

## Wheat Dwarf Virus Disease

JOSEF VACKE

Research Institute of Plant Production, Czechoslovak Academy of Agricultural Sciences,  
Ruzyně near Praha\*

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### Virová zakrslost pšenice

V práci je podán důkaz o virové povaze zakrslosti pšenice, vyskytující se v některých oblastech ČSSR. Virus se podařilo přenést přirozeně infikovanými imagy kříška *Psammotettix alienus* DAHLB. na jarní pšenici a jarní ječmen. Rovněž se ukázalo, že je ve vysokém procentu přenosný larvami vypěstovanými z vajec a uměle infikovanými na zdroji nákazy. Pokus o přenos viru mšicemi *Rhopalosiphon oxyanthae* (SCHRANK.) a *Sitobium granarium* (KIRBY) MORDV. byl negativní. Rovněž se nepodařilo přenést virus půdou, obilkami a mechanickou inokulací šťávy z nemocných rostlin.

Virus zakrslosti pšenice by mohl podle přenašeče stát nejbližší viru ruské mozaiky ozimé pšenice. Liší se však od něho symptomy vyvolávanými na hostitelských obilovinách, jiným vektorem a nedostatkem inklusí v cytoplasmě infikovaných rostlin. Tyto skutečnosti naznačují, že jde o nový doposud nepopsaný virus.

### Summary

Proof is given in this paper that dwarfing of wheat, which occurs in several regions of Czechoslovakia, is a virus infection. The virus was transmitted by means of naturally infected imagos of the leaf-hopper *Psammotettix alienus* DAHLB. to spring wheat and spring barley. It was also found that it is transmissible to a high degree by larvae grown from eggs which are then artificially infected at the source of infection. An attempt to transmit the virus by means of the aphids *Rhopalosiphon oxyanthae* (SCHRANK.) and *Sitobium granarium* (KIRBY) MORDV. was negative. Nor was it possible to transmit it by soil, wheat grains or by mechanical inoculation with sap from diseased plants.

The Russian mosaic virus of winter wheat would seem, in view of its vector, to be the most closely related to the wheat dwarf virus. It differs, however,

\* Address: Výzkumný ústav rostlinné výroby ČSAZV, Praha-Ruzyně 507.

as regards the symptoms produced on the host cereals, by having a different vector and by the absence of inclusions in the cytoplasm of infected plants. These facts indicate that we are probably dealing with a new virus, which has not previously been described.

### Introduction

In 1960 winter wheat suffered considerable damage in certain parts of Moravia (Haná, southern Moravia) and Bohemia (the vicinity of Prague). Attention was drawn to this by DLABOLA (1960), who pointed to the marked occurrence of the leaf-hopper *Psammotettix alienus* DAHLB. on the diseased crops and, according to his later personal communication, this insect is the cause of the disease. In the light of previous experience with virus diseases of cereals in Czechoslovakia it could be assumed that in this case, too, virus infection was involved. A general investigation of this question has been made during the vegetation period of 1960.

### Material and Methods

Imagos and larvae of the leaf-hopper *Psammotettix alienus* were used for the vector experiments. The imagos were naturally infected and were taken from diseased wheat crops in the Ruzyně and Jesenice localities near Praha. Larvae were reared under controlled conditions in a glasshouse from eggs that had been inserted into wheat plants. Four to six days after hatching some of the larvae were put on artificially diseased wheat for acquisition feeding. Following a period of fifteen days on the source of infection they were used for the infection experiments. The other group of larvae, which was not brought into contact with diseased wheat, was used as a standard as regards the effect of feeding on the plant. The imagos and both groups of larvae were then placed on healthy seedlings of the cereals for individual test feeding. Test feeding lasted 6—8 days in the individual passages. Silon cages, as already described (PRŮŠA, JERMOLJEV and VACKÉ 1959), were employed in order to prevent the insects from escaping.

In experiments on the transmission of the disease by means of aphids the species *Rhopalosiphon oxycanthae* (SCHRANK) and *Sitobium granarium* (KIRBY) Mordv. were used. Following two weeks' feeding on the diseased wheat, the aphids were transferred in groups of 5 to 10 to isolated healthy plants in silon cages. After 7 days the cages were removed from the plants and the aphids were killed with 0.05% solution of Systox. Forty two wheat plants were tested with *Rhopalosiphon oxycanthae* and 48 with *Sitobium granarium*.

Transmission of the virus by soil was investigated in an experiment in which grains from healthy plants were planted in 20 pots which had been filled with soil taken from the immediate vicinity of diseased wheat in the field. After the seedlings had sprouted, 10 were left in each flower pot and were observed up to the time of earing. The standard consisted of plants grown in soil taken from a field with root crops.

