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IDENTIFICATION OF THE VIRUS CAUSING PAPAW YELLOW CRINKLE WITH TOMATO BIG BUD VIRUS BY TRANSMISSION TESTS

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SUMMARY

A virus from naturally infected papaw showing symptoms of yellow crinkle disease was transmitted by a dodder (Cuscuta australis) **to tomato, white clover and** Datura stramonium. **The resulting symptoms included big bud, phyllody and proliferation in tomato, and little-leaf and phyllody in clover.**

A virus originally obtained from a naturally infected tomato plant showing big bud symptoms produced typical yellow crinkle disease in papaws when transmitted by the same method.

Incubation times with dodder transfer varied from 7 to 13 weeks and with graft transmission from 3 to 6 weeks in the various hosts.

I. INTRODUCTION

Yellow crinkle disease of papaw (*Carica papaya* L.) was first recorded in Queensland by Morwood (1927, unpublished report). It is characterized by the harsh, crinkled appearance of leaves which mature during early stages of the disease, while the interveinal and marginal parts of the younger leaves are at first abnormally translucent before becoming necrotic and tattered as growth proceeds. After older leaves are shed there remains only a tuft of small chlorotic leaves on short petioles; these leaves have a spidery appearance caused by the loss of most laminal tissue except that close to the main veins. Floral parts may show well-developed phyllody (McKnight and Everist 1948).

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The disease was early suspected of being caused by virus infection (Morwood 1931, p. 47; Simmonds 1934, p. 69, 1937). McKnight (1949) suggested tomato big bud virus as the causal agent on the basis of the virescence symptoms and correlated its occurrence with associated field infections of other plant species by this virus. However, as in the case of many other records of diseases involving virescence symptoms, there was no experimental evidence to support this hypothesis.

To determine if the causal agent was the same as that responsible for the better documented diseases in other families, transmission to plants such as tomato and clover was necessary. This was accomplished by the use of a parasitic plant of the dodder type. Insect transmission was not attempted.

II. TRANSMISSION EXPERIMENTS

The results of the transmission experiment are summarized in Table 1.

TABLE 1

RESULTS OF DODDER TRANSMISSION TESTS WITH PAPAW YELLOW CRINKLE AND TOMATO BIG BUD VIRUS

Source Plant	Test Plant	Healthy Dodder		Infected Dodder		Time for Symptom Appearance (weeks)
		Total No. of Plants Parasitized	No. of Plants Infected	Total No. of Plants Parasitized	No. of Plants Infected	
Papaw ..	Tomato	3	0	3	2	7–9
Papaw ..	White clover ..	9	0	9	3	7–10
Papaw ..	*Datura*	3	0	3	1	9
Tomato ..	Papaw (field) ..	8	0	8	3	11–13
Tomato ..	Papaw (green-house)	8	0	8	4 ?	9–12

(a) **Papaw to Tomato, White Clover and** Datura stramonium

Seeds of *Cuscuta australis* R. Br. and lucerne were sown together and after germination a suitable parasitized plant was transplanted. This clone of dodder was maintained in a green-house by occasional transfer to fresh lucerne seedlings.

Small plastic vials of water were tied to the crown of a naturally infected papaw plant showing typical yellow crinkle symptoms. Sturdy sprigs of healthy dodder were placed with the cut ends in the water and the growing terminals gently pushed between the lobes of small crown leaves on the infected papaw. The parasite established itself in about 1 week and was left to grow for a further 4 weeks before removal of sprigs to vials of water suitably positioned to allow their subsequent establishment on small tomato, white clover and *Datura stramonium* L. plants in the green-house.

Healthy dodder from the original clone was colonized on an equal number of plants of each species.

It was found necessary to prune the dodder periodically to reduce excessive growth, particularly on the clover. After 8 weeks, the dodder was removed from all plants and persistent haustoria were excised with a scalpel or by similar means.

Of 3 tomato plants (cv. Grosse Lisse) successfully colonized by dodder from infected papaw, 2 subsequently developed symptoms identical with those caused by tomato big bud virus. One of 3 *Datura* plants and 3 of 9 white clover plants which were successfully colonized developed symptoms. The time taken for symptoms to appear on these hosts varied from 7 to 10 weeks after the dodder was established.

