the leafhopper, *Macropsis trimaculata* (FITCH). (KUNKEL 1933)¹¹³⁾.

The relation of leafhopper to virus has been closely investigated in curly top of beet, streak disease of corn and aster yellows.

In curly top of beet SMITH and BONCQUET (1915)²⁰²⁾ demonstrated that non-viruliferous leafhoppers became highly infective after feeding upon diseased sugar beets for a period which at most needed not to exceed 3 hours. But the insects were unable to transmit the disease immediately after feeding on diseased plants and a period of at least 24 hours, but certainly no more than 48 hours, had to elapse before these insects could produce the disease. They came, therefore, to the conclusion that "the beet leafhopper does not mechanically transfer

* According to HOGGAN (1929),⁷⁹⁾ this species of aphid is not capable of transmitting tobacco mosaic.

the virus from diseased to healthy plants but that some development or change takes place in the virus within the body of the insect during the first few hours after it feeds upon the diseased plant." They were the first to suppose the possible existence of the incubation period of virus within the body of the insect carrier. According to them, as short a period as 5 minutes is sufficient for a single leafhopper to feed upon a healthy sugar beet in order to produce infection. SEVERIN (1921)¹⁸⁶⁾ states that the beet leafhopper can become infective by feeding only 1 or 2 minutes on a diseased beet and the minimum incubation period of the virus in the leafhopper requires 4 hours at the temperatures: maximum 103° F.; minimum 94° F. and mean 100° F. He later (1924)¹⁸⁷⁾ reports that infective beet leafhoppers retained their infectivity during all of the nymphal stages, after each molt, and during the entire adult life and (1931)¹⁸⁸⁾ that the shortest period necessary for a single leafhopper to transmit the disease was 7 hours. STAHL and CARNSER (1918)²⁰⁰⁾ report that newly emerged beet leafhoppers lifted from the plant before feeding were non-viruliferous but became carriers of the virus when fed on diseased beets. In their subsequent papers (1923, 1924)^{207, 34)} they state that the shortest period required for non-viruliferous leafhoppers to feed upon diseased beets in order to obtain the virus was 10 minutes, that the insects became infective within a period of 15 hours after they had fed upon diseased beets and that the minimum period necessary for a single leafhopper to transmit the disease was 20 minutes for sugar beets and 10 minutes for *Stellaria media*. They further state that "a viruliferous leafhopper may not produce infection each time it feeds on a healthy plant, even though the periods of feeding be 24 or 48 hours or even longer" and that the ability to produce the disease is not inherent in the leafhopper. They pointed out the apparent inability of any insects other than *Eutettix tenella* to transmit the disease.

The virus of aster yellows has been shown to require a much longer incubation period in its insect carrier, *Cicadula sexnotata* FALL. According to KUNKEL (1926),¹¹¹⁾ a period of at least 10 days, generally 12 to 17 days must elapse before the leafhoppers become infective, after they have fed on yellowed aster plants. A feeding period of 2 hours upon the yellowed plant is not long enough to render the leafhoppers viruliferous but an exposure of one day is sufficient to enable them to become virus-bearing. Considerable variation is shown in the certainty with which each infective leafhopper transmits the disease. In this

respect KUNKEL states: "Certain individuals transmitted the yellows to every plant on which they were confined for one day and some others transmitted it over a long period of time and with great certainty, whereas another group of insects did it with considerable irregularity failing to produce infection in many of the plants on which they were confined, and still another group transmitted yellows to a few plants and then apparently lost the virus." He further states that viruliferous leafhoppers retained the virus for as long a period as 75 days when they were confined on the rye plants that are immune to aster yellows, but that the virus is not transmitted through the eggs of the insect carrier. He attempted to transmit aster yellows by different species of leafhoppers and other insects and by the red mites but only negative results were obtained.

In streak disease of maize, STOREY (1928)²¹²⁾ has shown that the leafhopper, *Cicadulina mbila* (NAUDE) may acquire the virus after feeding for one hour on a diseased plant but in lower proportion than is the case when the feeding period is prolonged to several days. But "even when the feeding period on a diseased plant extends through the whole course of development of the insect from the first instar to the adult stage, a proportion of the hoppers remain uninfected." These insects are incapable of acting as vectors of the virus. The character of 'activity' in the transmission of maize streak has been found (1931)²¹⁴⁾ to behave in inheritance as dominant to that of 'inactivity' and to be sex-linked. According to STOREY, the cross, active male \times inactive female gave F₁ progeny of inactive males and inactive females. In the F₂ generation, active and inactive insects developed in approximately equal numbers in each sex. The reciprocal cross, inactive male \times active female, yielded an entirely active F₁ offspring, with segregation in the F₂ generation into active and inactive males in about equal numbers and only active females. The incubation period of the virus in the insect is variable but shortest at the higher temperatures. The minimum period observed was 6-12 hours at 30°C. Infective power is apparently not lost during the process of molting and it is retained during periods of feeding as long as 2 to 3 months upon immune plants. The virus, however, does not pass from parent to offspring through the eggs.

In curly top of beet and maize streak, it appears that the relation of the virus to its insect vector is not absolutely specific, because curly top has been shown to be transmissible by another leafhopper, *Agallia sticticollis* STAL. in Argentine (FAWCETT 1927)⁶²⁾ and maize streak is

transmitted by *Cicadulina zea* as well as by *C. mbila* (STOREY 1931).²¹³⁾

In dwarf disease of rice plant, the leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL. seems to be specific to the virus which is not transmitted by any other leafhoppers.

As a result of the writer's experiments evidence was obtained to indicate that all individuals of the leafhopper, *Nephotettix apicalis* var. *cincticeps* are not capable of acting as vectors of the virus of dwarf disease. Certain individuals of this leafhopper fail to transmit the disease even when they have been hatched and reared on diseased plants, although no morphological difference can be found between these leafhoppers and those which readily transmit the disease. A majority of the progeny from an infective female, which have been reared on diseased plants are capable of producing infections in healthy rice plants. Sometimes a period of 5 minutes was sufficient for a single leafhopper to feed upon a healthy plant to transmit the disease and a feeding period of 30 minutes was long enough to enable most of the infective leafhoppers tested to produce infections in healthy plants. Most of the leafhoppers retained their infective power as long as they lived and certain individuals produced infection in every healthy plant on which they were confined for 24 hours on consecutive days. In comparatively few instances the infectivity of a leafhopper was apparently reduced or lost even under favorable conditions. A viruliferous leafhopper may not produce infection in every plant on which it feeds, even though the periods of feeding may be 24 hours or longer. It seems, therefore, that certain viruliferous leafhoppers sometimes fail to produce infections in healthy plants, probably owing to the temporary exhaustion of the virus in the salivary glands or the anterior portion of the alimentary canal. Then the infections subsequently produced by the same individual must be attributed to the virus which had either multiplied in the salivary glands etc. or migrated from some other portion of the body to the salivary glands and the anterior part of the alimentary tract. This is an additional evidence to indicate that the leafhopper does not transfer the virus mechanically from a diseased to a healthy plant. The virus may be transmitted through the eggs of infective leafhoppers. The progeny from infective female leafhoppers are either viruliferous or free from the virus whereas those from the crosses between uninfected females and infective males are entirely non-viruliferous. It appears that all the ova in the ovaries of an infective leafhopper are not always affected by the virus. A period from one to 14

days must elapse before most of the infective nymphs become capable of producing infections in healthy plants although certain individuals may be infective immediately after emergence from eggs. This admits several interpretations: either (1) the newly emerged tiny nymphs may be able to transfer a dose of the virus insufficient to produce infection in a healthy plant, or (2) some developmental changes or multiplication of the virus may occur within the insect body before it is fully infective, or (3) the virus may migrate from the other parts of the insect body to the salivary glands and the anterior portion of the alimentary canal. Most of these infective nymphs retain their infectivity during all the nymphal stages and through the entire adult life, without access of the virus. Only a small proportion of non-viruliferous leafhoppers, both nymphs and adults, became infective after feeding on dwarf diseased rice plants under the writer's experimental conditions. The minimum period observed to be necessary for a non-viruliferous leafhopper to feed upon a diseased rice plant to pick up the virus was 3 days. But the leafhoppers acquired the virus more readily when they had been allowed to feed upon diseased plants for 10 to 50 days. Certain individuals of the leafhopper, however, remained uninfected even when the feeding period on diseased plants extended to 50 or 70 days. In the case of dwarf disease of rice plant, there also occurs an incubation period of the virus in the insect carrier, which could not be definitely determined but was apparently more than 10-40 days in certain cases where a temperature from 13° to 36°C. predominated while it was prolonged to more than two months in another case at a temperature ranging from 14° to 32°C. This furnishes further evidence to show that the virus is not mechanically transferred from diseased to healthy plants by the agency of leafhoppers but that some development or multiplication of the virus takes place within the body of the leafhopper before it becomes fully infective.

In short the conclusion seems to be justified that a relatively small proportion of the leafhoppers acquire the virus after feeding on dwarf diseased rice plants but the individuals which have once become infective generally retain the virus through their entire life and certain infective females transmit the virus to their progeny through the eggs. Thus it is evident that the leafhopper does not transfer the virus mechanically from diseased to healthy plants but that some development or multiplication of the virus takes place within the body of the leafhopper.

3. *Etiology of dwarf disease of rice plant*

Various theories have been advanced in order to account for the cause of dwarf disease of rice plant. KERNER et al. (1890),⁹²⁾ as mentioned before, ascribed this malady to the products of the reduction process which occurs in the soil. On account of the deficiency of oxygen in the soil, they assert, the sulphates contained in the soil, manure and likewise in the irrigation water are brought to reduction, producing iron sulphide which suppresses the growth of rice plants.

