Relationship of Turnip Mosaic Virus Susceptibility and Downy Mildew (*Bremia lactucae*) Resistance in Lettuce[1]

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Abstract. A mosaic disease of *Lactuca sativa* L. is described and the causal agent identified as turnip mosaic virus (TuMV). Extensive infection reduces the yield appreciably or may destroy entirely the value of the crop. A survey of *L. sativa* cultivars indicated that TuMV susceptibility is restricted to mildew resistant: crisphead types: 'Calmar', 'E-4', 'Imperial 410', 'Imperial Triumph', 'Valrio', 'Valtemp', and 'Valverde'. Circumstantial evidence indicates that TuMV susceptibility in cv. 'Calmar', 'Imperial 410', 'Valrio', 'Valtemp', and 'Valverde' stems from the mildew resistant *L. serriola* L. (P.I. 91532). TuMV and mildew resistant cultivars are: butterhead type 'May King', 'Meikoningin', 'Proeftuin's Blackpool', 'Ventura'; leaf type 'Red Salad Bowl', 'Salad Trim'; cos type 'Valmaine'. Seed collections of *L. serriola* from the Santa Clara and Salinas Valleys of California produced plants that fell into 3 classifications: a) TuMV-resistant, mildew-resistant; b) TuMV-resistant, mildew-susceptible, and c) TuMV-susceptible, mildew-resistant. No plants in *L. sativa* or *L. serriola* were susceptible to both TuMV and mildew. Extreme resistance to TuMV was demontrated in *L. sativa* and *L. serriola.* TuMV-susceptible *L. sativa* cultivars showed differences in tolerance to symptom expression and resistance to infection. In *L. serriola* a resistance connected with a hypersensitivity reaction was observed. All isolates of TuMV collected were capable of infecting susceptible *L. sativa* cultivars. *L. sativa* cv. 'Calmar' and 'Valverde' systemically infected with TuMV did not transmit the virus through the seed. *L. serriola* systemically infected with TuMV failed to produce seed.

Introduction

In 1966, a mosaic disease of lettuce, *Lactuca sativa* L., was observed in the Salinas Valley of California. It appeared to be restricted to the crisp-headed downy mildew resistant cultivar 'Calmar' (10) for it was not found in commercial plantings of 'Great Lakes' and 'Parris Island Cos'. The importance of 'Calmar', more than 25,000 acres planted annually in the central coastal districts, and the destructiveness of the disease led to investigation of the biological and physical characteristics of the causal virus. These investigations indicated that the causal virus, turnip mosaic virus (TuMV), has a close association with resistance in lettuce to downy mildew incited by *Bremia lactucate* Reg.

Materials and Methods

Nonviruliferous green peach aphids, *Myzus persicae* (Sulz.), were reared on radish, *Raphanus sativus* L. Other species of aphids used were reared on appropriate host plants within an insectary. Transfers of single insects were made with a moistened camel's-hair brush.

Host range studies were carried out by 2 methods. Recovery tests from field plants showing mosaic symptoms indicated some of the species to be naturally infected with TuMV. The virus was recovered by feeding nonviruliferous green peach aphids on collected field plants for 24 hr and transferring the insects to healthy indicator seedlings for 24 hr. Additional host-range studies were carried out by inoculation of at least 10 seedlings of a number of different species in an insectary compartment with viruliferous green peach aphids fed on TuMV-diseased shepherd's purse, *Capsella bursa-pastoris* (L.) Medic., for 24 hr. Recovery attempts from all inoculated plants were carried out to verify susceptibility. After tests with aphids, all plants were sprayed with nicotine sulfate and placed in greenhouses which were also fumigated at weekly intervals with nicotine sulfate.

Mechanical inoculation was by the common carborundum technique. Inoculum was prepared by grinding 1 part diseased tissue in about 5 parts of 0.02 *M* phosphate buffer, pH 7, containing 0.02 *M* sodium sulfite.

The local-lesion test plants used in assessing the effect of various treatments on virus infectivity were *Chenopodium amaranticolor* Coste and Reyn. or *C. quinoa* L. Most of the property tests were conducted with randomized-block or Latin-square designs on whole or half leaves of the test plants.

Downy mildew, *Bremia lactucae* Reg., inoculum was collected from commercial lettuce fields in the central coastal districts. Seed for plants to be scored for resistance was planted in flats or placed on moist filter paper in petri dishes. The young seedlings (cotyledon stage) were inoculated with a water suspension of spores of the fungus and tested by methods described by Rodenburg (8). To maintain an available supply of mildew for inoculation, cultures of mildew were maintained continuously on 'Great Lakes 118'.