White clover plants infected with the papaw virus during this experiment showed a range of symptoms, including red leaves, little leaf and phyllody. Early symptoms showed as a red pigmentation of leaves as they matured, followed by marked reduction in size of subsequent leaves, some of which also showed the red pigmentation. Infected plants were severely stunted (Figure 1). A few flowers with green leafy petals of the typical phyllody type were produced but most flowers were normal.

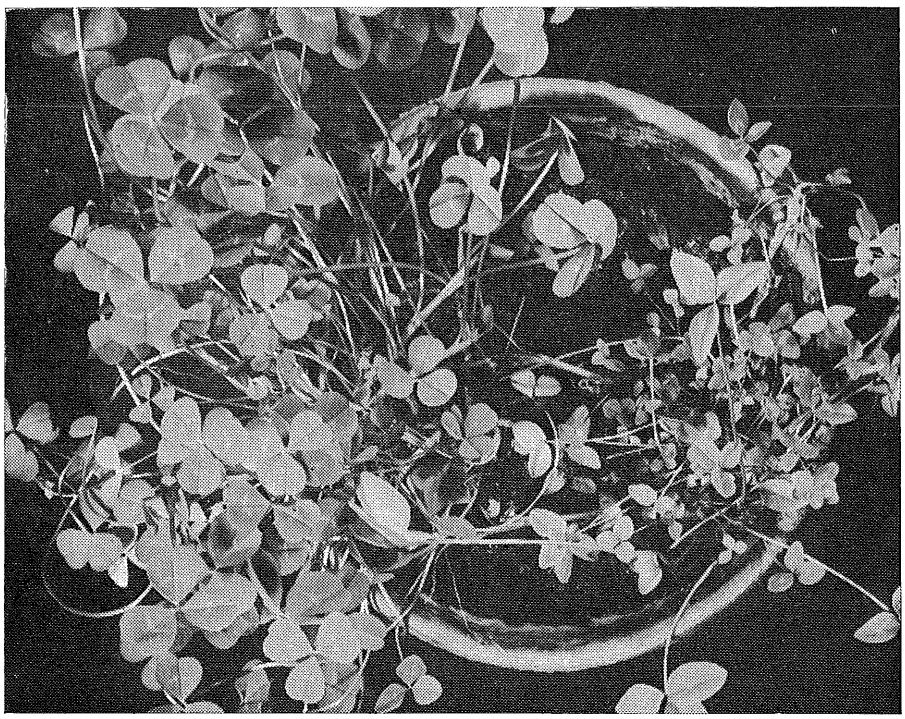

Fig. 1.—Right, white clover infected from papaw with yellow crinkle disease; left, healthy plant.

In tomato, slight swelling of the terminal bud and failure of young leaves to expand to a normal size were the first symptoms (Figure 2); they were followed by typical big bud and phyllody characteristics. Subsequent growth produced proliferation with many spindly axillary shoots in tight clusters. In *Datura,* the appearance of a few imperfect greenish flowers and interveinal chlorosis of the leaves was followed by cessation of flowering and production of small chlorotic leaves.