This hypothesis was disproved by TAKATA (1895-96),²¹⁹⁾ who looked upon this trouble as caused by the infestation of leafhoppers. As a result of more extensive studies performed by the Shiga Agricultural Experiment Station (1899-1905) the causal relation of the leafhopper, *Nephotettix apicalis* var. *cincticeps* to dwarf disease of rice plant was definitely established. A few years later, however, it was found that the disease is not due to the infestation of leafhoppers itself but that it is caused by certain infective principles which are transmitted by the agency of leafhoppers. The nature of the causative agent still remained unknown.

In the Annual Report of the Imperial Agricultural Experiment Station for the years 1913-1915,⁸⁵⁾ issued in 1917, it is briefly stated that the occurrence of certain protozoa was noticed in the tissues of diseased rice plant as well as in the blood of the leafhoppers. However, no detailed account has been given of these protozoa or of their relation to dwarf disease of rice plant.

DAIKUHARA (1904)³⁹⁾ was the first to recognize this trouble as a disease allied to serch of sugar cane, mosaic disease of tobacco and likewise to the stigmonose of carnations. On this point YONEMARU and Aso's view (1909)²³⁴⁾ was quite in agreement with DAIKUHARA's opinion. In recent years KASAI (1924)⁹⁹⁾ looked upon this disease as "allied to the virus diseases" and HINO (1927)⁷⁶⁾ also emphasized that this disease belongs to the mosaic disease group.

The symptoms of the disease, its infectiousness, the invisibility of the causative agent under the microscope, the intracellular bodies associated with the affected tissues, and the mode of transmission point to the fact that this disease is undoubtedly caused by a certain virus. Then what is the virus?

The properties of virus have been demonstrated in mosaic diseases of plants, particularly in those of tobacco, cucumber, potato, bean and

others. Undoubtedly, however, the best known and most thoroughly investigated virus is that of the mosaic disease of tobacco, and numerous hypotheses have been advanced as to its nature. As has been discussed by the writer (1929)⁶⁶⁾ before, some of these hypotheses have been nearly discarded, whereas three of them are considered to be more plausible, namely, the chlamydozoa hypothesis of PALM (1922),¹⁶¹⁾ HUNGER'S (1905)⁸³⁾ phytotoxin and allied views pointing to a certain toxin or enzyme-like substance, and the ultramicroorganism theory suggested by ALLARD (1916)⁵⁾ and advocated by D'HERELLE (1924).⁴⁰⁾ Although it cannot be denied that the virus group in all probability is not a homogeneous one, these three hypotheses may be adopted in considering the nature of the virus of dwarf disease of rice plant.

PALM (1922)¹⁶¹⁾ found certain foreign corpuscles and extraordinarily small granules in the cells of the mosaic diseased tissue of tobacco plant. He considered that these minute granules "agree in every respect with" the chlamydozoa of PROWAZEK (1907)¹⁶⁷⁾ or organisms occupying an intermediate systematic position between protozoa and bacteria, that the foreign corpuscles are homologous with the GUARNIERI bodies associated with variola, and further that these chlamydozoa represent the causal agency of mosaic disease of tobacco. However, no convincing evidence has been obtained to prove that these minute granules actually do represent the organisms which cause the disease. GOLDSTEIN (1924)⁷²⁾ suggested that these foreign corpuscles may represent a living organism or perhaps an aggregate of living organisms. But it appears that certain pathologists incline to consider them as the reaction products of the affected cells. (IWANOWSKI 1903,⁸⁹⁾ PALM 1922,¹⁶¹⁾ F. F. SMITH 1926,¹⁹⁶⁾ HOGGAN 1927⁷⁸⁾) As stated before, the occurrence of similar intracellular bodies has been demonstrated in several other virus diseases of plants and certain investigators looked upon them as the causal organisms. The general belief among plant pathologists, however, seems to be in favor of the view that these bodies are the reaction products of the host cells rather than the causative agents of the disease. The intracellular bodies which are found in the chlorotic tissues of leaves and likewise in the root and crown of dwarf diseased rice plant closely resemble those foreign corpuscles occurring in association with the mosaic disease of tobacco. These bodies cannot be considered as protozoans or other microorganisms because no indication of protoplasmic differentiation is shown in these bodies. Minute granules have been revealed scattering within these intracellular bodies. It was not, how-

ever, definitely known whether they represent the so-called chlamydozoa. They are not in all probability chondriosomes because no similar granules are found in the cytoplasm of the host cells and on the other hand these granules could not be demonstrated by means of the technique ordinarily recommended for the study of chondriosomes. The intracellular bodies are present also in the chlorotic tissues of leaves of wheat, rye, *Panicum miliaceum* L., and *Echinochloa Crus-galli* BEAUV. subsp. *colona* var. *edulis* HONDA and likewise in *Alopecurus fulvus* L. affected by the virus of dwarf disease of rice plant. In shape, dimension and structures, these intracellular bodies closely resemble those occurring in the affected tissues of rice plant. Accordingly it seemed desirable to determine whether similar inclusion bodies also occur in the viruliferous leafhopper. After a careful examination of the salivary glands, the alimentary tract, and the egg follicles in the ovarian tubules, and likewise in the mycetome and other organs of the viruliferous leafhoppers, the writer was unable to discover any inclusion bodies or any visual evidence of the presence of the virus.

Except the intracellular bodies, no microorganisms or like bodies have been found in constant association with dwarf disease of rice plant in spite of a careful examination of serial sections of all parts of affected rice plants. As a consequence it is considered that the causative agent may be of an ultramicroscopic dimension, either the so-called ultramicroorganism or some toxin-like substance.

As has been stated before, a period of 5 minutes was sufficient for a single leafhopper to feed upon a healthy plant in order to transmit the disease. The average weight of an infective male or female leafhopper being about 2.3 or 3.7 milligrams respectively, the amount of virus which a single leafhopper injects into the plant tissue during a period of 5 minutes is considered to be infinitesimally small. This extremely small quantity of the virus was capable of producing infection in a healthy rice plant whose average weight varied from 100 to 250 milligrams and developed a typical case of dwarf disease. The virus eventually becomes distributed throughout all parts of an affected plant as is obviously shown by the fact that the virus is present in the leaf tissues and is picked up by the leafhopper which feeds upon the leaf and that it exists also in the root since all the new growths show more conspicuous symptoms of the disease when the aerial parts of affected plants are cut back. In the green house the virus is persistent for years in the roots of affected plants and numerous diminutive tillers develop

invariably producing more and more striking symptoms of the disease when the plants are repeatedly cut back. The virus undoubtedly resides in the newly developed leaves, since a healthy leafhopper is capable of taking up the virus after feeding upon these leaves. Accordingly it is evident that the extremely small amount of virus which has been inoculated into a rice plant by the agency of leafhopper persists in the root forever and it is partly distributed throughout new growths of the plant. Such a condition is perpetuated for years in the green house and subsequently new growths invariably develop more and more conspicuous symptoms of the disease. This can be hardly explained, unless we assume that the virus multiplies in the plant body.

However, it is not certain whether the virus reproduces itself or the host cell affected with the virus produces the virus in turn,—that is some abnormal metabolic product of the affected cell may function as the virus and stimulate it to develop the same product. In this connection, it is interesting to refer to BAUR's view (1904)¹⁴⁾ as to the nature of viruses which cause the so-called infectious chloroses of plants or the variegation which is transmissible by means of grafting with infected twigs or even with single diseased leaves. He considered that the virus of the infectious chlorosis must multiply within the affected plant because, "durch Transplantation eines einzigen kranken Blattes können wir eine andere Pflanze infizieren, diese produziert eine unbegrenzte Menge kranker Blätter, mit jedem einzelnen von diesen Blättern können wir wieder eine andere Pflanze infizieren usf. ad infinitum." The virus, he argued, can not be an organism, since the disease can be transmitted by grafting only but not through the expressed juice of diseased leaves, nor by intimate contact with affected plants and otherwise, and since "ein parasitärer Organismus, der eine derartig geringe Fähigkeit hat, von einer Wirtspflanze in eine andere zu gelangen, ist überhaupt nicht existenzfähig." He suggested that the virus need not "aktiv wachsen" as an ordinary organism does, but that possibly some metabolic product of an affected plant may function as the virus and exert a formative stimulus on the embryonic leaf primordium, so that variegated leaves are produced instead of normal green foliage. Then these leaves may produce in turn the same abnormal metabolic product, which again exerts a stimulating action on the primordium of leaf. He looked upon certain infectious diseases, notably the mosaic disease of tobacco, as caused by such "ein stoffliches Virus."

HUNGER (1905),⁸³⁾ evidently in ignorance of BAUR's paper, ad-

vanced a similar hypothesis as to the nature of the virus of mosaic disease of tobacco. He considered that that virus is a toxin or phytotoxin which is primarily produced as some metabolic product of plant cells and stimulates the normal cells to produce the same toxin.

BAUR (1906)¹⁵⁾ later inclined to consider it more probable that the virus is a highly organized chemical substance which is produced as some metabolic product of the diseased plant and endowed with the ability of growth, splitting other compounds to produce the same substance or synthesizing it. This substance acts on certain groups of molecules in the embryonic cells of leaves in the same way as the toxin does which was postulated by EHRLICH in his widely known side-chain theory. The newly produced toxin migrates throughout the plant tissues to reach the cell wherein it finds the free side chain with which to unite, thus coming to enter the embryonic cells of leaves.