An antiserum against crude plant extracts was prepared from shepherd's purse plants showing typical symptoms induced by the original TuMV isolate from field lettuce. Plants were ground in a mortar in the presence of ½ volume 0.02 *M* phosphate buffer, pH 7, containing 0.02 *M* sodium sulfite. Crude extracts were clarified by low-speed centrifugation (10 minutes at 4220 *g*). Antiserum was prepared from the clarified crude extracts by 6 intramuscular injections of a rabbit at weekly intervals, using Freund's complete adjuvant (Difco Bacto).

Results

Symptoms on L. sativa. Susceptible lettuce cultivars of *L. sativa* mechanically or aphid-inoculated in the greenhouse developed symptoms similar to those observed on 'Calmar' exposed to natural infection. Early symptoms consist of numerous small light green circular to irregular lesions, distributed at random between and adjacent to the veins. Within a few days, chlorotic lesions on seedling lettuce become more numerous, and nearly replace the normal dark-green tissue, imparting a distinctly chlorotic color and a coarse mottle. Infection is often accompanied

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by curvature of the midrib and asymmetrical distortion of the leaf blade. The virus causes severe stunting of young lettuce and is occasionally lethal. During the reproductive phase of growth, necrotic lesions may develop on the seed stalk. Younger leaves have a yellow green mottle, and their margins frequently show considerable necrosis. Necrotic areas may form on the developing involucral bracts and peduncles, and many of the floral heads wither before they are able to mature. Both numbers of seed per flower head and seed production per plant are extremely low.

The disease is readily distinguished from lettuce mosaic by the presence of numerous circular to irregular lesions, by the absence of the downward roll of the leaf tips and by the dull yellow color of lettuce mosaic infected plants.

Host range. Plants found susceptible to TuMV are listed in alphabetical order. Species in which the virus was not fully systemic are marked with an asterisk (*).

AIZOACEAE. *Tetragonia expansa* Murr.

BORAGINACEAE. *Amsinckia douglasiana* DC.

CARYOPHYLLACEAE. *Spergula arvensis* L.

CHENOPODIACEAE. **Beta macrocarpa* Guss., **Chenopodium amaranticolor* Costa & Reyn., **C. capitatum* (L.) Asch., *C. quinoa* L., *C. urbicum* L., *Spinacia oleraceae* L. cv. 'Califlay'.

COMPOSITAE. *Cichorium endivia* L. cv. 'Broad Batavian Full Hearted' (escarolle), 'Green Curled'; *C. intybus* L. cv. 'Asparagus'; **Cynara scolymus* L.; *Lactuca sativa* L. cv. 'Calmar', 'Imperial 410', 'E-4', 'Imperial Triumph', 'Valrio', 'Valtemp', 'Valverde'; *L. serriola* L.; *Senecio vulgaris* L.; **Sonchus oleraceus* L.

CRUCIFERAE. *Brassica adpressa* Boiss.; *B. campestris* L.; *B. campestris* var. *napobrassica* (L.) DC. cv. 'American Purple Top'; *B. hirta* Moench; *B. juncea* (L.) Coss; *B. juncea* var. *crispfolia* Bailey cv. 'Southern Giant', 'Curled Long Standing'; B. kaber (DC.) L. C. Wheeler; *B. nigra* (L.) Koch; *B. oleracea* L. var. *botrytis* L. cv. 'Snowball Y', 'November-December'; *B. oleracea* var. *capitata* L. cv. 'Copenhagen Market Early'; *B. oleraceae* var. *gemmifera* DC. cv. 'Jade Cross'; *B. oleracea* var. *gongylodes* L. cv. 'Early White Vienna'; *B. oleracea* L. var. *italica* cv. 'Topper 43'; *B. oleracea* var. *viridis* L. cv. 'Dwarf Blue Curled' Vate's strain; *B. pekinensis* (Lour.) Rupr. cv. 'Michihli'; *B. rapa* L. cv. 'Purple Top White Globe'; *Capsella bursa-pastoris* (L.) Medic.; *Erysimum asperum* DC.; *Matthiola incana* (L.) R. Br.; *Nasturtium officinale* R. Br.; *Raphanus sativus* L. cv. 'White Icicle', 'Comet'; *R. raphanistrum* L.; *Sisymbrium irio* (L.) Britt.; *Thlaspi arvense* L.

