Relationship of a Non-Lethal Reaction to a Virulent Isolate of Lettuce Mosaic Virus and Turnip Mosaic Susceptibility in Lettuce[1]

F. W. Zink[2], James E. Duffus[3], and K. A. Kimble[4]
University of California, Davis, and Agricultural Research Service, U. S. Department of Agriculture, Salinas, California

Abstract. A mosaic disease of *Lactuca sativa* L. is described and the causal agent identified as a new virulent isolate of lettuce mosaic virus (LMV). The reservoir of infection was bristly oxtongue, *Picris echioides* L. *Lactuca sativa* L. cvs. Gallega, Calmar, Imperial 410, and Bibb systemically infected with virulent LMV did not transmit the virus through the seed. A survey of *L. sativa* cultivars indicated that the non-lethal reaction to the virulent isolate was restricted in the crisphead type to cultivars that are turnip mosaic virus (TuMV) susceptible, and downy mildew-resistant. A similar relationship was found in *L. serriola* lines. The non-lethal reaction was conferred by dominant complementary genes. 'Gallega', reported to be resistant to common LMV, was found to be susceptible to systemic infection by the virulent LMV isolate.

In 1968, a mosaic disease of lettuce, *Lactuca sativa* L., was observed in 2 fields in the northern end of the Salinas Valley of California. It appeared in crisp-head cv. Great Lakes 65, grown from a seed lot certified free from seed-borne lettuce mosaic virus (LMV). When found, the fields were near market maturity, but were a total loss, with a high percentage of dead or dying plants with severe necrosis of the leaf blades. A survey of the noncultivated areas adjacent to the fields revealed a population of bristly oxtongue, *Picris echioides* L., with mosaic mottling. Recovery tests from lettuce and bristly oxtongue plants with mosaic symptoms indicated the disease to be lethal on some lettuce cultivars, but not on others.

Because of the destructiveness of the disease, the biological and physical characteristics of the causal virus were investigated. The investigations indicated that the causal virus was a virulent isolate of lettuce mosaic virus designated herein as strain L (LMV-L). The nature of inheritance of the lethal LMV reaction, and investigations of cultivars with the non-lethal LMV-L reaction are reported.

Materials and Methods

Host-range studies were done by 2 methods. Recovery tests from field plants with mosaic symptoms indicated some species were naturally infected with LMV-L. The virus was recovered by feeding nonviruliferous green peach aphids, *Myzus persicae* (Sulz.), on collected field plants for 24 hr and then transferring the insects to healthy indicator seedlings for 24 hr. Additional host-range studies were done by inoculating at least 10 seedlings of a number of different species with viruliferous green peach aphids fed on LMV-L diseased bristly oxtongue for 24 hr. Recovery attempts from all inoculated plants were made to verify susceptibility.

Mechanical inoculations were made 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 done with a randomized-block or Latin-square design on whole or half leaves of the test plant.

Two cross protection tests were made between a typical seed-borne isolate of LMV and LMV-L isolate. The typical isolate and LMV-L were each mechanically inoculated into 2 separate groups of 'Great Lakes 118' in the 4th or 5th leaf stage. Two weeks after the first inoculation, the older leaves were removed from the plants in all treatments; leaving 3 or 4 leaves per plant. The inoculated plants showed mosaic symptoms in their remaining leaves. Half of the plants inoculated with the typical isolate were then inoculated with LMV-L. A second group of plants was inoculated with the typical or LMV-L. Each treatment consisted of 10 plants arranged in a randomized-block design. The plants were harvested 23 days and 38 days after the second inoculation in Tests 1 and 2, respectively. Fresh wt of the above-ground portion of the plants were subjected to analysis of variance and to Duncan's multiple range test.

The effect of plant age on susceptibility to infection by LMV-L was studied in 'Great Lakes 118', 'Imperial 410', and 'Gallega'. Ten plants of each cultivar were mechanically inoculated 28 days after planting (seedling stage), and at 56 (rosette stage), 84 (market maturity), and 112 days (seed stalks, but before flowering). Symptoms were noted and number of days from inoculation to lethal reaction was recorded.