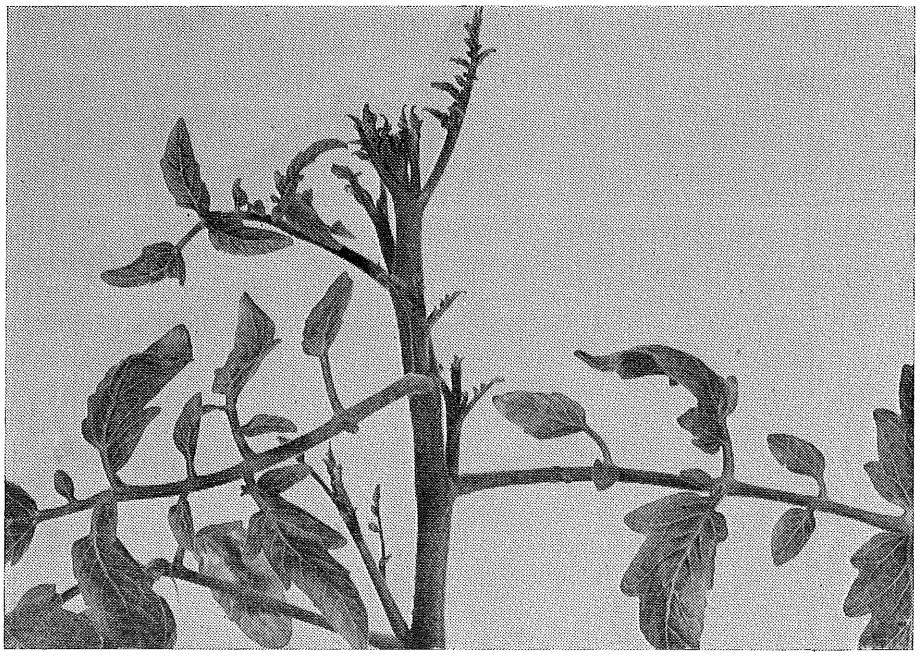

Fig. 2.—Early symptoms on tomato infected from papaw.

Attempts to transfer the virus from one papaw to another by grafting (bark patch with bud) were not successful, since infected grafts invariably became necrotic and died. Healthy material was comparatively easy to establish. However, graft transfers of the virus from tomato to tobacco, tomato and *Datura* were readily performed.

(b) **Tomato to Papaw**

In experiments to reproduce the disease in papaw in the glass-house, two sources of virus were used. In the first experiment the tomato plants infected above were recolonized by dodder, which was then established on papaw seedlings grown in pots. After a time approximating the expected latent period of 2-3 months, six papaw seedlings which had been colonized by infected dodder developed chlorotic symptoms; however, they soon succumbed to Pythium root rot. None of the eight control plants colonized by healthy dodder were affected.

In a second experiment involving both green-house and field inoculations, the source of virus was a field tomato plant showing typical big bud symptoms from which a graft transfer was made to a seedling tomato in the glass-house. This grafted plant was selected from similar ones after negative results were obtained from indexing for sap-transmissible viruses such as potato virus Y, cucumber mosaic virus, potato virus X, tomato mosaic virus and tomato spotted wilt virus. The plant was colonized with healthy dodder as in the earlier experiments, and after 6 weeks sturdy pieces of the parasite were used for infection purposes.

In the green-house, eight papaw seedlings approximately 18 in. high were each colonized by several sprigs of dodder from the infected tomato and four developed early yellow crinkle symptoms. All four of these, but none of eight uninoculated plants, subsequently died of root rot. A similar predisposition to root rot was not noticed with field infections by the virus.

In addition, a block of 200 papaw seedlings was established in the field, and when they were approximately 2 ft high, 16 adjacent plants were selected from these. Alternate plants were colonized by dodder from the same tomato plant as described above and the remaining eight by uninfected dodder. After 11 weeks, two papaw plants colonized by dodder from the infected tomato developed typical early symptoms of yellow crinkle disease and a third plant showed symptoms shortly after (Figure 3). The disease then continued to

Fig. 3.—Early symptoms on papaw infected from tomato.

develop within the normal range of symptom variation found in natural infections. None of the plants colonized by uninfected dodder, nor any of the remainder of the original block of 200 plants, developed similar symptoms.

III. DISCUSSION

In Great Britain several workers (e.g. Carr 1960; Davies 1964) have distinguished between clover phyllody and clover red leaf (potato stolbur?) viruses. Tomato big bud virus is also regarded as distinct. Several Australian workers (Hill 1943; Hutton and Grylls 1955; Helms 1962) have found that the same virus which causes phyllody and little-leaf in clover also produces big bud symptoms in tomato.

During the development of the disease in the plants infected in these experiments, a sequence of symptoms was noted which could possibly be explained as the result of infection by more than one virus component. However, it appears equally possible that these are only different expressions of symptoms during the course of a disease caused by a single virus. Further experiments with leafhopper transmission may be justified to clarify the position in Australia.

Posnette and Ellenberger (1963) concluded that tomato is an unsatisfactory host for distinguishing between some viruses of this type, due to the similarity of some of the symptoms produced. However, although many species of plants showing symptoms attributed to tomato big bud virus were available in Queensland, a source from a Solanaceous host was preferable because of the ease of grafting to related genera. This facilitated the maintenance of the source and comparison of symptoms among several other species in this family. Attempts were made to obtain tomato big bud virus from purple top wilt of potato, which has been attributed in Australia to this virus (Norris 1954). However, during the course of these experiments, a number of grafts to tomato from potatoes with symptoms of this disease (after rejecting those giving positive tests for sap-transmissible viruses) resulted in a yellows type of disease sometimes lethal to tomato and not typical of normal big bud disease.

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