GERANICEAE. *Geranium dissectum* L.

LEGUMINOSAE. *Phaseolus vulgaris* L. cv. 'Small White', 'Bountiful'.

MALVACEAE. **Gossypium hirsutum* L.

PAPAVERACEAE. **Eschscholtzia californica* Charm., *Papaver* somniferum L., P. rhoeas L. cv. 'Shirley'.

PORTULACACEAE. *Claytonia perfoliata* Donn.

SOLANACEAE. *Nicotiana clevelandii* Gray; **N. glutinosa* L.; *N. megalosiphon* Heurck & Meull.; **N. tabacum* L. cv. 'Turkish', 'Xanthi-nc'; *Physalis floridana* Rybd.; *P. ixocarpa* Brot.; *P. wrightii* Gray.

UMBELLIFERAE. *Conium maculatum* L.

Virus properties: Properties of the original isolate from field lettuce were determined by mechanical inoculation of extracts from lettuce, turnip, *Brassica adpressa,* and *Nicotiana clevelandii* to *Chenopodium amaranticolor,* and *C. quinoa* (local-lesion hosts) and shepherd's purse (systemic host).

1) *Thermal inactivation.* Extracts were heated in a water bath for 10 minutes at 45, 50, 55, 60, 65, and 70°C. Infectivity was greatly reduced at 55°, and was not detected after treatment at 60°.

2) *Tolerance of dilution.* Preparations used in dilution tests were extracted in the presence of buffer in the proportion of 1 part plant tissue to 5 parts diluent, and then diluted with the same buffer in series to 5^{-5}. Infectivity was low at dilutions of 5^{-4}, and was not found at 5^{-5}.

3) *Longevity in vitro.* Virus extracts *in vitro* lost activity rapidly at room temperature. Only a trace of activity remained in preparations aged for 48 hr.

4) *Serology.* An antiserum against crude plant extracts was prepared with a precipitin titer of 1/64. Precipitin tests were conducted with sap clarified by centrifugation and treatment with butanol (9). Precipitin tests with the same clarified sap were positive with antiserum (supplied by R. J. Shepherd) prepared against a Wisconsin isolate of the turnip mosaic virus.

5) *Electron microscopy.* Electron microscopy was done by Dr. A. H. Gold, University of California, Berkeley. Crude samples from lettuce, turnip, and *Nicotiana clevelandii* showed fairly numerous rods, about 800 mu long, and no other characteristic particles.

Transmission tests: 1) *Mechanical.* The virus isolate from field lettuce was readily transmitted by routine mechanical inoculation techniques. The efficiency of transmission in 239 shepherd's purse plants was 69%, in 173 *L. sativa* cv. 'Calmar' plants 38%, and in 174 cv. 'Valverde' 67%.

2) *Seed.* Seeds of each of 9 plants of *L. sativa* cv. 'Calmar' systemically infected with TuMV and 7 plants of cv. 'Valverde' were harvested separately and checked for rate of virus transmission. No seed transmission was observed in tests of 1688 'Calmar' and 1745 'Valverde' seedlings.

The seed produced by 5 *Brassica adpressa* plants systemically infected with TuMV was bulked, and indexed for seed-borne infection. No seed transmission was found in 784 seedlings indexed.

Ten systemically infected *L. serriola* plants from collection No. 8 and No. 14 (Table 2) were grown for seed production. They failed to produce seed although they flowered over a 2-month period.

3) *Dodder.* Transmission tests were made with 2 species of dodder, *Cuscuta californica* Choisy and *C. campestris* Yuncker. Dodder was established on diseased plants of *L. sativa* cv. 'Calmar', and stems of the parasite were trained to 'Calmar' and 'Great Lakes 118'. At least 15 healthy plants of each cultivar were parasitized by each species of dodder but neither cultivar developed symptoms of the disease.

4) *Insect.* To determine whether some of the common aphid species were vectors of the virus, tests were conducted with shepherd's purse used as both source and test plants. Various species of nonviruliferous aphids were placed on virus source plants for about 24 hr; then approximately 25 individuals were transferred to each of a number of test plants for an infection feeding period of about 24 hr. Under these conditions, the following aphid species transmitted the virus: *Brevicoryne brassicae* (L.), *Macrosiphum euphorbiae* (Thomas), and *Myzus persicae* (Sulz.).