In order to examine the transmissibility of the virus by seeds, grains of diseased spring wheat var. Ruzyněská II were used. They were sown in wooden boxes containing soil which had been sterilised by steam. Out of 453 seeds sown, 304 seedlings sprouted and were observed up to the time of earing.

In the experiments on mechanical transmission sap expressed from diseased wheat plants was used. The preparation of the inoculum and the method of inoculation were similar to those used in the investigation of mechanical transmission of the wheat striate virus (PRŮŠA and VACKÉ 1960). A total of 112 wheat plants and 45 barley plants were inoculated.

The material tested in the infection experiments consisted of spring wheat var. Ruzyněská II and spring barley var. Stupický plnozrný at the vegetative stage of 1—2 leaves. In order to

exclude the possibility of any other infection, the experimental plants were placed in silon isolators.

Diseased plants of wheat and barley, both naturally and artificially infected, were subjected to cytological examination. The method used for this purpose was that which gave such remarkable results in the U.S.S.R. during the examination of plants suffering from the "zakukli-vaniye" virus of cereals and the Russian mosaic virus of winter wheat (СУКHOV 1940, 1942, GOLDIN 1954).

## Results and Discussion

Both natural and artificial infection by the leafhopper *Psammotettix alienus* gave rise to symptoms in spring wheat and spring barley identical with those in diseased plants of these cereals under field conditions. With insects caught at Ruzyně transmission was successful in a relatively high proportion of cases (Table 1). The percentage transmission fluctuated according to the different periods of test feeding. Two leafhoppers completely failed to transmit the disease (one died at the fourth, the other at the fifth passage). Out of six insects caught at Jesenice near Prague, only one caused infection. This difference in the infectivity of the vectors from the two localities may have been due to the fact that the extent of the disease differed in these places. At Ruzyně the insects were caught on wheat crops in which the proportion of dwarfed plants was higher than 50%, while at Jesenice diseased plants occurred as individuals at the edges of the field.

In experiments with leafhopper larvae infected on diseased wheat plants a remarkably high proportion of positive transmissions was obtained (Table 1). In the first test with 56 plants, 46 became diseased (82.1%), in the second out of 44 plants 32 were diseased (72.7%). Plants on which non-infected

Table 1. Results of Experiments with the Leafhopper *Psammotettix alienus*

Species of test plant		Wheat				Barley				Wheat	
		I		II		III		IV		V	
Test feeding		Plants		Plants		Plants		Plants		Plants	
		fed on	infected	fed on	infected	fed on	infected	fed on	infected	fed on	infected
Imagos from	Ruzyně	9	5	7	3	6	4	5	4	2	1
	Jesenice	6	1	6	1	5	1	—	—	—	—
Artificially infected larvae		56	46	44	32	—	—	—	—	—	—
Non-infected larvae		31	—	28	—	—	—	—	—	—	—

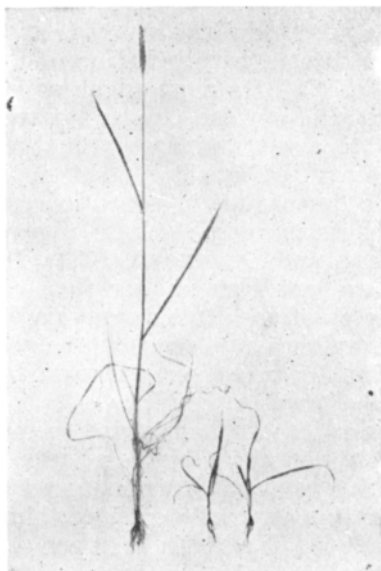


Fig. 1. Wheat infected with wheat dwarf virus; left a healthy plant. (Photo M. Novák.)

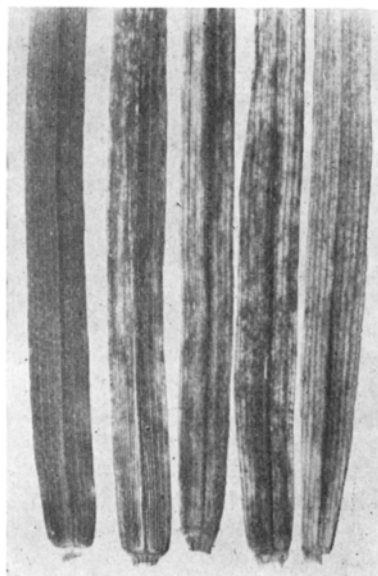


Fig. 2. Spots on wheat leaves affected by wheat dwarf virus disease; left, leaf from a healthy plant. (Photo M. Novák.)

larvae and non-infected adults of *Psammotettix alienus* were put to feed showed only a temporary retardation of growth. This provided proof of the virus nature of the disease under investigation.