Crosses were made between the homozygous, LMV-L lethal reacting, downy mildew-susceptible, turnip mosaic virus (TuMV)-resistant 'Great Lakes 118' and the homozygous LMV-L non-lethal reacting, mildew-resistant, TuMV-susceptible 'Calmar' or 'Imperial 410'. In each cross 'Calmar' or 'Imperial 410' was used as the pollen parent. The F_1 plants were identified by their resistance to downy mildew infection, and were not tested for their reaction to LMV-L.

Seeds for plants to be assayed for lethal reaction were germinated on moist filter paper in petri dishes at 20 to 22°C under continuous light. The young seedlings were transplanted into flats at the cotyledon stage.

Nonviruliferous green peach aphids were reared on radish, *Raphanus sativus* L. The nonviruliferous aphids were transferred to LMV-L infected bristly oxtongue plants for 24 hr; then approx 20 individuals were transferred to each F_2 or F_3 lettuce seedling for an infection feeding period of 24 hr. The plants were then sprayed with nicotine sulfate and placed in greenhouses which were fumigated at weekly intervals, also with nicotine sulfate.

The F_2 seedlings from 5 crosses involving 3 parent cultivars, and F_3 seedlings involving 2 parent cultivars, were classified with respect to disease reaction into 2 categories: 1) plants with the lethal reaction within 38 days after inoculation and 2) plants with no lethal reaction. Fifty surviving seedlings from each of the F_2 families were grown for seed production.

The procedures for inoculation and scoring plants for the non-lethal reaction in the survey of *L. sativa* cultivars and *L.*

[1]Received for publication, May 30, 1972.

[2]Specialist in the Experiment Station, Department of Vegetable Crops.

[3]Plant Pathologist, Plant Science Research Division.

[4]Staff Research Associate in the Experiment Station, Department of Plant Pathology.

serriola lines were similar to those used in the inheritance study.

Results

Identification of the causal virus. Early studies of symptomology and host range indicated that the virus isolate which was lethal on 'Great Lakes 65' in the field was similar in several ways to typical isolates of LMV. Investigations were undertaken to determine the relationship between LMV and the virulent virus isolate.

1) *Host range.* Limited comparative host range studies between a typical seed-borne LMV isolate and the virulent isolate were conducted. A similar reaction on all host plants tested was recorded for the common LMV isolate and the virulent isolate. Plants found susceptible to the virulent isolate are listed in alphabetical order.

AMARANTHACEAE. *Gomphrena globosa* L.

CHENOPODIACEAE. *Chenopodium amaranticolor* Costa and Reyn.; *C. capitatum* (L.) Asch.; *C. murale* L.; *C. quinoa* Willd; *Spinacia oleracea* L.

COMPOSITAE. *Callistephus chinenis* (L.) Nees; *Cichorium endivia* L.; *C. intybus* L.; *Lactuca sativa* L.; *L. serriola* L.; *L. virosa* L.; *Picris echioides* L.; *Senecio vulgaris* L.; *Zinnia elegans* L.

LEGUMINOSAE. *Lathyrus odoratus* L.; *Pisum sativum* L.

2) *Virus properties.* Properties of a typical LMV isolate and the virulent isolate were determined by mechanical inoculation of extracts from lettuce and bristly oxtongue into *Chenopodium amaranticolor* and *C. quinoa.*

a) *Thermal inactivation.* Extracts of both isolates were heated in a water bath for 10 min at 20, 45, 50, 55, 60, and 65°C. Infectivity of both isolates was greatly reduced at 45°C, and was not detected after treatment at 50°C.

b) *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 were diluted with the same buffer in series to 5^{-5}. Infectivity of both the virulent isolate and the typical LMV isolate was low at dilutions of 5^{-3}, and was not found at 5^{-4}.

c) *Longevity in vitro.* Virus extracts *in vitro* of both virus isolates lost activity rapidly at room temp. Only a trace of activity remained in preparations aged for 48 hr.

3) *Serology.* Antiserum against virus preparations from concentrated butanol-clarified *Chenopodium quinoa* L. infected with seed-borne LMV was produced in rabbits. Double-gel diffusion tests were conducted using unclarified sap of leaves ground 1:1 in 0.05M phosphate buffer, pH 7.6, containing 1.0% thioglycolic acid, made to 15% pyridine. A positive relationship in double-gel diffusion was obtained when the antiserum made against the seed-borne isolate of LMV was compared with the virulant LMV or with the seed-borne isolate but not with healthy.