Virus-vector relationships: Although several aphid species are capable of transmitting TuMV, virus-vector relationships were studied only with the green peach aphid, *M. persicae.*

1) *Transmission efficiency.* Nonviruliferous green peach aphids, after a 24-hr acquisition feeding on diseased shepherd's purse plants, were transferred singly to healthy seedlings not only of this species but also of *L. sativa* cv. 'Calmar' and 'Valverde'. The insects were allowed a 24-hr feeding period on the test plants and then killed with a nicotine spray. The efficiency of transmission in 60 shepherd's purse plants was 22.7%, in 148 'Calmar' plants 9.5%, and in 146 'Valverde' plants 25.3%.

2) *Persistence.* Retention of the virus by the green peach aphid was determined by daily serial transfers to healthy shepherd's purse seedlings. Viruliferous aphids transmitted the virus to healthy plants only during the first day.

Table 1. Susceptibility of *L. sativa* cultivars to infection by TuMV and *Bremia lactucae.*

Cultivar and type	Plants inoculated with TuMV	Plants susceptible to TuMV[a]	Downy-mildew reading[b]
	(number)	(number)	(R = resistant) (S = susceptible)
Crisphead type			
Calmar	215	215	R
Carvan	10	0	S
Climax	10	0	S
Cosberg	20	0	S
E-4	70	70	R
Francisco	10	0	S
Forty-Niner	10	0	S
Fulton	10	0	S
Golden State A	10	0	S
Golden State B	10	0	S
Golden State D	10	0	S
Great Lakes A-36	10	0	S
Great Lakes 54	10	0	S
Great Lakes 65	10	0	S
Great Lakes 66	10	0	S
Great Lakes 118	230	0	S
Great Lakes R-200	10	0	S
Great Lakes 366	10	0	S
Great Lakes 407	10	0	S
Great Lakes 659	10	0	S
Great Lakes 6238	10	0	S
Green Bay	10	0	S
Greenland	10	0	S
Imperial D	30	0	S
Imperial F	20	0	S
Imperial 17	20	0	S
Imperial 44	20	0	S
Imperial 101	20	0	S
Imperial 152	30	0	S
Imperial 615	20	0	S
Imperial 410	80	80	R
Imperial 456	19	0	S
Imperial 847	30	0	S
Imperial 850	20	0	S
Imperial Triumph	70	70	R
Merit	10	0	S
Mineto	10	0	S
New York 12	20	0	S
New York 515	20	0	S
Oswego	10	0	S
Penlake	10	0	S
Progress	10	0	S
Spartan Lakes	20	0	S
Vanguard	10	0	S
Valrio	60	60	R
Valtemp	60	60	R
Valverde	115	115	R
Butterhead type			
Bibb	20	0	S
Big Boston	17	0	S
Blondine	20	0	S
Buttercrunch	10	0	S
Dark Green Boston	10	0	S
Dark Green Mignonette	10	0	S
Matchless	10	0	S
May King	40	0	R
Meikoningin	40	0	R
Midas	10	0	S
Neckarreuzen	20	0	S
Proeftuin's Blackpool	40	0	R
Tom Thumb	10	0	S
Ventura	40	0	R
Wonder van Voorburg	20	0	S
Leaf type			
Black Seeded Simpson	20	0	S
Grand Rapids	40	0	S
Oak Leaf	10	0	S
Prize Head	20	0	S
Red Salad Bowl	40	0	R
Ruby	10	0	S
Salad Bowl	20	0	S
Salad Trim	40	0	R
Slowbolt	10	0	S
Cos type			
Dark Green Cos	20	0	S
Ghaffari	20	0	S
Parris Island Cos	20	0	S
Valmaine	40	0	R
Latin type			
Fordhook	20	0	S
Gallega	20	0	S
Stem type			
Celtuce	10	0	S
Chinese	10	0	S

[a]Systemic infection. TuMV was recovered from susceptible cultivars, but not from resistant cultivars.
[b]*Bremia lactucae* collection from the central coastal districts of California.

Prevalence of lettuce affecting TuMV isolates. To determine whether isolates capable of causing severe disease symptoms in susceptible lettuce cultivars were widespread or restricted in distribution, a limited survey was conducted. Susceptible cultivars of lettuce were infected by 52 field isolates of TuMV in *Brassica adpressa, B. campestris, B. hirta, B. nigra, Nasturtium officinale* (watercress), *Raphanus sativus* (wild radish), *Senecio vulgaris,* and *Cichorium endivia* (escarolle) from 20 different locations in the central coastal lettuce districts and the San Joaquin Valley. The isolates differed in host range, especially with regard to their ability to infect radish. The isolates also differed in severity of symptoms on common hosts, including lettuce. All isolates of TuMV thus far collected are capable of infecting susceptible lettuce cultivars.