BEIJERINCK (1898)¹⁷⁾ considered that the virus of tobacco mosaic is a contagium *vivum* fluidum, because it was possible, by starting with an extremely small quantity of the virus, to infect serially an endless number of plants and accordingly the virus must multiply within the living plant. ALLARD (1916)⁵⁾ also considered that the virus of tobacco mosaic was capable of increasing indefinitely within affected plants, and therefore "there was every reason to believe that it is an ultramicroscopic parasite of some kind."

BAUR and HUNGER, as stated before, also admitted that the virus multiplies within the affected plant; nevertheless their views are opposite to the considerations of BEIJERINCK and ALLARD that the virus is after all, of a living nature.

Now leaving as they are, the conflicting views whether the viruses of tobacco mosaic and infectious chloroses of plants are animate or inanimate, it should be desirable to return to the subject in question.

As stated before, some individuals of the progeny from the viruliferous female leafhopper carry the virus by nature and retain it during all the nymphal stages and through the entire adult life or for a period ranging from 53 to 97 days, without access of the virus. Furthermore, certain leafhoppers produced infections in almost every healthy plant on which they were singly confined for 24 hours when each leafhopper was transferred daily to a new healthy rice plant. The average weight of a nymph just emerged from the egg being 0.06 milligrams, the amount of virus contained in the insect body must be extremely small as compared with the inoculated plant which ranged from 150 to 300 milli-

grams by weight. Without assuming that the virus multiplies within the insect body, it can be hardly explained how such an insect is capable of producing infections in 35 to 71 healthy plants on which it was confined for 24 hours on consecutive days.

A period of more than 10 days must elapse before certain viruliferous nymphs become capable of producing infections in healthy plants whereas some other individuals may be infective immediately or a few days after their emergence from eggs. In such a case it is most reasonable to consider that the virus may multiply within the body of insect before it is fully infective as has been already pointed out.

Non-viruliferous leafhoppers are not capable of transmitting the disease immediately after they have fed upon diseased rice plants but a period of 10 to 40 days must elapse before these leafhoppers become infective. Under certain conditions the virus requires a much longer incubation period within the insect body. This seems to indicate that the virus must either multiply in the insect or undergo a definite change there.

Certain viruliferous leafhoppers, either nymphs or adults, occasionally fail to produce infections in healthy plants. It is in all probability due to the temporary decrease or exhaustion of the virus in the salivary glands and like organs. Then the infections subsequently produced by the same individual must be attributed to the virus which has multiplied in the salivary glands etc.

All the evidence available at present, therefore, seems to indicate that the virus of dwarf disease of rice plant multiplies within the body of the leafhopper.

Hereupon the question again may be advanced whether the virus reproduces itself or the host cell affected by the virus produces it in turn as some metabolic product of cells. If we assume the latter view it is almost impossible to elucidate why comparatively few individuals of the leafhopper which have fed upon diseased plants are capable of producing the virus and why a period of as long as one or two months must elapse before the leafhopper can produce the virus as the metabolic product of cells. How can the rice plant be affected by the metabolic product of the leafhopper, an exceedingly small quantity of which is injected through the proboscis into the plant tissues? Is it a toxin? If that be granted, is it then readily acceptable that the affected cells of the rice plant produce the same toxin? On the other hand, the concept cannot be admitted that the toxin produced by the affected cells of the

rice plant may be different from what is injected by the leafhopper, because it is almost impossible to conceive that both the toxin injected into the plant tissues and the toxin which is subsequently produced in the cells and different from the originally injected toxin stimulate the cells to liberate one and the same toxin as the metabolic product. If we assume that the virus of dwarf disease of rice plant reproduces itself within the insect body, it cannot be acceptable to suppose that the same virus is produced by the rice plant as a product of cell metabolism. Thus the evidence seems to lend support to the view that the virus reproduces itself both within the insect body and the rice plant.

Consequently it cannot be denied that the virus is autonomous and multiplies utilizing the substances found in its environment, transforming them into its own substance. Furthermore the virus has the power of adaptation to reproduce itself both within the insect body and the plant tissues. D'HERELLE (1924, 1926)^{40, 41)} asserts that the power of assimilation and the power of adaptation are two fundamental properties of life. On the other hand, MCKINLEY (1929)¹³¹⁾ points out that reproduction is perhaps the most fundamental of all functions of organisms, because life would cease to exist after a time if there were no reproduction and that adaptation is "closely following in importance the power of reproduction," since living beings would die if they did not possess the power of adapting themselves to their environment. According to these concepts it is inevitable to conclude that the virus of dwarf disease of rice plant is of living nature. The virus, as stated before, in all probability lies beyond the limits of microscopic visibility. However, nobody can deny with certainty the existence of organized beings of ultramicroscopic dimensions, since the limits of microscopic visibility cannot be the line of demarcation between living and non-living beings.

Briefly stated, the sole hypothesis which is not in contradiction with any of the experimental results and which provides rational elucidation of all these facts is that the virus of dwarf disease of rice plant is a living entity of ultramicroscopic dimensions.

VIII. Summary

1. In the present paper a detailed account of dwarf disease of rice plant is given, presenting descriptions of the intracellular bodies associated with it, the results of transmission experiments and discus-

sions on the etiology of the disease.

2. The symptoms of the disease, its infectiousness, the invisibility of the causative agent under the microscope, the intracellular bodies associated with the affected tissues and the mode of transmission are considered to indicate that this disease belongs undoubtedly to the virus disease group.

3. Certain intracellular bodies are invariably present in the chlorotic tissue of the leaf as well as in the crown and root of the rice plant affected with dwarf disease. Similar bodies are found in the chlorotic tissues of the leaves of wheat, rye, *Panicum miliaceum* L., *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA, and *Alopecurus fulvus* L. affected with the virus of dwarf disease of rice plant. These bodies occur near or in close contact with the host cell-nucleus. They are usually round to oval but occasionally irregular in shape and much larger than the host nucleus. They are of rather homogeneous structure containing many vacuoles of various size and resemble the intracellular bodies in association with certain virus diseases of plants and animals. All attempts to demonstrate either a nucleus or chromatin granules in these bodies were unsuccessful but minute granules were successfully revealed in these structures, the exact nature of which remains unknown.

4. Any inclusion bodies or microorganisms of etiological significance, or visual evidence of the presence of the virus could not be discovered in the salivary glands and the alimentary canal, egg follicles in the ovarian tubules, nor likewise in the mycetome and other organs of the viruliferous leafhoppers, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL., the insect vector of the disorder.

5. The disease is not transmitted through the soil or the seeds of rice plant. All attempts to transmit the disease by mechanical inoculations with unfiltered juice of the diseased plants or by means of leaf mutilation have failed. Artificial inoculation of healthy rice plants with the macerated tissues and body fluid of viruliferous leafhoppers yielded also only negative results.

6. The leafhopper, *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL. is the sole means of transmission of the disease, acting as the vector. However, all individuals of this leafhopper are not capable of acting as carriers of the virus. Certain individuals fail to transmit the disease even when they have been hatched and reared on diseased plants. A majority of the offspring from infective females, which have been reared

on diseased plants are capable of producing infections in healthy rice plants. Sometimes a feeding period of 5 minutes was sufficient for a single leafhopper to transmit the disease and a feeding period of 30 minutes was long enough to enable most of the infective leafhoppers tested to produce infections in healthy plants. Most of the infective leafhoppers retained their infective power as long as they lived and certain leafhoppers produced infections in every healthy plant on which an individual was confined for 24 hours on consecutive days. In comparatively few instances the infectivity of leafhopper was apparently reduced or lost even under favorable conditions. A viruliferous leafhopper may not produce infections in every plant on which it feeds, even though the period of feeding may be 24 hours or longer. It seems, therefore, that certain viruliferous leafhoppers sometimes fail to transmit the disease, probably owing to the temporary exhaustion of the virus in the salivary glands or the anterior portion of the alimentary canal. Then the infections subsequently produced by the same individual must be attributed to the virus which either multiplied in the salivary glands etc. or migrated from some other portions of the body to these organs.

The progeny from infective female leafhoppers are either viruliferous or free from the virus while those from the crosses between uninfected females and infective males are entirely non-viruliferous. It appears that the eggs are not affected by the virus after they have been deposited but they are probably attacked at an earlier stage of development in the ovaries of the maternal insect. All the ova in the ovaries of an infective leafhopper are not always affected by the virus. A period from 1 to 14 days must elapse before most of the viruliferous nymphs become infective although certain individuals may transmit the disease immediately after emergence from eggs. Most of these viruliferous nymphs retain their infectivity during all the nymphal stages and through their entire adult life, without access of the virus. Only a small proportion of nonviruliferous leafhoppers, both nymphs and adults, became infective after feeding on dwarf diseased rice plants. The minimum period observed to be necessary for a non-viruliferous leafhopper to feed upon a diseased rice plant to pick up the virus was 3 days but the leafhoppers acquired the virus more readily after 10 to 50 days' feeding. Certain individuals, however, remained uninfected even when the feeding period on diseased plants extended to 50 or 70 days. The incubation period of the virus within the insect body could not be

definitely determined but it was apparently at least 10 to 40 days in certain cases where a temperature from 11° to 38°C. predominated while it was prolonged to more than 2 months in another case at a temperature ranging from 14° to 32°C. In short a relatively small portion of the number of leafhoppers acquire the virus after feeding on dwarf diseased rice plants but the individuals which have once become infective generally retain the virus through their entire life and certain infective females transmit the virus to their offspring through the eggs. It is evident, therefore, that the leafhopper does not mechanically transfer the virus from diseased to healthy plants but that some development or multiplication of the virus takes place within the body of the leafhopper.

