SPECIAL ARTICLES

TRANSMISSION OF POTATO WITCHES' BROOM TO TOMATOES AND POTATOES

(Preliminary Report)

A. TRANSMISSION FROM POTATOES TO TOMATOES

In the greenhouse of the Montana Agricultural Experiment Station at Bozeman, Montana, stems of 18 Earliana tomato plants 20 to 75 cm. tall were inarch-grafted on to the stems of 18 potato plants which showed severe symptoms of witches' broom. The potatoes were grown from seed pieces inoculated with tuber plugs from potato tubers infected with witches' broom. These potato plants were selected because they developed large stems before the witches' broom appeared. Seven of these 18 grafted tomato plants showed definite symptoms of a peculiar disease within 53 days after grafting. Two of the other grafted tomato plants exhibited the disease symptoms within 73 days after grafting. The top of one plant became chlorotic within 13 days, and another within 30 days after grafting. Of the nine grafts which transmitted the disease, five were known to be true grafts, and graft unions probably occurred also in the others. The witches' broom potato plants which transmitted the disease to the tomatoes were of the following varieties: Bliss Triumph, Russet Burbank, Perfect Peachblow, Blue Victor and Rural New Yorker.

The other nine grafted tomato plants of the above series, and 15 ungrafted tomato plants of the same age and variety showed no symptoms of the disease. Two of the checks were in the same pots with tomatoes which developed the disease.

The symptoms of the disease transmitted from witches' broom potato plants to tomato plants were: The new leaflets had very prominent, yellow-chlorotic margins; they were extremely dwarfed and often rugose. In a small percentage of the leaflets, the lower sides of the veins were purple. Many of the new leaflets had very narrow leaf blades. None of the leaflets was upward rolled. In many cases the rhachises of the leaves were prominently downward curled. The new tops of the affected plants were very chlorotic. They bloomed profusely and produced many small tomatoes; the buds and flowers developed normally.

Western yellow tomato blight and linear and rugose tomato mosaics have some of the symptoms described above. Since the disease considered here does not have enough of the diagnostic symptoms of any of these three tomato viroses for classification in one of them, and because the symptoms are very similar to the first symptoms of witches' broom in the potatoes from which the disease was transmitted, this disease is tentatively called tomato witches' broom. This is probably the first report of the transmission of the virus of potato witches' broom to a different host.

B. TRANSMISSION TO VARIOUS POTATO VARIETIES

Potato witches' broom was shown by Young and Morris¹ to be transmissible from diseased to healthy potatoes. The primary and secondary symptoms of the disease were well described by Hungerford and Dana.²

Seed pieces of rogued stocks of several varieties of potatoes were inoculated with witches' broom by inserting cork borer plugs from witches' broom tubers into holes made in the seed pieces. Thirty-four of the resulting plants developed the first symptoms of witches' broom within 35 to 114 days after inoculation. These tuber plugs formed graft unions with the seed pieces in many cases.

The following data include the cases of transmission previously reported and summarize the data secured: Of the 138 seed pieces plugged with witches' broom scions, 45 produced plants with typical symptoms of witches' broom. The progeny of two other plants showed severe symptoms of witches' broom although the parents exhibited no symptoms of disease. From tuber scions of Bliss Triumph and Jersey Peachblow potatoes, witches' broom was transmitted to Bliss Triumph, Russet Burbank, Perfect Peachblow, Idaho Rural, Green Mountain, Irish Cobbler, Blue Victor, Six Weeks, Mills' Prize, Colorado Pearl and Rural New Yorker potato plants.

Similar tuber plug inoculations failed to transmit the disease to Norwegian Yam, Early Michigan, Early Rose, Producer and Up-to-date potato plants. Russet Burbank and Rural New Yorker plants affected with witches' broom did not exhibit marginal chlorosis of the leaflets. The other nine varieties affected with this disease showed prominent marginal chlorosis, at least in the early stages of the disease.

Forty-five normal seed pieces of Bliss Triumph potatoes, plugged with scions from healthy tubers of this variety, were planted in the greenhouse where they grew for 84 days. None of them developed any symptoms of viroses (virus diseases). Besides these, four Bliss Triumph seed pieces were plugged with scions from Irish Cobbler and Netted Gem tubers,

¹ Young, P. A., and H. E. Morris. "Potato Witches" Broom is a Transmissible Disease," U. S. D. A. Plant Disease Reporter 10 (3): 26-28, 1926.