Resistant to TuMV and Bremia lactucae: To determine how widespread susceptibility to TuMV is in cultivars of lettuce and to determine the possible relationship of mildew resistance to TuMV susceptibility, a survey of cultivars of *L. sativa* and prickly lettuce *L. serriola* was conducted.

1) *Survey of cultivars of L. sativa.* Seventy-nine cultivars of lettuce were evaluated for resistance to TuMV and mildew (Table 1). Some cultivars were susceptible to TuMV, and others resistant, with no segregation observed within the population of a given cultivar. TuMV-susceptible cultivars were all of the crisphead type: 'Calmar', 'E-4', 'Imperial 410', 'Imperial Triumph', 'Valrio', 'Valtemp', and 'Valverde'.

The cultivars tested were either susceptible or resistant to mildew with no segregation within the population of a given cultivar. Downy-mildew-resistant cv. were: crisphead type-'Calmar', 'E-4', 'Imperial 410', 'Imperial Triumph', 'Valrio', 'Valtemp', 'Valverde'; butterhead type-'May King', 'Meikoningin', 'Proeftuin's Blackpool', 'Ventura'; leaf type-'Red Salad Bowl', 'Salad Trim'; and cos type-'Valmaine'. The results indicate that mildew race 5 was present, since cultivars resistant to race 4 were susceptible (5, 6). The number of seedlings tested ranged from 63 to 87 for each cultivar.

2) *Survey of L. serriola.* Seed was harvested from several *L. serriola* plants at each of 21 locations in the Santa Clara and Salinas Valleys. A minimum of 10 seedlings from each location were tested for resistance to TuMV and mildew (Table 2). Although the populations tested were small the plants fell into 3 classifications: a) TuMV-resistant, mildew-resistant; b) TuMV-resistant, mildew-susceptible, and c) TuMV susceptible, mildew-resistant. No plants were classified as TuMV-susceptible, mildew-susceptible.

Nature of resistance to TuMV in L. sativa: 1) *Extreme resistance.* The TuMV susceptible cv. 'Calmar', 'Valverde', and 'Imperial 410' and the resistant cv. 'Great Lakes 118' were used for test. Young lettuce plants were

approach grafted. The following combinations were obtained: a) susceptible "Calmar' to resistant 'Great Lakes 118', b) susceptible 'Valverde' to resistant 'Great Lakes 118', and c) susceptible 'Calmar' to susceptible 'Imperial 410'. After 3 weeks green peach aphids carrying TuMV were caged on several leaves of the susceptible cv. 'Calmar' and 'Valverde'. Plants were watched for symptoms, and virus recovery was attempted at intervals. The susceptible inoculated cultivars showed symptoms in about 2 weeks. No TuMV symptoms developed on resistant 'Great Lakes 118'. TuMV was recovered from the susceptible inoculated cultivars but not from resistant 'Great Lakes 118'. TuMV was transmitted from susceptible 'Calmar' to susceptible 'Imperial 410' in 5 approach grafts.

Two months after the TuMV inoculations, lettuce mosaic virus (LMV) was inoculated mechanically into the 'Calmar' or the 'Valverde' side of grafts onto 'Great Lakes 118'. Since 'Calmar', 'Valverde', and 'Great Lakes 118' are susceptible to LMV, development of LMV symptoms and recovery of the virus from the 'Great Lakes 118' would indicate a graft union. LMV symptoms developed in 'Great Lakes 118' and the virus was recovered from 'Great Lakes 118' in 2 of the 'Calmar'-'Great Lakes 118' grafts, and in 3 of the 'Valverde'-'Great Lakes 118' grafts. As further evidence of a graft, the scion 'Great Lakes' was cut below the union and continued to live.

These data indicate that 'Great Lakes 118' has extreme resistance ("immunity") to TuMV. It would appear that the TuMV-resistant cultivars reported in Table 1 are also extremely resistant, since no local lesions developed and in no case was the virus recovered.