4) *Cross-protection.* The 'Great Lakes 118' plants infected with the typical isolate of LMV and later inoculated with the virulent isolate had only typical isolate symptoms. Also, there were no significant differences in mean fresh wt between plants infected with the typical isolate only and those infected with the typical isolate and later inoculated with the virulent isolate (Table 1). Recovery attempts from the double-inoculated plants yielded only the typical isolate. Thus the typical LMV isolate completely protected against infection by the virulent isolate. Host range, physical properties, and immunological studies indicate a close relationship between the virulent isolate and typical LMV. The virulent isolate is designated strain L (LMV-L).

Symptoms of LMV-L on L. sativa. Cultivars showing the lethal reaction developed symptoms similar to those observed on 'Great Lakes 65' exposed to natural infection in 1968. An early symptom was vein clearing of young leaves 6 to 7 days after inoculation. Subsequently, the older leaves developed severe necrosis, thickened, and became distorted. The young leaves took on a dark green, water-soaked appearance, and then wilted. The plant appeared to die from the young leaves outwardly to the older leaves.

Cultivars not showing the lethal reaction developed vein clearing followed by mild to severe necrosis and a distortion of the leaf blades (Fig. 1). After the initial shock of infection, the plants recovered to a degree. They were stunted, and the tips of the outer leaves rolled downward, giving the plants a wilted appearance. During the reproductive phase of growth the younger leaves had a yellow green mottle, and their margins frequently had considerable necrosis. Necrotic areas sometimes formed on the developing involucral bracts and peduncles, and many of the floral heads withered before they matured. The number of seeds per flower head and seeds per plant were extremely low.

Transmission tests. 1) *Mechanical.* The LMV-L strain from bristly oxtongue was readily transmitted by routine mechanical inoculation techniques. The efficiency of transmission in 97 'Calmar' plants was 66%, in 98 'Imperial 410' plants 63%, in 95 'Great Lakes 118' plants was 74% and in 98 'Gallega' plants 60%.

Table 1. Cross-protection by a typical LMV isolate against a virulent strain of LMV-L in 'Great Lakes 118' lettuce.

Test no.	Treatment: LMV isolate & inoculation date	Mean fresh wt per plant[z]
		(g)
1	Noninoculated	90.9a
	Typical LMV 1/10	48.2b
	Typical LMV 1/10 + LMV-L 1/24	45.9b
	LMV-L 1/24	20.5c
2	Noninoculated	131.4a
	Typical LMV-L 4/17	95.0b
	LMV-L 4/17	0.0d
	Typical LMV 5/1	99.9b
	Typical LMV 4/17 + LMV-L 5/1	102.3b
	LMV-L 5/1	14.1c

[z]Two means having any letter in common are not significantly different from each other at the 1% level.

2) *Seed.* Seeds from each of 20 plants of 'Gallega' systemically infected with LMV-L and from 10 infected plants each of 'Calmar', 'Imperial 410', and 'Bibb', were harvested

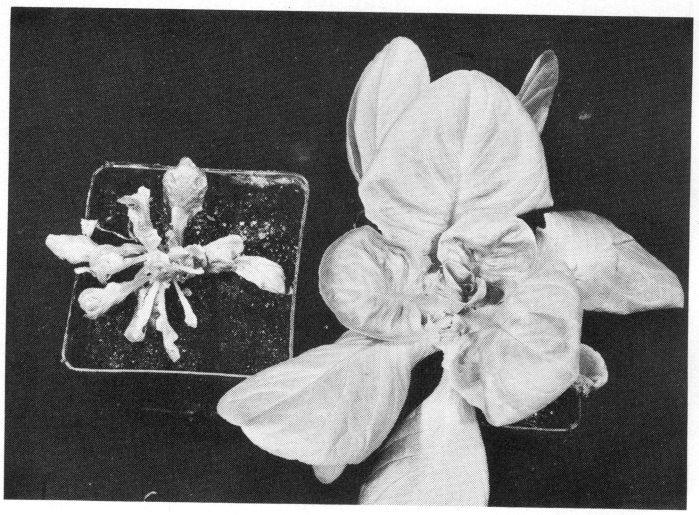

Fig. 1. Symptoms of lettuce mosaic virus virulent strain (LMV-L) in 'Gallega' 28 days after inoculation, and noninfected 'Gallega' plant the same age.

separately and checked for virus transmission. No seed transmission was observed in 6,356 'Gallega' and 1,942 'Bibb' seedlings. Four of 976 seedlings of 'Calmar' and 3 of 1,692 seedlings of 'Imperial 410' appeared to have seed-borne infection. However, it is possible that these diseased seedlings were the result of early secondary infection. Recovery tests from these seedlings indicated that they were infected with a typical isolate of LMV. Thus, LMV-L appears not to be seed-borne in these cultivars.