The incubation period for the wheat dwarf virus was 14—15 days on the average for both wheat and barley under glasshouse conditions. It fluctuated, however, within the limits of 10 to 25 days.

The attempt to transmit wheat dwarf virus by means of the aphids *Rhopalosiphon oxycanthae* and *Sitobium granarium* was not successful. Experiments to transmit the virus by soil grains of wheat and by mechanical inoculation with sap from diseased plants also gave negative results.

Simultaneously with the etiological study of the wheat and barley disease, symptomatological observation was carried out on both naturally and artificially infected plants.

The most important symptom of wheat dwarf virus disease in the cereals examined is the marked dwarfing (Fig. 1); plants infected at the stage of 1—2 leaves, under glasshouse conditions, attained a height of only 10—15 cm., while the standard plants were over one metre (measured after the earing of healthy plants). Tillering was repressed in wheat and barley infected in the glasshouse. On the other hand, plants infected in the field showed a somewhat greater number of tillers than the healthy plants.

The ears of diseased plants under field conditions were very frequently barren, in many cases they did even emerge from the sheath. In so far as grains were formed in the ears, they were shrivelled.

The retardation of growth caused by the wheat dwarf virus was accompanied by colour symptoms. Fine spots and patches with indistinct outlines were irregularly scattered over the leaves (Fig. 2). At a more advanced stage of the disease the leaves went yellow, starting from the tips and margins. This yellowing later spread to the leaf sheath and stalk. At the final stage of the disease the plants began to go brown and to die off.

The cereal virus that has been determined during this investigation could, in view of its vector, be related to the Russian mosaic virus of winter wheat as known in the U.S.S.R. (ZAZHURILLO and SITNIKOWA 1939). It is, however, very probable that two viruses are not identical, for they differ from each other in many respects. The vector of the Russian mosaic virus of winter wheat (*Psammotettix striatus* L.) is of the same genus as the vector of our virus, but a different species. The differences in the symptoms produced in the host plants may be regarded as being of greater significance.

Lemon-yellow, sharply defined, conspicuous spots and stripes running parallel to the leaf veins develop on the leaves of wheat plants attacked by the Russian mosaic virus of winter wheat (SUKHOV and RAZVYAZKINA 1955). In the case of the present virus, however, only indistinctly outlined, blurred spots are to be found scattered over the whole leaf blade. Their colour varies from light green, dirty yellow to light brown.

As distinct from the Russian mosaic virus of winter wheat, which strongly stimulates tillering, the present virus either does not stimulate tillering at all, or only very slightly.

The fact that under the influence of acid fixation agents the crystals characteristic for the Russian mosaic virus of winter wheat (SUKHOV 1940, GOLDIN 1954) were not formed in the cells of chlorotic tissues of dwarfed wheat also points to a lack of identity between the two viruses. Not even by the preparation of herbarised material was it possible to produce crystal formation. DLABOLA (personal communication) also failed to find crystals in the alimentary tract of leafhoppers from the areas where the occurrence of the disease was most pronounced.

Finally, the behaviour of the viruses in their vectors is not similar. The larvae of *Psammotettix alienus* acquired a high ability of infecting healthy plants with the wheat dwarf virus by feeding on the source of infection. Larvae of *Psammotettix striatus*, however, only rarely transmit the Russian mosaic of winter wheat (GORLENKO 1951).

The facts that have been established indicate that we are probably dealing with a new virus which has not been previously described. It has been named wheat dwarf virus according to the most characteristic symptoms and the first plant identified as its host.

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Й. Вацке (Научно-исследовательский институт растениеводства, Чехословацкая академия сельскохозяйственных наук, Прага-Рузыне)

## Вирусная карликовость пшеницы

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В работе приводится доказательство вирусной природы карликовости пшеницы, распространенной в некоторых областях ЧССР. Вирус удалось перенести естественно инфицированными имаго цикадки *Psammotettix alienus* Dahlb. на яровую пшеницу и яровой ячмень. Оказалось также, что он в высоком проценте переносится личинками вылупившимися из яиц и искусственно инфицированными на источнике заражения. Опыт переноса вируса тлями *Rhopalosiphon oxycanthae* Schrank. и *Sitobium granarium* (Kirby) Mordv. дал негативные результаты. Также не удалось перенести вирус почвой, семенами и механической инокуляцией сока из больных растений.

Вирус карликовости пшеницы по своему переносчику может стоять ближе всего к вирусу русской мозаики озимой пшеницы. Однако, отличается от него симптомами, вызываемыми на злаках-хозяевах, другим переносчиком и отсутствием включений в цитоплазме инфицированных растений. Эти данные свидетельствуют о том, что мы имеем дело с новым, пока не описанным вирусом.