2) *Tolerance.* Early observations indicated that severity of symptoms differed between susceptible lettuce cultivars. Experiments were conducted to test susceptible *L. sativa* cultivars for tolerance to TuMV. Each treatment consisted of 5 matched plants. Plants were inoculated in the fifth-true-leaf stage with strain S or strain B of TuMV. Symptom scoring was as follows: 0 = no symptoms, 1 = trace, 3 = slight, 5 = moderate, 7= severe, and 9 = death. The plants were examined periodically, and each plant was given a symptom rating. Curves were drawn to fit the average of all plant ratings. The cultivars differed in tolerance to the 2 TuMV strains (Fig. 1). Tolerance in 'Calmar' to strain S, and in 'Calmar' and 'Imperial 410' to strain B, however, does not appear high enough for use in a breeding program for TuMV tolerance. Severity of symptoms on 'Valrio' and 'Valtemp' were similar to those observed on 'Valverde'.

3) *Resistance to infection.* Two experiments were conducted to determine whether 'Calmar' and 'Valverde' differed in resistance to TuMV infection. A paired plot design was used, with 6 replications in the mechanical inoculation study and 5 replications in the aphid inoculation study. Each plot had 24 to 30 plants. Mechanical inoculation was carried out using inoculum from diseased 'Calmar' plants. In the insect inoculation a single green peach aphid was transferred to each healthy lettuce seedling after a 24-hr acquisition feeding period on diseased shepherd's purse plants. Each lettuce plant was caged and the aphid allowed to feed 24 hr. Per cent infection was noted 28 days after inoculation. Percentages were converted to the appropriate angle of transformation and subjected to analysis of variance. Per cent infection in the mechanically inoculated plants was: 'Calmar', 38.2%; 'Valverde', 67.2%. In the aphid inoculated plants, the per cent infection was: 'Calmar', 9.5%; 'Valverde', 25.3%. 'Valverde' had a significantly higher per cent infection, at the 1% level, than 'Calmar' with both mechanical and aphid inoculation.

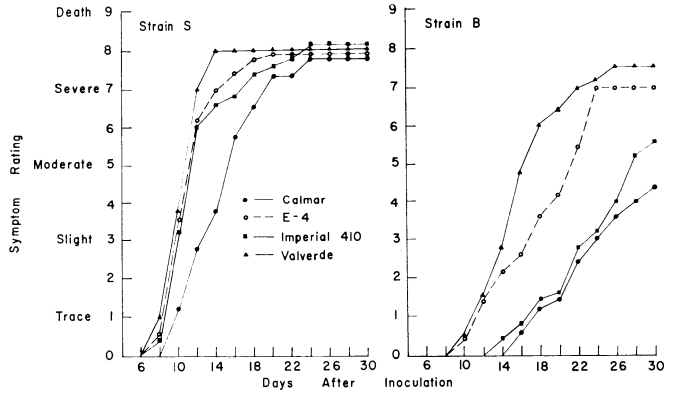

Fig. 1. Effect of 2 strains of Turnip Mosaic Virus on symptom severity of 4 *L. sativa* cultivars.

Nature of resistance in L. serriola. Systemically infected *L. serriola* plants from collection No. 20 and resistant plants from collection No. 6 were used for tests. The following bud grafts were obtained: a) bud from systemically infected plant grafted to resistant plant; b) bud from resistant plant grafted to systemically infected plant. Two successful grafts were made from systemically infected plants to resistant stock, and 3 from resistant plants to systemically infected stock. In no combination did the resistant bud or stock plant develop TuMV symptoms over a 3-month period. New growth in systemically infected stock plants developed TuMV symptoms, and systemically infected buds grafted to resistant plants developed symptoms. TuMV virus was recovered from susceptible scion and stock, but in no graft was the virus recovered from resistant scion or stock. The results indicate that within the population of *L. serriola* extreme resistance to TuMV is present.

A second type of resistance, connected with a hypersensitivity reaction, was observed in *L. serriola.* Several plants developed necrotic local lesions but the virus did not become systemic.

Discussion

A survey of 79 *L. sativa* cultivars indicated that TuMV susceptibility is restricted to mildew-resistant crisphead

Table 2. Resistance to TuMV and downy mildew in *Lactuca serriola,* (number of plants).