3) *Insect.* The LMV-L strain was readily transmitted by *Myzus persicae* utilizing short (1 hr) acquisition and test feeding intervals. Retention of the virus by the green peach aphid was determined by daily serial transfers to healthy lettuce seedlings of 'Great Lakes 118'. Viruliferous aphids transmitted the virus to healthy plants only during the first day.

Effect of plant age on susceptibility to infection. 'Imperial 410' and 'Gallega' became systemically infected with LMV-L at the 4 ages that inoculations were made. However, neither cultivar responded with the lethal reaction. 'Great Lakes 118' also became systemically infected at all inoculation dates, but did give the lethal reaction. The mean numbers of days from inoculation to death were 20.8, 31.7, 34.7, and 32.7 for plants inoculated at 28, 56, 84, and 112 days, respectively.

Occurrence of the lethal LMV reaction. To determine how widespread the lethal and non-lethal reaction to LMV-L is in cultivars of lettuce a survey of *L. sativa* and prickly lettuce, *L. serriola,* was conducted.

1) *Survey of cultivars of L. sativa.* Sixty-seven cultivars were evaluated for the lethal reaction to LMV-L (Table 2). Some cultivars had a lethal reaction, others did not, but no segregation was observed within a population of a given cultivar. All cultivars inoculated with LMV-L, however, became systemically infected. Crisphead types with the non-lethal reaction were 'Calicel', 'Calmar', 'E-4', 'Imperial 410', 'Imperial Triumph', 'Valrio', 'Valtemp', and 'Valverde'. These cultivars are all TuMV-susceptible and downy mildew (incited by *Bremia lactucae* Reg.) resistant (12). The crisphead cultivars with the lethal reaction are TuMV-resistant, and mildew-susceptible. All the butterhead, leaf, cos, Latin, and stem type cultivars tested are TuMV-resistant. The mildew-resistant cvs. Bourguignonne, Red Salad Bowl, and Salad Trim had a lethal reaction, whereas the mildew-resistant 'May King', 'Proeftuin's Blackpool', and 'Valmaine' had a non-lethal reaction. There appears to be no clear association of the lethal reaction to virulent LMV-L with TuMV-resistance, mildew-resistance or susceptibility in the butterhead, leaf, cos, Latin, and stem type cultivars.

2) *Survey of L. serriola.* From a collection of seed made in the Santa Clara and Salinas Valleys (12), several *L. serriola* lines were selected which were homozygous TuMV-resistant, mildew-resistant; TuMV-resistant, mildew-susceptible; or TuMV-susceptible, mildew-resistant. Twelve lines were evaluated for the lethal reaction to LMV-L (Table 3). Lines that were TuMV-resistant, mildew-resistant, and TuMV-resistant, mildew-susceptible, had a lethal reaction. Lines which were TuMV-susceptible, mildew-resistant did not have a lethal reaction. All plants in each of the 12 lines became systematically infected with LMV-L.

Inheritance of the lethal reaction. The parental combinations, the observed number of F_2 plants with the lethal and non-lethal reaction to LMV-L and Chi square and probability values are given in Table 4. These data indicate that the F_2 progenies of the several crosses segregated approx 9 non-lethal to 7 lethal. Only 16 F_2 plants which survived infection of LMV-L produced enough seed to test the F_3 progenies. These were from the 'Great Lakes 118' x 'Imperial 410' cross. Although the F_3 population was small it could be separated into 3 classes: (a) homozygous non-lethal, (b) segregating 9 non-lethal to 7 lethal, and (c) segregating 3 non-lethal to 1 lethal (Table 5). Data on the parents, the F_2,

Table 2. Reaction of *L. sativa* cultivars to a virulent strain of LMV-L.