² Hungerford, C. W., and B. F. Dana. "Witches' Broom of Potatoes in the Northwest," Phytopath. 14: 372-383, 1924. and three Netted Gem and two Irish Cobbler seed pieces were plugged with scions from healthy Bliss Triumph tubers. None of these seed pieces plugged with scions from healthy tubers developed plants with disease symptoms. About 4,000 potato plants uninoculated with witches' broom were closely observed while they developed to maturity. Only one of them developed witches' broom symptoms, and it very probably represented late infection of the parent plant in the field. Hundreds of tubers produced by potato plants affected with witches' broom were grown in the field and the greenhouse; only one of the resulting plants failed to develop witches' broom symptoms.

Mealy bugs (Coccidae) were colonized on potato vines severely affected with witches' broom, and then transferred to four healthy Bliss Triumph potato plants and two healthy Earliana tomato plants; no evidence of disease transmission was seen. Aphids taken from potato vines with severe witches' broom were allowed to feed on the sprouts of two Irish Cobbler potatoes. Neither the resulting plants nor their progeny produced vines with witches' broom symptoms. Seven other attempts to transmit witches' broom by means of aphids all failed. In the field in 1925 and 1926, normal Bliss Triumph potato plants grew beside potato plants severely affected with witches' broom. Since aphids were abundant on both the normal and the diseased potato plants late in the season, tubers from eight of the normal plants were planted in the greenhouse at different times. All the plants grown from these tubers remained healthy, showing that aphids had not carried the disease to their parents from the adjacent vines which had witches' broom.

By leaf mutilations 62 Bliss Triumph, Netted Gem and Irish Cobbler potato plants 12 to 30 cm. tall were inoculated with witches' broom. The inoculations were repeated on many of the plants two or three times, at intervals of a few days. These experiments were performed in the greenhouse during two winters. None of the inoculated plants developed witches' broom symptoms. Eighteen normal potato plants were grown in pots with plants severely affected with witches' broom, but none of the normal plants became diseased. The method of field dissemination of witches' broom is not yet determined.

Most of the witches' broom plants seen in many fields have shown severe symptoms so that the disease was recognizable soon after the sprouts appeared. Tubers infected with witches' broom produce many chlorotic, spindly sprouts. There now appears to be no constant difference between the upright and prostrate forms of witches' broom in the field. In one row of 98 hills of Bliss Triumph potatoes, 56 of the hills were severely affected with witches' broom. The seed pieces weighed about 50 g. apiece and were quarters of tubers. The large size of the seed tubers indicated that they very probably came from parent vines infected late in the season. Plants infected early in the season rarely produce tubers of marketable size.

In the greenhouse the development of a potato plant successfully inoculated with witches' broom by tuber grafting is as follows: The plugged seed piece produces a plant which remains normal in growth and appearance for 31 to 114 days. If the mosaic virus be present in either the stock or the scion, its symptoms appear within the first month or two and precede the witches' broom symptoms. The presence of mild or crinkle mosaic does not interfere with the development of witches' broom. The first symptom of witches' broom consists of an increasingly prominent chlorosis of the new leaflets on one or more stems. In most varieties of potatoes the margins of the new leaflets are chlorotic yellow. Red or purple colors often occur in the new leaflets, particularly under field conditions. The top of the plant then elongates with abnormal rapidity, and produces a stem which is cylindrical with enlarged nodes; it is chlorotic and often purpled.

Many chlorotic, spindly axillary branches develop all along the stem and bear typical witches' broom leaves. The abnormally numerous axillary branches, sometimes borne by plants with severe spindle tuber, are thick, green, and not spindly, so they are unlike those borne on witches' broom plants. Filamentous aerial stems are valuable in diagnosis when they occur; they are often numerous. Small numbers of these peculiar stems occur in the field.

Spindly sprouts appear as aerial or subterranean stem branches at the base of the plant and grow very slowly, representing witches' broom in severe form. The tubers already formed often sprout and add to the number of spindly basal sprouts. Vigorous plants may have 200 or more very slender stems, and a large number of very small tubers. In most cases the top of the plant continues to grow rather rapidly for the next few months and may reach a height of 1 m. Such plants frequently bloom earlier and more profusely than normal plants. The old leaflets produced while the plant was normal slowly die and are replaced by the dwarfed, chlorotic leaves produced by the spindly tops and branches. The dwarfed, new branches often bear only minute, simple leaves. Filamentous aerial stems and aerial tubers commonly occur on all parts of the stems. Flowers were borne on the aerial tubers of a few plants, but this is an uncommon symptom.