Collection number[a]	TuMV-resistant		TuMV-susceptible[b]	
	Downy-mildew-resistant	Downy-mildew-susceptible	Downy-mildew-resistant	Downy-mildew-susceptible
1	0	5	5	0
2	8	2	0	0
3	1	0	9	0
4	0	20	0	0
5	20	0	0	0
6	20	0	0	0
7	5	0	5	0
8	0	0	20	0
9	20	0	0	0
10	20	0	0	0
11	20	0	0	0
12	5	5	0	0
13	0	10	0	0
14	0	0	20	0
15	0	20	0	0
16	0	10	0	0
17	0	20	0	0
18	1	0	9	0
19	9	1	0	0
20	0	0	20	0
21	8	2	0	0
Total	137	95	79	0

[a]Collections from 21 locations in the Santa Clara and Salinas Valleys of California.
[b]Systemic infection. TuMV was recovered from susceptible plants, but not from resistant plants.

types. The cv. 'Calmar', 'Imperial 410', 'Valrio', 'Valtemp', and 'Valverde' are derived from crosses made in 1932 by the late Dr. I. C. Jagger. Resistance to mildew in these cultivars stems from a *L. serriola* collection reported in the literature (4, 6, 10, 11) as P.I. 104854. However, according to H. F. Winters, New Crops Research Branch, ARS, U.S.D.A., the correct number is P.I. 91532. Jagger and Whitaker (6) have shown that immunity to mildew physiologic race 5 is dependent upon a single dominant gene from P.I. 91532. 'Imperial 410' also has a second source of resistance to mildew derived from a strain of 'Chinese Stem' lettuce (4).

'Valverde' 's pedigree includes P.I. 91532, 'Imperial D', 'Grand Rapids', 'Imperial 152', 'Imperial 847', 'Imperial 850', and 'Cosberg' (11, Fig. 3). 'Calmar' has an early pedigree similar to 'Valverde' but with additional crosses to 'Great Lakes 6238' and 'Great Lakes A-36' (10; 11, Fig. 3). 'Valrio' and 'Valtemp' pedigrees include P.I. 91532, 'Imperial D', 'Imperial F', 'Imperial 152', and 'Great Lakes 66' (2; 3; 11, Fig. 2). 'Imperial 410' pedigree includes P.I. 91532, 'Chinese Stem', 'Imperial D', 'Imperial F', 'Imperial 152', and 'Imperial 847' (4, Fig. 5). Unfortunately, seed of P.I. 91532 and Jagger's 'Chinese Stem' lettuce is no longer available. Seed of the other items in the above pedigrees, however, was obtained from the National Seed Storage Laboratory and from Dr. T. W. Whitaker. All proved susceptible to mildew and resistant to TuMV (Table 1). These findings suggest that TuMV susceptibility was introduced into the pedigrees by P.I. 91532. This hypothesis is supported by finding of mildew-resistant and TuMV-susceptible plants in the native *L. serriola* population (Table 2). A further tie between mildew resistance and TuMV susceptibility was sought in tests of advanced breeding lines of Dr. J. E. Welch, University of California, Davis, involving crosses of 'Great Lakes' cultivars (TuMV-resistant, mildew-susceptible) with a mildew resistant P.I. 177418 from Turkey. The following resistant combinations were observed: a) TuMV-susceptible, mildew-resistant; b) TuMV-resistant, mildew-susceptible; c) TuMV-resistant, mildew-resistant. Seed of P.I. 177418 was obtained from the North Central Regional Plant Introduction Station in 1967. Nearly 1,500 seedlings were tested. All plants were susceptible to mildew and resistant to TuMV. An earlier sample of P.I. 177418 tested in 1961 yielded 3 mildew-resistant plants in the population of 84 tested. The mildew-resistant portion of the population appears to have been lost in subsequent seed increase.

Cultivar 'E-4' was released in 1943 by the late Dr. LeRoy E. Weaver, Growers Ice and Development Company, Salinas, California. The source of resistance to mildew or susceptibility to TuMV is not known, because no pedigree record is available. 'Imperial Triumph' is reported to be a selection from 'E-4'.

The authors know of no crisphead cultivars that are resistant to both mildew and TuMV. Within the cos type 'Valmaine' is mildew-and-TuMV-resistant. The 'Valmaine' source of mildew resistance is P.I. 167150, introduced from Turkey in 1949 (7). Leaf-type 'Salad Trim' and 'Red Salad Bowl', introduced by Dessert Seed Company, are mildew-and-TuMV resistant. 'May King', developed by Max Kretchner in 1902, is resistant to mildew and TuMV. Jagger (6), however, found 'May King' susceptible to certain mildew races in England and in the Imperial Valley of California. 'Meikoningin' is a selection from 'May King' and is mildew-and-TuMV resistant. The Dutch glasshouse cv. 'Proeftuin's Blackpool' and 'Ventura' are mildew-and-TuMV resistant. Rodenburg (8), however, reports 'Proeftuin's Blackpool' and 'Ventura' resistant to mildew race 1 in the Netherlands, but susceptible to race 2. The source of mildew resistance in 'Salad Trim', 'Red Salad Bowl', 'May King', 'Meikoningin', 'Proeftuin's Blackpool', and 'Ventura' is not known.