Cultivar and type	Plants inoculated	Plants with lethal reaction	TuMV[z]	Mildew[y]
	(number)	(number)	(R=resistant) (S= susceptible)	
Crisphead type				
Calicel	40	0	S	R
Calmar	40	0	S	R
Caravan	20	20	R	S
Climax	20	20	R	S
Cosberg	20	20	R	S
E-4	39	0	S	R
Empire	20	20	R	S
Francisco	20	20	R	S
Forty-Niner	20	20	R	S
Fulton	20	20	R	S
Golden State A	20	20	R	S
Golden State B	20	20	R	S
Golden State D	20	20	R	S
Great Lakes 54	20	20	R	S
Great Lakes 65	20	20	R	S
Great Lakes 66	20	20	R	S
Great Lakes 118	212	212	R	S
Great Lakes 366	20	20	R	S
Great Lakes R-200	20	20	R	S
Great Lakes 659	20	20	R	S
Great Lakes 6238	20	20	R	S
Green Bay	20	20	R	S
Greenland	20	20	R	S
Hanson	20	20	R	S
Imperial D	20	20	R	S
Imperial F	20	20	R	S
Imperial 44	20	20	R	S
Imperial 101	20	20	R	S
Imperial 152	20	20	R	S
Imperial 410	215	0	S	R
Imperial 615	20	20	R	S
Imperial 847	20	20	R	S
Imperial Triumph	40	0	S	R
Ithaca	20	20	R	S
Merit	20	20	R	S
Minetto	20	20	R	S
Oswego	20	20	R	S
Vanguard	20	20	R	S
Valrio	40	0	S	R
Valtemp	40	0	S	R
Valverde	40	0	S	R
Butterhead type				
Bibb	35	0	R	S
Big Boston	40	0	R	S
Bourguignonne	20	20	R	R
Dark Green Mignonette	40	0	R	S
May King	40	0	R	R
Midas	35	0	R	S
Proeftuin's Blackpool	35	0	R	R
Tom Thumb	35	0	R	S
Wayahead	20	20	R	S
White Boston	40	0	R	S
Leaf type				
Black Seeded Simpson	40	0	R	S
Grand Rapids	40	0	R	S
Oak Leaf	40	0	R	S
Prize Head	20	20	R	S
Red Salad Bowl	20	20	R	R
Salad Bowl	20	20	R	S
Salad Trim	20	20	R	R
Slowbolt	20	20	R	S
Cos type				
Dark Green Cos	40	0	R	S
Parris Island Cos	20	20	R	S
Valmaine	40	0	R	R
White Paris Cos	40	0	R	S
Latin type				
Fordhook	40	0	R	S
Gallega	40	0	R	S
Stem type				
Celtuce	20	20	R	S
Chinese	20	20	R	S

[z]Turnip mosaic virus from Salinas Valley, California.
[y]Downy mildew (*Bremia lactucae*) collection from the central coastal districts of California.

Table 3. Reaction of *L. serriola* lines to a virulent strain of LMV-L.

Collection number[z]	Phenotype[y]		LMV-L isolate	
	TuMV	Mildew	Plants inoculated	Plants with lethal reaction
			(number)	(number)
5	R	R	40	40
6	R	R	40	40
9	R	R	40	40
10	R	R	40	40
11	R	R	38	38
4	R	S	40	40
13	R	S	40	40
15	R	S	39	39
16	R	S	40	40
8	S	R	40	0
14	S	R	40	0
20	S	R	40	0

[z]Collection numbers from literature citation (13).
[y]R = resistant, S = susceptible.

and the F_3 from surviving F_2 plants indicate that the mode of inheritance of the non-lethal reaction is one of dominant complementary genes.

Discussion

Seed transmission has been indicated as the major factor in the spread of lettuce mosaic virus in England (1) and the United States (3). The efficacy of virus-free lettuce seed for the control of common lettuce mosaic has been demonstrated in other tests (15), and by the mosaic-free seed program in the Salinas Valley (2). In the last several years occasional outbreaks of severe variants of LMV occurred. These outbreaks were not associated with seed transmission, but were mainly local phenomena associated with reservoirs in wild hosts. Over 550 lots of commercial lettuce seed were tested, and plants observed for seed transmission. From each lot, 30,000 seedlings were examined, with no apparent evidence of severe lettuce mosaic symptoms (A. Greathead, Farm Advisor, Monterey County, California, personal communication). The lack of seed transmission of LMV-L in our study confirms these observations.