Finally, the tall main stems die completing the transformation of the normal potato plant into a

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plant severely affected with witches' broom. Thus, the primary and secondary symptoms of witches' broom intergrade. All that remains is the group of spindly basal sprouts only 5 to 30 cm. tall which, with proper care, remain alive for several months longer. If kept for eight or more months, the old stems successively die and are replaced by new sprouts and their branches which continue to arise from the tubers in or near the surface of the soil.

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STUDIES ON THE GOLGI APPARATUS OF THE MAMMARY GLAND

THE present study is a corroboration and extension of the work of Da Fano on the Golgi apparatus in different physiological conditions of the mammary glands. The primary objective of this work was to find if any evidence could be obtained as to the supposed transformation of the Golgi apparatus into a granular material that would be extruded from the cells together with the products of their activity.

It was found by using Da Fano's cobalt-silvernitrate method that the epithelial cells of the mammary glands of rats during the later stages of pregnancy and full activity show a great hypertrophy of the apparatus together with a transformation into a reticular and granular-like structure. During lactation a part of the apparatus migrates from near the nucleus and spreads throughout the cytoplasm of the cell, concentrating at its sides in the direction of the lumen.

By use of Brouha's modified Flemming fixative it was found that the Golgi bodies could be demonstrated with ease in the lumen of the actively secreting gland —a condition unknown to Da Fano. The technique used to demonstrate the Golgi bodies in the lumen is as follows:

(1) Pieces of mammary glands of the white rat were fixed for two days in the modified Flemming fixative, which consists of:

Sol. A. Saturated sol. of Corrosive sub-

	limate	600	gr.
Sol. B.	Glacial acetic acid	4 0	gr.
	Osmic acid	1	gr.
	Chromic acid	1	gr.
	Distilled water	100	gr.
			-

To four parts of solution A add one part of solution B.

(2) The tissue is then washed in water from one half to one hour.

(3) Dehydrate, clear in xylol and embed in paraffin.

(4) Section from 3 to 5 microns and fix sections on slides by the albumen method.

(5) Transfer sections through xylol and decreasing strengths of alcohols into distilled water.

(6) Sections are then placed in 0.2 per cent. solution of gold chloride plus 1 drop of acetic acid to every 10 cc. of solution.

(7) Wash slides in distilled water and place in 5 per cent. solution of sodium hyposulphite for two minutes and wash over again in distilled water.

(8) Dehydrate and clear in xylol.

(9) Mount in balsam.

Slides prepared by the above described technique shows the Golgi apparatus in the lumen in the form of small round bodies. These bodies are apparently located in the presumably cytoplasmic layer surrounding the fat droplet which is derived from the cytoplasm of the secreting epithelium. When the fat is dissolved the Golgi apparatus remains as granularlike bodies marking the outer limiting membrane of the droplet. Large numbers of these figures may appear in one lumen which, before the fat is dissolved, presents a deeply stained mass varying from approximately 2 to 15 microns in diameter. In some cells which were fixed just before extrusion of the fat droplet the Golgi bodies may be seen in the bulging limiting membrane of the cell describing a convex arc into the lumen of the gland.

It is also possible to demonstrate the Golgi apparatus in both the cell, and in the lumen of the actively secreting gland by use of Lundford's modified osmic acid method, although it is much more difficult to dissolve the fat from within the droplet after such long osmic acid impregnation. However, the Golgi bodies may be seen quite clearly in the peripheral layer surrounding the fat droplet before it is extruded from the cell and also in the lumen of the active secreting gland.

It would seem from this evidence that the Golgi bodies play a part in the phenomena of secretion. However, the exact part they play in this process is yet imperfectly known. That they are reformed within the cell after extrusion with the secretory products from a fragmentation of the remaining apparatus would seem the most plausible explanation from the evidence at hand. These observations would likewise suggest that the Golgi apparatus is a definite structure capable of a morphological existence without its cellular environment.

These studies are being continued.

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