Several facts established by this investigation merit further discussion, since they are of general genetic and pathological significance. Strong circumstantial evidence indicates that TuMV susceptibility in *L. sativa* is associated with mildew-resistant progenies derived from P.I. 91532 and P.I. 177418. In *L. serriola* TuMV susceptibility was also associated with mildew resistance. In none of the *L. sativa* cultivars and breeding lines or in *L. serriola* were plants found that were susceptible to both TuMV and mildew. This suggests a linkage between genes for mildew resistance and TuMV susceptibility.

Genes for mildew resistance in cv. of the butterhead, leaf, and cos types and in *L. serriola* may be different than the single gene for resistance from P.I. 91532 and P.I. 177418. Such genes could be used to broaden the genetic base for mildew resistance against mutations of new physiological races. Resistance to TuMV should be incorporated into mildew-resistant crisphead types, particularly cultivars adapted to the Salinas-Watsonville area, where both diseases are important.

Species found to be susceptible to TuMV, but not, to our knowledge, previously reported are: *Amsinckia douglasiana, Beta macrocarpa, Claytonia perfoliata, Conium maculatum, Cynara scolymus, Erysimum asperum, Eschscholtzia californica, Geranium dissectum, Gossypium hirsutum, Lactuca sativa, L. serriola, Nicotiana megalosiphon, Phaseolus vulgaris, Physalis ixocarpa, P. wrightii, Senecio vulgaris, Sisymbrium irio, Sonchus oleraceus, Spergula arvensis,* and *Tetragonia expansa.*

The susceptibility of certain cultivars of lettuce and the resistance of others to all isolates of TuMV thus far tested may prove of value as a diagnostic tool for the identification of this virus.

Literature Cited

1. Anonymous. 1963. Valmaine, a new mildew immune romaine lettuce variety. *Tex. Agr. Exp. Sta. Leaflet* 610:1–3.
2. ———. 1968. Notice to primary seed producers of the release of the low temperature adapted, downy mildew resistant lettuce variety Valrio for the lower Rio Grande Valley. *Tex. Agr. Exp. Sta., U.S. Dept. Agr. Res. Serv. Release Sheet.*
3. ———. 1968. Notice to primary seed producers of the release of Valtemp, a downy mildew resistant variety of head lettuce adapted to high temperature-early season planting in the lower Rio Grande Valley. *Tex. Agr. Exp. Sta., U.S. Dept. Agr. Res. Serv. Release Sheet.*
4. Bohn, G. W., and Thomas W. Whitaker. 1951. Recently introduced varieties of head lettuce and methods used in their development. *U.S. Dept. Agr. Res. Serv. Circ.* 881:1–27.
5. Jagger, Ivan C., and Norman Chandler. 1933. Physiologic forms of *Bremia lactucae* on letuce. *Phytopathology* 23:18–19. (Abst.)
6. ———, and Thomas W. Whitaker. 1940. The inheritance of immunity from mildew *(Bremia lactucae)* in lettuce. *Phytopathology* 30:427–433.
7. Leeper, Paul W., Thomas W. Whitaker, and G. W. Bohn. 1959. Lettuce mildew resistant variety. *Amer. Veg. Grower* 7 (9):18.
8. Rodenburg, C. M. 1966. Some testing procedures and research experiences on resistance to *Bremia lactucae* Regel. in lettuce. *Euphytica* 15:141–148.
9. Shepherd, Robert J., and Glenn S. Pound. 1960. Purification of turnip mosaic virus. *Phytopathology* 50:797–803.
10. Welch, J. E., R. G. Grogan, F. W. Zink, G. M. Kihara, and K. A. Kimble. 1965. Calmar, a new lettuce variety resistant to downy mildew. *Calif. Agr.* 19 (8):3–4.
11. Whitaker, Thomas W., G. W. Bohn, J. E. Welch, and R. G. Grogan. 1958. History and development of head lettuce resistant to downy mildew. *Proc. Amer. Soc. Hort. Sci.* 72:410–416.