Table 4. F_2 populations, showing mode of inheritance to non-lethal LMV-L reaction.

Family	Parentage	Observed		X^2	P
		Non-lethal	Lethal	9:7[z]	
164	GL 118 X Imp. 410	175	143	0.191	0.70-0.50
165A	GL 118 X Imp. 410	229	193	0.675	0.50-0.30
166A	GL 118 X Imp. 410	167	144	0.823	0.50-0.30
63B	GL 118 X Calmar	86	56	1.073	0.50-0.30
56B	GL 118 X Calmar	80	76	1.564	0.30-0.20
Total		737	612	1.353	0.30-0.20

[z]Homogeneity of families: X^2=4.356, P=0.50-0.30.

Four variants of LMV, designated as variants 1, 2, 3, and 4, were reported by McLean and Kinsey (5, 6) from the Salinas Valley. Symptoms produced by variant 4 were more severe than those produced by any one of the 3 previously described variants (5). McLean and Kinsey (6) reported that plants of 'Great Lakes', 'Dark Green Cos', 'Parris Island Cos', 'Eiffel Tower Cos', 'White Boston', and 'Prize Head' infected with variant 4 died a short time after symptom expression. Variant 4 was recovered from an infected plant of lambs'-quarters, *Chenopodium album* L., growing in the vicinity of a 'Dark Green Cos' lettuce planting. In contrast, in our study the weed host was bristly oxtongue, and 'Great Lakes 65' was infected. Furthermore, 'Dark Green Cos' and 'White Boston' did not die when infected with our LMV-L strain. The area where LMV-L was recovered was approx 5 miles from the semi-isolated area where variant 4 was found. It seems probable that there are several virulent isolates of LMV in a number of wild hosts in the Salinas Valley.

Table 5. Observed segregation in F_3 families from F_2 plants which survived LMV-L infection.

Family	Lethal LMV reaction - observed		X^2	P
	Non-lethal	Lethal		
Homozygous non-lethal				
164-2A	189	0	---	---
164-5D	142	0	---	---
165A-3F	138	0	---	---
166A-2A	156	0	---	---
Total	625	0		
Segregating 9 non-lethal to 7 lethal.				
164-5A	52	32	1.091	0.30-0.20
165A-5F	24	17	0.087	0.95-0.70
166A-2C	64	54	0.194	0.70-0.50
166A-2H	36	20	1.469	0.30-0.20
166A-3J	31	16	1.799	0.20-0.10
Total	207	139	1.798	0.20-0.10
Homogeneity of families			2.842	0.50-0.30
Segregating 3 non-lethal to 1 lethal				
164-2F	59	14	1.319	0.30-0.20
164-3H	67	27	0.695	0.50-0.30
165A-1A	28	14	0.555	0.30-0.20
165A-1J	52	12	0.812	0.50-0.30
165A-5F	71	15	2.620	0.20-0.10
166A-5H	45	18	0.428	0.70-0.50
166A-1J	44	10	1.209	0.30-0.20
Total	366	110	0.907	0.50-0.30
Homogeneity of families			7.731	0.30-0.20

A survey of 67 *L. sativa* cultivars indicated that non-lethal reaction to LMV-L in the crisphead types is restricted to TuMV-susceptible and mildew-resistant cultivars. 'Calicel', 'Calmar', 'Imperial 410', 'Valrio', 'Valtemp', and 'Valverde', are derived from crosses originally made in 1932 by the late Dr. I. C. Jagger. Resistance to mildew in these cultivars stems from a *L. serriola* collection P.I. 91532. 'E-4' was released in 1943 by the late Dr. Le Roy E. Weaver, and no pedigree record is available. 'Imperial Triumph' is reported to be a selection from 'E-4'. Pedigrees of the above mentioned cultivars have been reviewed by Whitaker et al. (11) and Zink and Duffus (12) and therefore are omitted here. Downy mildew resistance to physiologic race 5 is dependent upon a single dominant gene (4, 12). Zink and Duffus (13) have reported that resistance to TuMV is dependent upon a single dominant gene, and that the TuMV reaction gene (*Tu, tu*) is linked with the mildew reaction gene (*Dm, dm*). A similar genetic relationship was found in *L. serriola* lines that are TuMV-susceptible and mildew-resistant (14). The *L. serriola* lines that are non-lethal reacting to LMV-L are TuMV-susceptible and mildew-resistant, as were the *L. sativa* crisphead cultivars. This suggests that the genes for non-lethal reaction to LMV-L were introduced into the crisphead cultivars from *L. serriola* P.I. 91532. Furthermore, the complex pedigrees of the crisphead cultivars with the non-lethal reaction suggest that the genes for non-lethal LMV-L reaction are linked to the TuMV-susceptible and downy mildew-resistant genes.