7. *Panicum miliaceum* L., *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA, *Poa pratensis* L. and *Alopecurus fulvus* L. are subject to attack by the virus causing dwarf disease of rice plant. The symptoms produced on these plants closely resemble those on rice plants. Rye, wheat and oats are slightly susceptible to the disease, infection having been produced in only a small portion of inoculated plants. This may be partly due to the fact that these plants are uncongenial food plants for the leafhopper. The plants which have given negative results are corn, Italian millet, barley and Sorghum. These plants were also apparently unfavorable as food plants for the leafhopper under consideration. Since *Alopecurus fulvus* L. is a biennial wild grass abundantly growing on the rice fields throughout Japan, it is possible that the virus may be carried over the winter in this wild grass. However, there is no record to show the occurrence of symptoms similar to those of dwarf disease of rice plant on wild grasses.

8. Certain leafhoppers which had been collected on *Astragalus sinicus* L. growing on rice fields in the spring proved to be viruliferous. Hence it is more probable that the virus survives over winter in its insect carrier unless it is definitely established that the virus actually overwinters in *Alopecurus fulvus* and other biennial or perennial wild grasses under natural conditions or that *Astragalus sinicus* L. harbors the virus during the winter without showing any manifestations of disease.

9. All the evidence available at present seems to lend support to the view that the virus of dwarf disease of rice plant is autonomous and multiplies both in the insect body and in the plant tissues and that the virus in all probability lies beyond the limits of microscopic visibility.

The sole hypothesis, therefore, which is not in contradiction with any of the experimental results and which provides rational elucidation of all the facts is that the virus under consideration is a living entity of ultramicroscopic dimensions.

LITERATURE CITED

1. ALLARD, H. A. The mosaic disease of tobacco. *Sci. n. s.* 36: 875-876. 1912.
2. ——— The mosaic disease of tobacco. U.S. Dept. Agric. Bull. 40. 33 p. illust. 1914.
3. ——— Distribution of the virus of the mosaic disease in capsules, filaments, anthers, and pistils of affected tobacco plants. *Journ. Agric. Res.* 5: 251-256. illust. 1915.
4. ——— The mosaic disease of tomatoes and petunias. *Phytop.* 6: 328-335. 1916.
5. ——— Some properties of the virus of the mosaic disease of tobacco. *Journ. Agric. Res.* 6: 649-674. illust. 1916.
6. ——— Further studies of the mosaic disease of tobacco. *Journ. Agric. Res.* 10: 615-632. illust. 1917.
7. ANDÔ, H. On dwarf disease of rice plant. (in Japanese) *Journ. Jap. Agric. Soc.* No. 347: 1-3. 1910.
8. ARCHIBALD, E. S. Report of the acting Dominion botanist. *Rept. Dom. Exp. Farms* (1920): 58-64. 1921 (bean mosaic p. 62).
9. ATANASOFF, D. Stipple-streak disease of potato. *Meded. Landbouwhoogeschool Wageningen*, deel 24 verhandel. 5. 32 p. illust. 1922.
10. ——— Mosaic disease of flower bulb plants. *Bull. Soc. Bot. Bulgarie* (1928): 51-59. 1928.
11. BALL, E. D. The beet leafhopper. *Utah Agric. Exp. Sta. Ann. Rept.* 16: 16. 1906.
12. ——— The genus *Eutettix*. *Proc. Davenport Acad. Sci.* 12: 27-94. illust. 1907.
13. ——— The leafhoppers of the sugar beet and their relation to the "curly leaf" condition. U.S. Dept. Agric. Bur. Entomolog. Bull. 66: 33-52. illust. 1909.
14. BAUR, E. Zur Aetiologie der infektiösen Panachierung. *Ber. Deutsch. Bot. Ges.* 22: 453-460. 1904.
15. ——— Über die infektiöse Chlorose der Malvaceen. *Sitzungsber. Königl. Preuss. Akad. Wiss.* reprint 19 p. 1906.
16. BECKWIRTH, C. S. and HULTON, S. B. Cranberry false blossom and the blunted leafhopper. *New Jersey Agric. Exp. Sta. Bull.* 491. 16 p. illust. 1929.
17. BEIJERINCK, M. W. Über ein Contagium vivum fluidum als Ursache der Fleckenkrankheit der Tabaksblätter. *Verhandel. Kon. Akad. Wetensch. Amsterdam* II 6 (5): 1-22. illust. 1898.
18. BEWLEY, W. F. and CORBETT, W. The control of cucumber and tomato mosaic disease in glasshouses by the use of clean seed. *Ann. Appl. Biol.* 17: 260-266. 1930.
19. BLAKE, M. A., COOK, M. T. and CONNORS, C. H. Recent studies on peach

- yellow and little peach. New Jersey Agric. Exp. Sta. Bull. 356. 62 p. illust. 1921.
20. BLATTNY, C. Mosaika Konvalinky (*Convallaria majalis* L.). Ochrana Rostlin 9: 19-21. Illust. 1929. (original not seen)
 21. BOKURA, U. Dwarf disease of rice plant. Journ. Japanese Agric. Soc. No. 593: 56-59. 1930. (in Japanese)
 22. BONCQUET, P. A. and HARTUNG, W. J. The comparative effect upon sugar beets of *Eutettix tenella* BAKER from wild plants and from curly top beets. Phytop. 5: 348-349. illust. 1915.
 23. —, and STAHL, C. F. Wild vegetation as a source of curly top infection of sugar beets. Journ. Econ. Ent. 10: 392-397. illust. 1917.
 24. BÖNING, K. Die Mosaikkrankheit der Ackerbohne (*Vicia Faba* L.) Forsch. a. d. Gebiet der Pflanzenkr. 4: 43-111. illust. 1927.
 25. — Die Mosaikkrankheit der Rübe. Forsch. a. d. Gebiet d. Pflanzenkr. 3: 81-128. illust. 1927. (auch in Zeitschr. Vereins Deutsch. Zucker-Industrie 77: 13-72. illust. 1927)
 26. BOTJES, J. G. O. De Bladrolziekte van de Aardappel-plant. 136 p. (German summary 123-136) 1920.
 27. BRANDES, E. W. The mosaic disease of sugar cane and other grasses. U.S. Dept. Agric. Bull. 829. 26 p. illust. 1919.
 28. — Mosaic disease of corn. Journ. Agric. Res. 19: 517-522. illust. 1920.
 29. — Artificial and insect transmission of sugar cane mosaic. Journ. Agric. Res. 19: 131-138. 1920.
 30. —, and KLAPHAAK, P. J. Cultivated and wild hosts of sugar-cane or grass mosaic. Journ. Agric. Res. 24: 247-262. illust. 1923.
 31. BREHMER, W. v. und BÄRNER, J. Über die Viruskrankheiten bei der Kartoffel. Arbeit. Biol. Reichsanst. Land- u. Forstwirts. 18: 1-54. illust. 1930.
 32. BUCHNER, P. Tier und Pflanze in Symbiose. 2te Aufl. 900 p. 1930.
 33. CARSNER, E. Susceptibility of various plants to curly-top of sugar beet. Phytop. 9: 413-421. 1919.
 34. —, and STAHL, C. F. Studies on curly-top disease of the sugar beet. Journ. Agric. Res. 28: 297-319. illust. 1924.
 35. CLAYTON, E. E. A study of the mosaic disease of crucifers. Journ. Agric. Res. 40: 263-270. illust. 1930.
 36. CLINCH, P. Cytological studies of potato plants affected with certain virus diseases. Sci. Proc. Roy. Dublin Soc. n. s. 20: 143-172. illust. 1932.
 37. CLINTON, G. P. Chlorosis of plants with special reference to calico of tobacco. Connecticut Agric. Exp. Sta. Ann. Rept. 1914. Part VI: 357-424. illust. 1915.
 38. COOK, M. T. Histology and cytology of sugar cane mosaic. Journ. Dept. Agric. Porto Rico 9: 5-27. illust. 1925.
 39. DAIKUHARA, G. On dwarf disease of rice plant. Journ. Tokyo Chem. Soc. 25: 215-253. 1904 (also in Imp. Agr. Exp. Sta. Bull. 29: 163-193. 1904; Journ. Jap. Agric. Soc. 255: 4-8. 1902.) (in Japanese)
 40. D'HERELLE, F. Immunity in natural infectious disease. (English ed. by G. H. SMITH) 399 p. 1924.
 41. — The bacteriophage and its behavior. (English ed. by G. H. SMITH) 629 p. 1926.
 42. DICKSON, B. T. Studies concerning mosaic diseases. MacDonald Coll. Tech.

