

## SEED-BORNE BEAN YELLOW MOSAIC VIRUS OF FABABEAN IN CANADA

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### Abstract

Seed lots of fababean (*Vicia faba*) obtained from several commercial sources in Canada were found to contain seed infected with a seed-borne virus. The virus was identified as a strain of bean yellow mosaic virus (BYMV). The BYMV was readily transmitted from infected to healthy fababeans by the cowpea aphid (*Aphis craccivora*). In a field trial of fababeans at Guelph natural spread of this virus by aphids from infected to healthy beans was first apparent in early July. At the end of August all of the field grown beans showed symptoms of BYMV. Seeds harvested in October and grown under greenhouse conditions from these plants indicated that on average 1.7% of the seed was infected with seed-borne BYMV.

### Introduction

In recent years fababeans, *Vicia faba* L., have assumed a new importance in Canada as a relatively high protein animal feed crop. A number of cultivars or strains of this bean are currently being tested for their yield and suitability at various locations, primarily in the Western provinces and in the Maritimes.

Fababeans were grown at Guelph for the purpose of rearing *Aphis craccivora* Koch, the cowpea aphid. Periodically virus-infected bean plants would develop from commercially obtained seed. In light of this fact and the possible future importance of fababeans in Canada a study was undertaken on this seed-borne virus.

### Materials and methods

#### Virus identification

Several lots of seed of the cultivars Broad Windsor and Longpod fababean were obtained from three commercial seed companies in Canada. In all instances 1 to 3 seedlings from each lot of 500 seeds of the two varieties grown under aphid-free greenhouse conditions developed a distinctive mosaic, (Figure 1), indicating a seed-borne virus.

In a host range study mechanical inoculations were made by dusting plant species with 400-mesh Carborundum and rubbing the leaves with a sterile cheesecloth pad dipped into tap water diluted juice from infected bean plants. A minimum of 10 individuals of each plant species was inoculated. After 3 weeks, all inoculated test plant species, irrespective of symptom expression, were checked for virus infection



Figure 1. Symptoms of bean yellow mosaic in fababean; top right and below, typical mosaic pattern in leaves; top left, healthy leaf.

by mechanical sub-inoculation onto pea seedlings, *Pisum sativum* L. cv. Alaska.

The cowpea aphid reared on virus-free fababeans was used in transmission studies.

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Nonviruliferous aphids were starved for 3-4 hours and transferred collectively in groups of 50 or more individuals to virus-infected fababean leaves. After an initial access period of 2 minutes aphids that appeared to be probing were transferred singly to healthy test plants. In all studies with aphids fababean seedlings were used as test plants. Nonviruliferous starved aphids transferred directly to healthy bean seedlings were used as controls.

The physical properties of the virus which included the thermal inactivation point, longevity in vitro and dilution end point were determined in accordance with the procedure suggested by Ross (1964) using Alaska pea seedlings for assaying infectivity.

Epidermal strips taken from the leaves of healthy and virus-infected plants were stained and examined for the presence of virus-induced inclusions according to a method described by Christie (1967). The virus particle morphology was determined by cutting up small pieces of infected or healthy leaf tissue in a few drops of 1% phosphotungstic acid neutralized to pH 6.8 with KOH and containing 0.025% bovine serum albumen. The resulting liquid was transferred to a Formvar coated specimen grid, excess fluid was removed with filter paper, and the specimen was examined with an electron microscope.

#### Transmission of virus under field conditions

In May of 1972 Broad Windsor fababean seeds were planted in 8 rows 40 m long running in a north to south direction. On June 1 when the beans were approximately 15 cm in height, plants showing virus symptoms were removed. The field plot was then divided into two 20-m halves. The plants in the northern half of the plot in an area where the prevailing wind is southwesterly were all mechanically inoculated with a virus isolate maintained under greenhouse conditions in Broad Windsor. Adjacent to the fababeans, single 40-m rows of Phaseolus vulgaris L. cultivars Bountiful and Dark Red Kidney, and pea cultivars Alaska and Thomas Laxton were planted in mid-June.

Seeds harvested from the fababeans were germinated during the winter of 1973 and checked for seed-borne virus.

## Results

### Virus identification

The following hosts were susceptible to the seed-borne virus from fababean: Chenopodium amaranticolor Coste & Reyn., and C. quinoa Willd. gave local lesions; Frenchbean (Phaseolus vulgaris) cv. Kentucky Wonder Wax, Kentucky Wonder: pea (Pisum

sativum) cv. Alaska, Thomas Laxton, and Crimson clover (Trifolium incarnatum L.) developed a severe systemic mosaic symptom following infection by the virus.

Nonsusceptible hosts were mustard (Brassica juncea Coss.) cv. Tendergreen, cucumber (Cucumis sativus L.) cv. Chicago Pickling, dimes violet (Hesperis matronalis L.), Nicotiana clevelandii Gray, N. rustica L., N. tabacum L. cv. Samsun NN, French bean (Phaseolus vulgaris) cv. Bountiful, Dark Red Kidney, Richgreen, Romano, Royalty, and Topcrop, pea (Pisum sativum) cv. Little Marvel, white clover (Trifolium repens L.), and cowpea (Vigna sinensis (L.) Endl.) cv. Black Local. Noninfection of the Phaseolus beans in the host range study indicates the probability that this is a pea mosaic strain of BYMV.