- Bull. 2. 125 p. illust. 1922.
43. ——— Mosaic studies IV. *Phytop.* 14: 346. 1924.
 44. ——— Tobacco and tomato mosaic. *Sci.* 62: 398. 1925.
 45. ———, and McROSTIE, G. P. Further studies on mosaic II. *Phytop.* 12: 42. 1922.
 46. DOBROSKY, I. D. Is the aster yellows virus detectable in its insect vector? *Phytop.* 19: 1009-1015. illust. 1929.
 47. ——— Cranberry false blossom disease spread by a leafhopper. *Sci.* 70 (1826): 635. 1929.
 48. ——— Studies on cranberry false blossom disease and its insect vector. *Contrib. Boyce Thompson Inst.* 3: 59-83. illust. 1931.
 49. DOOLITTLE, S. P. The mosaic disease of cucurbits. U.S. Dept. Agric. Bull. 879. 69 p. illust. 1920.
 50. ——— Soil transmission of tomato mosaic and streak in the green house. *Phytop.* 18: 155. 1928.
 51. ———, and GILBERT, W. W. Seed transmission of cucurbit mosaic by the wild cucumber. *Phytop.* 9: 326-327. 1919.
 52. ———, and JONES, F. R. The mosaic disease in the garden pea and other legumes. *Phytop.* 15: 763-772. illust. 1925.
 53. ———, and WALKER, M. N. Cross inoculation studies with cucurbit mosaic. *Sci.* 57: 477. 1923.
 54. ———, and ——— Further studies on the overwintering and dissemination of cucurbit mosaic. *Journ. Agric. Res.* 31: 1-58. illust. 1925.
 55. EARLE, F. S. Eradication as a means of control in sugar cane mosaic or yellow stripe. Porto Rico Dept. Agr. Ins. Exp. Sta. Bull. 22: 1-17. 1919.
 56. ECKERSON, S. H. An organism of tomato mosaic. *Pot. Gaz.* 81: 204-209. illust. 1926.
 57. ELZE, D. L. Die Übertragbarkeit mit dem Samen von Aukuba-Mosaik sowie Blattroll (Phloemnekrose) der Kartoffel. *Phytop. Zeitschr.* 3: 449-457. illust. 1931.
 58. ——— De overgang van virusziekten met het zaad, in het bijzonder bij de aardappel. *Tijdschr. Plantenziekten* 37: 189-199. illust. 1931. (original not seen)
 59. FAJARDO, T. G. Progress on experimental work with the transmission of bean mosaic. *Phytop.* 18: 155. 1928.
 60. ——— Studies on the mosaic disease of the bean (*Phaseolus vulgaris* L.). *Phytop.* 20: 469-494. illust. 1930.
 61. ——— Studies on the properties of the bean mosaic virus. *Phytop.* 20: 883-888. 1930.
 62. FAWCETT, G. L. The curly top of sugar beet in the Argentine. *Phytop.* 17: 407-408. 1927.
 63. FRANÇA, C. La flagellose des euphorbes. *Arch. Protistenkunde* 34: 108-132. 1914.
 64. FREIBERG, G. W. Studies in the mosaic diseases of plants. *Ann. Missouri Bot. Gard.* 4: 175-232. illust. 1917.
 65. FROMME, F. D., WINGARD, S. A. and PRIODE, C. N. Ringspot of tobacco; an infectious disease of unknown cause. *Phytop.* 17: 321-328. 1927.
 66. FUKUSHI, T. On the cause of mosaic disease of tobacco. (In Japanese) *Agric.*

- and Hort. 4: 1273-1283. illust. 1929.
67. — On broad bean mosaic. Journ. Pl. Protect. 17: 707-712, 779-784. illust. 1930. (in Japanese)
68. — On the intracellular bodies associated with dwarf disease of rice plant. Trans. Sapporo Nat. Hist. Soc. 12: 35-41. illust. 1931.
69. GARDNER, M. W. and KENDRICK, J. B. Soy bean mosaic. Journ. Agric. Res. 22: 111-115. illust. 1921.
70. —, and — Turnip mosaic. Journ. Agric. Res. 22: 123. illust. 1921.
71. —, and — Overwintering of tomato mosaic. Bot. Gaz. 73: 469-485. illust. 1922.
72. GOLDSTEIN, B. Cytological study of living cells of tobacco plants affected with mosaic disease. Torrey Bot. Club. Bull. 51: 261-273. illust. 1924.
73. — A cytological study of the leaves and growing points of healthy and mosaic diseased tobacco plants. Bull. Torrey Bot. Club 53: 499-599. illust. 1926.
74. — The x-bodies in the cells of dahlia plants affected with mosaic disease and dwarf. Bull. Torrey Bot. Club 54: 285-293. illust. 1927.
75. HENDERSON, R. G. Transmission of tobacco ring spot by seed of *Petunia*. Phyt. 21: 225-229. illust. 1931.
76. HINO, I. Early important records on phytopathological science in the Orient. (in Japanese) Agric. and Hort. 2: 1223-1232. illust. 1927.
77. Hiroshima Agr. Exp. Sta. Experiments with dwarf disease of rice plant. Hiroshima Agr. Exp. Sta. Bull. 7: 21-39. 1903. (in Japanese)
78. HOGGAN, I. A. Cytological studies on virus diseases of solanaceous plants. Journ. Agric. Res. 35: 651-671. illust. 1927.
79. — The peach aphid (*Myzus persicae* SULZ.) as an agent in virus transmission. Phyt. 19: 109-123. illust. 1929.
80. — Some viruses affecting spinach, and certain aspects of insect transmission. Phyt. 23: 446-474. illust. 1933.
81. HOLMES, F. O. The relation of *Herpetomonas elmassiani* (MIGONE) to its plant and insect hosts. Biol. Bull. 49: 323-337. illust. 1925.
82. — Cytological study of the intracellular body characteristic of Hippeastrum mosaic. Bot. Gaz. 86: 50-58. illust. 1928.
83. HUNGER, F. W. T. Neue Theorie zur Ätiologie der Mosaikkrankheit des Tabaks. Ber. Deutsch. Bot. Ges. 23: 415-418. 1905.
84. Hyōgo Agr. Exp. Sta. Experiments with dwarf disease of rice plant. (in Japanese) Results of Agricultural Experiments. Report 1: 68-82. 1895, Rept. 3: 16-18. 1896, Rept. 5: 21-22. 1897, Rept. 7: 139-141. 1898, Rept. 9: 121-123. 1899.
85. Imperial Agric. Exp. Sta. Ann. Rpts. for 1906: 53-54, for 1909: 59, for 1912: 29 (1914), for 1913-1915: 40-41. (1917), for 1916: 28. (1918), for 1922: 68-70 (1924), for 1923: 33 (1925) (in Japanese)
86. ISHIKAWA, T. The merit of HATSUZO HASHIMOTO, the earliest investigator of dwarf disease of rice plant. Journ. Pl. Protect. 15: 218-222. 1928. (in Japanese)
87. IWANOWSKI, D. Über zwei Krankheiten der Tabakspflanze. Land- und Forstwirtschaft (Russisch) 1892 (original not seen, abstract in Beih. Bot. Centralbl. 3: 266-268. 1893).

88. ——— Über die Mosaikkrankheit der Tabakspflanze. Bull. Acad. Imper. Sci. St. Petersburg. II 3: 67-70. 1892. (original not seen)
89. ——— Über die Mosaikkrankheit der Tabakspflanze. Zeitschr. Pflanzenkr. 13: 1-41. illust. 1903.
90. Japanese Department of Agriculture and Forestry Bur. Agric. A survey on the distribution of plant diseases and insect pests. (in Japanese) 2, 341 p. 1929. (dwarf disease of rice plant pp. 16-19)
91. ——— Distribution of green manure crops. 21 p. 3 graphs. 1932. (in Jap.)
92. Japanese Dept. Education Dwarfed rice plant, its cause and control measures. (in Japanese) Official Gazette No. 2192: 231. Oct. 18, 1890.
93. JOHNSON, J. Transmission of viruses from apparently healthy potatoes. Wisconsin Agr. Exp. Sta. Res. Bull. 63. 12 p. illust. 1925.
94. ——— The classification of plant viruses. Wisc. Agr. Exp. Sta. Res. Bull. 76. 16 p. illust. 1927.
95. ———, and OGDEN, W. B. The overwintering of the tobacco mosaic virus. Wisc. Agr. Exp. Sta. Res. Bull. 95. 25 p. 1929.
96. JONES, L. K. The mosaic disease of beets. Washington Agr. Exp. Sta. Bull. 250. 16 p. illust. 1931.
97. Kagoshima Agr. Exp. Sta. Ann. Rpts. for 1912: 46 (1913), for 1913: 48 (1914), for 1914: 39-43 (1915), for 1915: 94 (1916). (in Japanese)
98. KASAI, M. Observations and experiments on the leafroll disease of the Irish potato in Japan. Ber. Ohara Inst. landw. Forsch. 2: 47-77. 1921.
99. ——— Investigations on the Nelson's bodies as observed in the leafroll, mosaic, and healthy plants. Ber. Ohara Inst. Landw. Forsch. 2: 443-461. illust. 1924.
100. KENDRICK, J. B. and GARDNER, M. W. Soy bean mosaic: seed transmission and effect on yield. Journ. Agr. Res. 27: 91-98. 1924.
101. KIRKPATRICK, T. W. Preliminary note on leaf-erinkle of cotton in the Gezira Area, Sudan. Bull. Entomol. Research 21: 127-137. 1930.
102. ——— Further studies on leaf-curl of cotton in the Sudan. Bull. Entom. Res. 22: 323-363. illust. 1931.
103. KLEBAHN, H. Die Alloiophyllie der *Anemone nemorosa* und ihre vermutliche Ursache. Ber. Deutsch. Bot. Ges. 43: (32)-(37) illust. 1926 (auch in *Planta* 1: 419-440. illust. 1926)
104. ——— Experimentelle und cytologische Untersuchungen im Anschluss an Alloiophyllie und Viruskrankheiten. *Planta*, Arch. Wiss. Bot. 6: 40-95. illust. 1928.
105. KUNKEL, L. O. A possible causative agent for the mosaic disease of corn. Hawaiian Sugar Plant. Assoc. Exp. Sta. Bull. 3: 44-58. illust. 1921.
106. ——— Amoeboid bodies associated with Hippeastrum mosaic. Sci. 55: 73. 1922.
107. ——— Insect transmission of yellow stripe disease. Hawaiian Planters' Record 26: 58-64. illust. 1922. (original not seen)
108. ——— Insect transmission of aster yellows. Phytop. 14: 54. 1924.
109. ——— Histological and cytological studies on the Fiji disease of sugar cane. Hawaiian Sugar Planters' Assoc. Exp. Sta. Bull. 3: 99-107. illust. 1924.
110. ——— Further studies on the intracellular bodies associated with certain mosaic diseases. Hawaiian Sugar Planters' Assoc. Exp. Sta. Bull.

- 3: 108-114. illust. 1924.
111. ——— Studies on aster yellows. Amer. Journ. Bot. 13: 646-705. illust. 1926.
112. ——— The corn mosaic of Hawaii distinct from sugar cane mosaic. Phytop. 17: 41. 1927.
113. ——— Insect transmission of peach yellows. Contrib. Boyce Thompson Inst. 5: 19-28. illust. 1933.
114. KURIBAYASHI, K. On the seed transmission of bean mosaic. Journ. Pl. Protect. 13: 199-210. (in Japanese) 1926.
115. ——— Studies on the stripe disease of rice plant. (in Japanese) Nagano Agr. Exp. Sta. Bull. 2: 45-69. illust. 1931.
116. ——— On the relation of *Delphacodes striatellus* FALL. to the transmission of the stripe disease of rice plant. (in Japanese) Journ. Pl. Protect. 18: 565-571, 636-640. 1931.
117. Kyôto Agr. Exp. Sta. On dwarf disease of rice plant. Special Bull. 11. 12 p. 1910. (in Japanese)
118. LIKHITÉ, V. The nature and relations of the intracellular inclusions present in the mosaic of tobacco. Meded. Landbouwhoogeschool Wageningen. 33 (1) 30 p. illust. 1929. (original not seen)
119. ——— Cytological aspects of the virus diseases in plants. Meded. Landbouwhoogeschool Wageningen 33 (2) 12 p. 1929. (Original not seen)
120. LIND, J. Runkelroernes Mosaiksyge. (Mosaikkrankheit der Runkelrüben) Tidskr. Planteavl. 22: 444-457. 1915. (original not seen)
121. LIRO, J. I. Über die Mosaikkrankheit der *Prunella vulgaris*. Ann. Soc. Zool. Bot. Fenn. Vanams 11: 143-149. 1930. (original not seen)
122. LYON, H. L. A new canker disease now epidemic in Fiji. Hawaiian Planters' Record 3: 200-205. 1910.
123. ——— Three major cane diseases: mosaic, sereh, and Fiji disease. Hawaiian Sugar Planters' Assoc. Exp. Sta. Bull. 3: 1-43. illust. 1921.
124. MAYER, A. Über die Mosaikkrankheit des Tabaks. Landwirt. Versuchsst. 32: 451-467. illust. 1886.
125. MATSUMOTO, T. Further studies on legume mosaic. Journ. Pl. Protect. 9: 517-520. 1922. (in Japanese)
126. MATZ, J. Infection and nature of the yellow stripe disease of cane (mosaic, mottling, etc.) Journ. Dept. Agric. Porto Rico 3: 65-82. illust. 1919.
127. McCLINTOCK, J. A. Lima bean mosaic. Phytop. 7: 60-61. 1917.
128. ——— Peach rosette, an infectious mosaic. Journ. Agr. Res. 24: 307-315. illust. 1923.
129. ——— and SMITH, L. B. True nature of spinach-blight and relation of insects to its transmission. Journ. Agr. Res. 14: 1-60. illust. 1918.
130. MCKAY, M. B., BRIERLEY, P. and DYKSTRA, T. P. Tulip 'breaking' is proved to be caused by mosaic infection. U.S. Dept. Agric. Yearbook of Agric. 1928: 596-597. illust. 1929.
131. MCKINLEY, E. B. Filterable viruses and rickettia diseases. Philippine Journ. Sci. 39: 1-416. illust. 1929.
132. MCKINNEY, H. H. Investigations of the rosette disease of wheat and its control. Journ. Agric. Res. 23: 771-800. illust. 1923.
133. ——— A mosaic disease of winter wheat and winter rye. U.S. Dept. Agr. Bull. 1361. 10 p. 1925.
134. ——— Certain aspects of the virus diseases. Phytop. 15: 189-202. 1925.

135. — Virus mixtures that may not be detected in young tobacco plants. *Phytop.* 16: 893. 1926.
136. — Quantitative and purification methods in virus studies. *Journ. Agr. Res.* 35: 13-38. illust. 1927.
137. —, ECKERSON, S. H. and WEBB, R. W. The intracellular bodies associated with the rosette disease and a mosaic like leaf mottling of wheat. *Journ. Agr. Res.* 26: 605-608. illust. 1923.
138. —, —, and — Intracellular bodies associated with a "mosaic" of *Hippeastrum Johnsonii*. *Phytop.* 13: 41-42. 1923.
139. —, WEBB, R. W. and DUNGAN, G. H. Wheat rosette and its control. *Illinois Agr. Exp. Sta. Bull.* 264: 275-296. illust. 1925.
140. —, WEBB, R. W. The dilution method as a means for making certain quantitative studies of viruses. *Phytop.* 16: 66. 1926.
141. McWHORSTER, F. P. The nature of the organism found in the Fiji galls of sugar cane. *Philippine Agriculturist* 11: 103-111. illust. 1922.
142. MERKEL, L. Beiträge zur Kenntnis der Mosaikkrankheit der Familie der Papilionaceen. *Zeitschr. Pflanzenkr.* 39: 289-347. illust. 1929.
143. MEULEN, J. G. J. Voorloopig onderzoek naar de specialisatie en de infectiebronnen der mosaikziekten van landbouwgewassen. *Tijdschr. Plantenziekten* 34: 155-176. 1928. (original not seen)
144. Miyazaki Agr. Exp. Sta. *Ann Repts. for 1913: 32 (1914), for 1914: 29-30 (1915).* (in Japanese)
145. — Dwarf disease of rice plant. *Special Bull.* 5. 26 p. illust. 1915. (,)
146. MOUTIA, A. Sur un des modes de transmission de la mosaïque du tabac. *Rev. Agric. de l'Île Maurice* 40: 179-180. 1928. (original not seen)
147. MULVANIA, M. Studies on the nature of the virus of tobacco mosaic. *Phytop.* 16: 853-871. 1926.
148. MURATA, T. Insect pests of the rice and barley and their control. (in Japanese) 364 p. 1915.
149. — Dwarf disease of rice plant. *Journ. Japanese Agr. Soc.* No. 604: 47-50. 1931. (in Japanese)
150. MURPHY, P. A. Investigation of potato diseases. *Dominion Exp. Farms Bull.* 44. (2nd ser.) 86 p. illust. 1921.
- 151a. — Investigations on the leaf-roll and mosaic diseases of the potato. *Journ. Dept. Agric. & Tech. Instruct. Ireland* 23: 20-34. illust. 1923.
- 151b. —, and MCKAY, R. Investigations on the leafroll and mosaic diseases of the potato. *Journ. Dept. Agric. & Tech. Instruct. Ireland* 23: 344-364. illust. 1924.
152. Nagano Agr. Exp. Sta. *Ann. Rept. for 1915: 130-134.* (in Japanese)
153. Nara Agr. Exp. Sta. *Results of Agricultural Experiments. Report 3: 51-61.* (Experiments with dwarf disease). 1897. (in Japanese)
154. NARASIMHAN, M. J. Note on the occurrence of intracellular bodies in spike disease of sandal (*Santalum album* LINN.) *Phytop.* 18: 815-817. illust. 1928.
155. — Cytological investigations on the spike disease of sandal, *Santalum album*. *Phytop.* 23: 191-202. Illust. 1933.
156. NELSON, R. The occurrence of protozoa in plants affected with mosaic and related diseases. *Michigan Agr. Exp. Sta. Tech. Bull.* 58. 30 p. illust. 1922.