In aphid transmission tests 37 out of 100 cowpea aphids given 2 or more minutes access to an infected fababean leaf transmitted the virus to healthy fababean seedlings.

A study of physical properties revealed that the virus was inactivated at 59 C in crude sap taken from infected fababeans and was not infectious to Alaska pea at sap dilutions greater than 10<sup>-3</sup>. The virus remained infectious for 60 h but not for 72 h at 21 C in crude sap extracts.

Stained epidermal leaf strips taken from virus-infected fababean leaves and examined with a light microscope revealed the presence of amorphous cytoplasmic inclusion bodies similar to those reported by Bos (1969) and Evans (1969) for bean yellow mosaic virus (BYMV). Electron microscope examination of negatively stained leaf dip preparations revealed the presence of filamentous rods similar to those reported by others for BYMV (Brandes and Bercks 1965, Brandes and Wetter 1959, Taylor and Smith 1968).

### Transmission of virus under field conditions

In the field approximately 45% of the mechanically inoculated fababeans showed mosaic symptoms by July 1. These mechanically infected plants were tagged so that seed pods removed at harvest could be collected separately. Natural spread of virus by aphids in the field was first apparent on scattered plants by mid-July. At the end of July all plants in the mechanically inoculated half of the plot showed symptoms of virus infection. About 10% of fababean plants in the downwind noninoculated half of the plot showed evidence of virus infection. This information concurs with the general observation that aphid vectors of plant viruses move in the direction of the prevailing wind; healthy fababeans upwind of virus-infected plants become infected later on in the season than plants downwind (Swenson 1968). By late August all fababeans in the field plot were infected with virus. In the adjacent rows all of the Alaska peas

were virus-infected but no virus infection was apparent in Bountiful or Dark Red Kidney bean or Little Marvel pea. Only one species of aphid, *Aphis fabae* Scopoli, could be identified as colonizing the field-grown plants. Colonies of *A. fabae* never built up to more than a few hundred individuals on scattered plants apparently due to decimation of colonies by predaceous insects.

In early October fababean seed was collected from mechanically infected plants, from plants that showed virus infection from natural aphid transmission by late July, and from plants that did not show infection until late August. Seeds grown from mechanically infected plants resulted in 8 out of 454 or 1.8% of seedlings being infected with the virus. From the naturally infected plants 7 out of 510 or 1.4% showed evidence of virus in seed harvested from plants infected by late July, and 9 out of 463 plants or 1.9% had virus in seed harvested from those plants which had shown symptoms by late August. Thus, less than 2% of the seed taken from virus-infected plants was infected with seed-borne virus in this cultivar of fababean.

## Discussion

It is concluded from the above evidence that the seed-borne virus in fababean is a strain of BYMV. Its failure to infect cultivars of *Phaseolus* bean such as Bountiful and Dark Red Kidney formerly would have led to classifying this virus as a strain of the pea mosaic virus, but Box (1970) now regards this virus as a variant of BYMV. The resistance of Little Marvel pea to infection by the virus and the susceptibility of the Alaska and Thomas Laxton cultivars is in agreement with the reports of Corbett (1958) and Ford (1963) for BYMV.

Seed transmission of BYMV in fababean and other legumes has been demonstrated previously (Bos 1970, Corbett 1958). In several Middle Eastern countries this seed-borne virus presents serious problems in the cultivation of fababean (Izadpanah et al. 1969, Kaiser et al. 1967). In Iran, Kaiser (1972) showed that several pathogenic strains of this virus could be isolated from broad beans in different areas of that country. In a planting of 56 fababean types from 14 countries, he showed that BYMV was seed-borne in 80% of the lines with an incidence of 0.1% to 2.4%. In addition to BYMV, at least two other viruses are known to be seed-borne in fababean (Gibbs and Paul 1970, Gibbs and Smith 1970).

At Guelph it has been demonstrated that commercially available fababean in Canada may be infected with seed-borne BYMV. In the field trial natural spread of the virus occurred rapidly in July and August probably as a result of transmission by several species of aphid vectors (Kennedy et al. 1962).

Plantings of fababeans from virus-free seed could become infected with indigenous strains of BYMV from weed or crop sources such as naturally infected red clover and white sweet clover. This is a problem that will have to be looked into if fababeans are to become an established crop, but fababeans imported into Canada and those cultivars currently undergoing agronomic evaluation should be checked for the presence of seed-borne virus. Strains of BYMV present in imported seed lots might be far more pathogenic to fababean than isolates of the virus occurring naturally in North America. However, virus, if present in seed lots, can be controlled under field plot conditions if infected plants are rogued out as soon as symptoms become evident. If these infected plants, which never number much more than 2% of a planting, are removed by early June, then aphid transmission to healthy plants will be prevented and the cultivars freed from virus. This is due to the fact that the virus is totally dependent on the presence of aphid vectors for spread in the bean crop. In most locations in Canada little if any aphid build-up occurs before the end of June (Swenson 1968) and consequently spread of BYMV would not take place in the fababean crop before this time.

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