157. NEWHALL, A. G. Seed transmission of lettuce mosaic. *Phytop.* 13: 104-106. 1923.
158. OGILVIE, L. and GUTERMAN, C. E. F. A mosaic disease of the Easter lily. *Phytop.* 19: 311-315. illust. 1929.
159. Okayama Agr. Exp. Sta. Experiments with dwarf disease. Results of Agricultural Experiments. Report 3: 62-70. 1902, 5: 57-64. 1903, 7: 51-53. 1904, 11: 131-138. 1906, 13: 169-171. 1907, 15: 109-111. 1908, 19: 69. 1909. (in Japanese)
160. Government of Okayama Prefecture. Rice of the Prefecture of Okayama. 300 p. 1910. (see pp. 189-193). (in Japanese)
161. PALM, B. T. De mozaiekziekte van de tabak een chlamydozoonose? (voorloopige mededeeling) Deliproefsta. Medan-Sumatra Bull. 15. 10 p. (English transl. 7-10). 1922.
162. PETRI, L. Sull' "arriciamento" della vite. *Boll. R. Staz. Patol. Veg.* 11: 61-83. illust. 1931. (original not seen)
163. PIERCE, W. H. and HUNGERFORD, C. W. Symptomatology, transmission, infection and control of bean mosaic in Idaho. *Idaho Agric. Exp. Sta. Res. Bull.* 7. 37 p. illust. 1929.
164. PLAKIDAS, A. G. Strawberry xanthosis (yellows), a new insect-borne disease. *Journ. Agr. Res.* 35: 1057-1090. illust. 1927.
165. ——— Strawberry dwarf. *Phytop.* 18: 439-444. illust. 1928.
166. PRIODE, C. N. Further studies in the ring-spot disease of tobacco. *Amer. Journ. Bot.* 15: 88-93. illust. 1928.
167. PROWAZEK, S. Chlamydozoa. I Zusammenfassende Übersicht. *Arch. Protistenkunde* 10: 336-358. 1907.
168. RANKIN, W. H. Leaf curl and mosaic or yellows of the cultivated red raspberry. *Interim. Rept. Domin. Bot. Canada Dept. Agr.* 1921-22: 30-60. 1922.
169. RAWLINS, T. E. Cytology of root tips from sugar beets having the curly-top disease. *Phytop.* 16: 761. 1926.
170. ———, and JOHNSON, J. Cytological studies of the mosaic disease of tobacco. *Amer. Journ. Bot.* 12: 19-32. illust. 1925.
171. REDDICK, D. and STEWART, V. B. Transmission of the virus of bean mosaic in seed and observations on thermal death-point of seed and virus. *Phytop.* 9: 445-450. 1919.
172. REINKING, O. A. Fiji disease of sugar cane. *Sugar Cent. and Plant News* 2: 41-48. 1921. (original not seen)
173. RIDGWAY, R. Color standards and color nomenclature. 1912.
174. ROBBINS, W. W. Mosaic disease of sugar beets. *Phytop.* 11: 349-365. 1921.
175. ROSEN, H. R. The mosaic disease of sweet potatoes with special reference to its transmissibility. *Arkansas Agr. Exp. Sta. Bull.* 213: 2-16. 1926.
176. SAMUEL, G., BALD, J. G., and PITTMAN, H. A. Investigations on "spotted wilt" of tomatoes. *Com. Australia Coun. Sci. Indust. Res. Bull.* 44. 1930. (original not seen)
177. SCHAFFNIR, E. und JÖHNSEN, A. Beiträge zur Kenntnis der Blattrollkrankheit der Kartoffel. *Phytop. Zeitschr.* 5: 603-612. illust. 1933.
178. ———, und WEBER, H. Über das Vorkommen von intrazellularen Körpern in den Geweben mosaikkranker Rüben. *Forsch. a. d. Gebiet Pflanzenkr.* 4: 23-42. illust. 1927.

179. SCHULTZ, E. S. A transmissible mosaic disease of Chinese cabbage, mustard and turnip. Journ. Agr. Res. 22: 173-177. illust. 1921.
180. —, and FOLSOM, D. Transmission of the mosaic disease of Irish potatoes. Journ. Agr. Res. 19: 315-337. illust. 1920.
181. —, and — Leafroll, net-necrosis, and spindling sprout of the Irish potato. Journ. Agr. Res. 21: 47-80. illust. 1921.
182. —, and — Transmission, variation, and control of certain degeneration diseases of Irish potatoes. Journ. Agr. Res. 25: 43-117. illust. 1923.
183. —, and — Infection and dissemination experiments with degeneration diseases of potatoes, observations in 1923. Journ. Agr. Res. 30: 493-528. illust. 1925.
184. —, —, HILDEBRANDT, F. M. and HAWKINS, L. A. Investigations on the mosaic disease of the Irish potato. Journ. Agr. Res. 17: 247-273. illust. 1919.
185. SEIN, F. A new mechanical method for artificially transmitting sugar cane mosaic. Journ. Dept. Agr. Porto Rico 14: 49-68. 1930.
186. SEVERIN, H. H. P. Minimum incubation periods of causative agent of curly leaf in beet leafhopper and sugar beet. Phytop. 11: 424-429. illust. 1921.
187. — Curly leaf transmission experiments. Phytop. 14: 80-93. illust. 1924.
188. — Modes of curly-top transmission by the beet leafhopper, *Eutettix tenellus* (BAKER). Hilgardia 6: 253-276. illust. 1931.
189. SHAW, H. B. The curly-top of beets. U.S. Dept. Agr. Bur. Pl. Ind. Bull. 181. 46 p. illust. 1910.
190. SHEFFIELD, F. M. L. The formation of intracellular inclusions in solanaceous hosts infected with aucuba mosaic of tomato. Ann. Appl. Biol. 18: 471-493. illust. 1931.
191. —, and SMITH, J. H. Intracellular bodies in plant virus diseases. Nature 125 (3145): 200. 1930.
192. Shiga Agr. Exp. Sta. Experiments with the leafhoppers. Results of experiments with insect pests. Reports 1: 111-169. 1899, 2: 1-26. 1900, 3: 25-55. 1901, 4: 19-65. 1902, 5: 1-37. (1)-(36). 1904, 6: 1-43. 1906, 7 and 8: 1-43. and (1)-(50). 1908.
193. — Experiments with dwarf disease of rice plant. Spec. Bull. 56-65. 1906. No. 2: 35-51. 1910.
194. — Ann. Reports for 1907: 55-59. 1908, for 1908: 52-56. 1909, for 1909: 52-54. 1910, for 1910: 54-56. 1911, for 1911: 58-60. 1912, for 1912: 75-78. 1913, for 1913: 128-130. 1914, for 1914: 181-183. 1915, for 1915: 167-168. 1916.
195. SMITH, E. F. Peach yellows: a preliminary report. U.S. Dept. Agr. Bot. Div. Bull. 9. 254 p. illust. 1888.
196. SMITH, F. F. Some cytological and physiological studies of mosaic diseases and leaf variegations. Ann. Missouri Bot. Gard. 13: 425-484. illust. 1926.
197. SMITH, J. H. Experiments with a mosaic disease of tomato. Ann. Appl. Biol. 15: 155-167. illust. 1928.
198. — Intracellular inclusions in mosaic of *Solanum nodiflorum*. Ann. Appl. Biol. 17: 213-222. illust. 1930.
199. — Virus diseases in plants. Biol. Rev. 5: 159-170. 1930. (original not

- seen)
200. SMITH, K. M. On a curious effect of mosaic disease upon the cells of the potato leaf. *Ann. Bot.* 38: 385-388. illust. 1924.
 201. ——— Studies on potato virus diseases. IX. Some further experiments on the insect transmission of potato leafroll. *Ann. Appl. Biol.* 18: 141-157. illust. 1931.
 202. SMITH, R. E. and BONCQUET, P. A. New light on curly top of the sugar beet. *Phytop.* 5: 103-107. 1915.
 203. ———, and ——— Connection of a bacterial organism with curly leaf of the sugar beet. *Phytop.* 5: 335-342. 1915.
 204. SOROKIN, H. Phenomena associated with the destruction of the chloroplasts in tomato mosaic. *Phytop.* 17: 363-379. illust. 1927.
 205. STAHL, C. F. Corn stripe disease in Cuba not identical with sugar cane mosaic. *Trop. Plant Res. Found. Bull.* 7. 12 p. illust. 1927.
 206. ———, and CARLSNER, E. Obtaining beet leafhoppers nonvirulent as to curly-top. (Preliminary paper) *Journ. Agr. Res.* 14: 393-394. 1918.
 207. ———, and ——— A discussion of *Eutettix tenella* BAKER as a carrier of curly top of sugar beets. *Journ. Econ. Entom.* 16: 476-479. 1923.
 208. STOREY, H. H. Streak disease of sugar cane. *Union S. Africa Dept. Agric. Sci. Bull.* 39. 30 p. illust. 1925.
 209. ——— Streak disease, an infectious chlorosis of sugar cane not identical with mosaic disease. *Rept. Proc. Imp. Bot. Conf. London 1924*: 132-144. illust. 1925.
 210. ——— The transmission of streak disease of maize by the leafhopper, *Balclutha mbila* NAUDE. *Ann. Appl. Biol.* 12: 422-439. illust. 1925.
 211. ——— Recent researches on plant virus diseases (Summary) *So. African Journ. Sci.* 23: 307. 1927. (original not seen)
 212. ——— Transmission studies of maize streak disease. *Ann. Appl. Biol.* 15: 1-25. illust. 1928.
 213. ——— The bearing of insect vectors on the differentiation and classification of plant viruses. *Deux. Congr. Internat. Path. Comp. Paris. Compt. rend. et Communic.*: 471-479. 1931.
 214. ——— The inheritance by a leafhopper of the ability to transmit a plant virus. *Nature* 127 (3216): 928. 1931.
 215. ———, and BOTTOMLEY, A. M. The rosette disease of peanuts. *Ann. Appl. Biol.* 15: 26-45. illust. 1928.
 216. SWEZY, O. Factors influencing the minimum incubation periods of curly top in the beet leafhopper. *Phytop.* 20: 93-100. illust. 1930.
 217. ———, and SEVERIN, H. H. P. A Rickettsia-like microorganism in *Eutettix tenellus* (BAKER), the carrier of curly top of sugar beets. *Phytop.* 20: 169-178. illust. 1930.
 218. TAKAMI, N. On dwarf disease of rice plant and *Nephotettix apicalis* MOTSCH. var. *cincticeps* UHL. *Journ. Jap. Agric. Soc. No.* 241: 22-30. 1901. (in Japanese)
 219. TAKATA, K. Results of experiments with dwarf disease of rice plant. (in Japanese) *Journ. Jap. Agric. Soc. No.* 171: 1-4. 1895; No. 172: 13-32. 1896.
 220. TAUBENHAUS, J. J. Mosaic disease of the sweet pea. *Delaware Agr. Exp. Sta. Bull.* 106: 53-61. 1914.

221. TOWNSEND, C. O. Curly-top, a disease of the sugar beet. U.S. Dept. Agric. Bur. Pl. Ind. Bull. 122. 39 p. illust. 1908.
222. VALLEAU, W. D. Tobacco seed beds in Kentucky. Pl. Dis. Reporter 14: 113. (mimeographed) 1930 (original not seen)
223. ——— Two seed-transmitted ring-spot disease of tobacco. Phytop. 22: 28. 1932.
224. ——— Seed transmission and sterility studies of two strains of tobacco ringspot. Kentucky Agr. Exp. Sta. Bull. 327: 43-80. illust. 1932.
225. WALKER, M. N. A comparative study of the mosaic disease of cucumber, tomato and *Physalis*. Phytop. 16: 431-458. 1926.
226. WEBB, R. W. Soil factors influencing the development of the mosaic disease in winter wheat. Journ. Agr. Res. 35: 587-614. illust. 1927.
227. ——— Further studies on the soil relationships of the mosaic disease of winter wheat. Journ. Agr. Res. 36: 53-75. illust. 1928.
228. WESTERDIJK, J. Die Mosaikkrankheit der Tomaten. Meded. Phytop. Lab. "Wille Commelin Sholten" 20 p. illust. 1910.
229. WILBRINK, G. en LEDEBOER, F. Bijdrage tot de Kennis der Gele Strepenziekte. Arch. Suikerindust. Nederl. Indie. 18: 465-518. 1910 (also in Meded. Proefstat. Java-Suikerindust. deel. 2. No. 39: 443-495. illust. 1910). (original not seen)
230. WINGARD, S. A. Hosts and symptoms of ring spot, a virus disease of plant. Journ. Agr. Res. 37: 127-154. illust. 1928.
231. WOODS, M. W. Intracellular bodies in ring spot. Phytop. 23: 38. 1933.
232. WORTLEY, E. J. Potato leaf-roll: its diagnosis and cause. Phytop. 8: 507-529. 1918.
233. Yamanashi Agr. Exp. Sta. Ann. Repts. for 1916: 88-91 (1917); for 1917: 87-89, 146-147 (1918); for 1918: 48, 69 (1919); for 1919: 39 (1920) (in Japanese)
234. YONEMARU, C. and Aso, K. Report of the investigations of dwarf disease of rice plant. Kyôto Agr. Exp. Sta. Special Bull. No. 1. 178 p. illust. 1905. (in Japanese)
235. ZELLER, S. M. Dwarf disease of blackberries. Phytop. 17: 629-648. 1927.
236. BRANDENBURG, E. Ueber Mosaikkrankheiten an Compositen. Forsch. a. d. Gebiet d. Pflanzenkr. 5: 39-72. illust. 1928.
237. GOLDSTEIN, B. Nuclear form as related to functional activities of normal and pathological cells. Bot. Gaz. 86: 365-383. illust. 1928.
238. MAYER, A. Heilung der Mosaikkrankheit des Tabaks. Landwirts. Versuchstationen. 35: 339-340. 1888.

EXPLANATION OF PLATE

Plate I.

- Fig. 1. Healthy (left) and affected (right) rice plants.
Fig. 2. Healthy (right) and affected (left) plants of *Echinochloa Crus-galli* BEAUV. subsp. *colona* HONDA var. *edulis* HONDA.
Fig. 3. Healthy (right) and affected (left) wheat plants.

Plate II.

- Fig. 4. Diseased leaves of the rice plant showing the typical streaks along the veins.
Fig. 5. Healthy (on the left end) and diseased leaves of *Echinochloa Crus-galli* subsp. *colona* var. *edulis*.
Fig. 6. Diseased leaves of *Panicum miliaceum* L.
Fig. 7. Diseased leaves of the wheat plant.

Plate III.

- Fig. 8. A portion of a longitudinal section of the diseased leaf of rice plant, showing the intracellular bodies marked × (Fixed in GILSON'S fluid and stained with GIEMSA'S stains)
Fig. 9. do.
Fig. 10. A portion of a longitudinal section of the root of the affected rice plant, showing the intracellular bodies marked × (Fixed in chromo-acetic solution and stained with safranin and gentian violet)
Fig. 11. A portion of a cross section of diseased leaf showing the chlorotic tissue (Fixed in chromo-acetic solution and stained with safranin)
Fig. 12. A portion of the section shown in Fig. 8, highly magnified.
Fig. 13. A portion of the section shown in Fig. 9, highly magnified.

Plate IV.

- Fig. 14. A portion of the section shown in Fig. 9, highly magnified.
Fig. 15 and 16. A portion of a cross section of the affected leaf of rice plant, showing an intracellular body in the mortar cell photographed at different optical levels to show the host cell-nucleus and the vacuoles in the body (Fixed in formol acetic alcohol and stained with safranin and gentian violet)
Fig. 17. A portion of a longitudinal section of a leaf of the wheat plant affected by the virus of dwarf disease of rice plant (Fixed in GILSON'S fluid and stained with GIEMSA'S stains) x: intracellular body, n: cell-nucleus.
Fig. 18. do.
Fig. 19. do.

Plate V.

- Fig. 20. The leafhopper, *Nephotettix apicalis* MOTSCH. var. *cineticeps* UHL., male (left) and female (right)

- Fig. 21. A longitudinal section of a male leafhopper (Fixed in GILSON's fluid and stained with MALLORY's stains)
- Fig. 22. A longitudinal section of the head and prothorax of a male leafhopper, showing salivary glands (s), oesophagus (oe), cephalic ganglion (g) and muscles (m)
- Fig. 23. A longitudinal section of the abdomen of a male leafhopper, showing the testes (t), mycetomes (myc), fat body (f) and midintestine (i)
- Fig. 24. A longitudinal section of the head and prothorax of a male leafhopper, showing salivary glands (s)
- Fig. 25. A longitudinal section of the abdomen of a male leafhopper showing testes (t) and midintestine (i)
- Fig. 26. A longitudinal section of the abdomen of a male leafhopper showing the mycetome (myc), midintestine (i), fat bodies (f) and Malpighian tubules (M)

Plate VI.

- Fig. 27. A portion of a longitudinal section of the abdomen of a female leafhopper showing the ovarian tubules (o), mycetomes (myc) and midintestine (i)
- Fig. 28. A portion of the mycetome, showing the mycelium-like strands which fill up the mycetocytes.
- Fig. 29. A portion of the mycetome. Note the inner mycetocytes which have collapsed, leaving the nuclei among clumps of cytoplasm.

Fig. 3

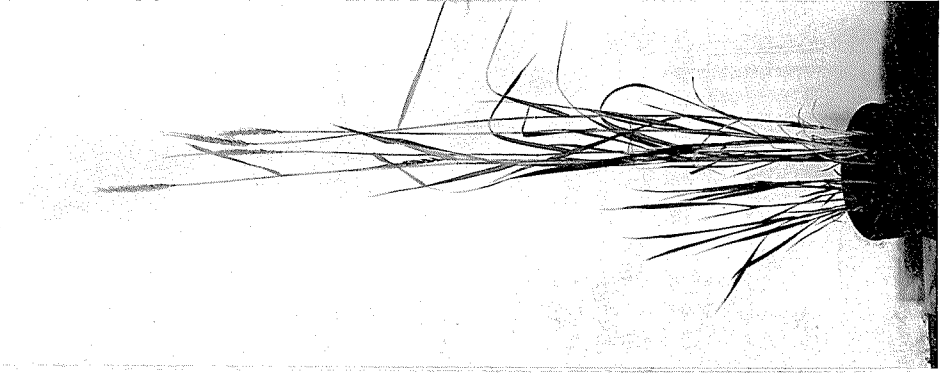


Fig. 2

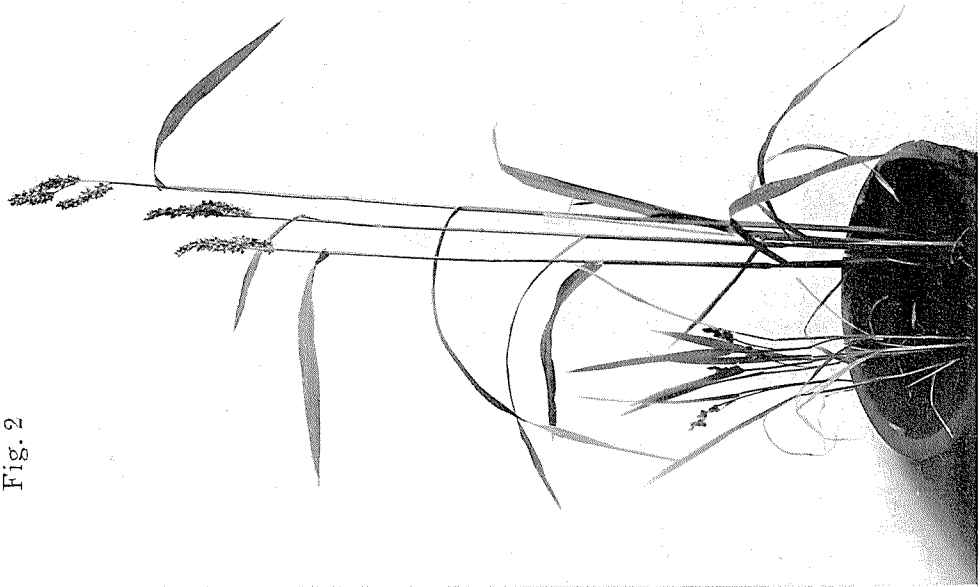


Fig. 1