Two sources of resistance to LMV have been reported. Von der Pahlen and Crnko (10) found that 'Gallega' is resistant and Ryder (7) reported finding resistance in 3 lines of a wild or primitive form of lettuce introduced into the United States from Egypt. Resistance is inherited as a single recessive gene, and the same gene occurs in both 'Gallega' and the Egyptian plant introduction lines (9). Ryder (9) reported that the resistance appeared to be either tolerance of, or resistance to, viral increase and spread, as the LMV infection in mosaic-resistant plants was apparently systemic. The source of LMV used in Ryder's investigations (7, 8, 9) is reported as one

causing typical symptoms in Salinas Valley lettuce fields. The symptoms he described indicate that these were relatively mild isolates and not a necrotic or virulent variant.

In our study, 'Gallega' became systemically infected with LMV-L inoculated at 4 stages of growth. The sequence of symptom development and severity of symptoms in 'Gallega' were similar to those observed on 'Imperial 410' and 'Calmar'. The resistance in 'Gallega' appears to be somewhat effective against the typical seed-borne LMV isolates, but not against the more virulent isolate.

Genes for LMV resistance are of value in a lettuce breeding program as long as they confer resistance to the prevalent strains of the virus. The success of the mosaic-free seed program in controlling the typical seed-borne isolates indicates the need for resistance to the more virulent isolates that occur in wild hosts.

Literature Cited

1. Broadbent, L., T. W. Tinsley, W. Buddin, and E. T. Roberts. 1951. The spread of lettuce mosaic in the field. *Ann. Appl. Biol.* 38:687-706.
2. Greathead, Arthur S. 1969. Lettuce mosaic control program. *Western Grower* 40(11):99-101.
3. Grogan, R. G., J. E. Welch, and R. Bardin. 1952. Common lettuce mosaic and its control by the use of mosaic-free seed. *Phytopathology* 42:573-578.
4. Jagger, Ivan C., and Thomas W. Whitaker. 1940. The inheritance of immunity from mildew *(Bremia lactucae)* in lettuce. *Phytopathology* 30:427-433.
5. McLean, D. L., and M. G. Kinsey. 1962. Three variants of lettuce mosaic virus and methods utilized for differention. *Phytopathology* 52:403-406.
6. _____, and _____. 1963. Transmission studies of a highly virulent variant of lettuce mosaic virus. *Plant. Dis. Rpt.* 47:474-476.
7. Ryder, E. J. 1968. Evaluation of lettuce varieties and breeding lines for resistance to common lettuce mosaic. *USDA Tech. Bul.* 1391:8 p.
8. _____. 1970. Screening for resistance to lettuce mosaic. *HortScience* 5:47-48.
9. _____. 1970. Inheritance of resistance to common lettuce mosaic. *J. Amer. Soc. Hort. Sci.* 95:378-379.
10. Von der Pahlen, A., and J. Crnko. 1965. El virus del mosaico de la lechuga (*Marmor lactucae* Holmes) en Mendoza y Buenos Aires. *Rev. Invest. Agropecuaria.* 11:25-31.
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.
12. Zink, F. W., and James E. Duffus. 1969. Relationship of turnip mosaic virus susceptibility and downy mildew *(Bremia lactucae)* resistance in lettuce. *J. Amer. Soc. Hort. Sci.* 94:403-407.
13. _____, and _____. 1970. Linkage of turnip mosaic virus susceptibility and downy mildew, *(Bremia lactucae)* resistance in lettuce. *J. Amer. Soc. Hort. Sci.* 95:420-422.
14. _____, and _____. 1973. Inheritance and linkage of turnip mosaic virus and downy mildew *(Bremia lactucae)* reaction in *Lactuca serriola. J. Amer. Soc. Hort. Sci.* 98:49-51.
15. _____, R. G. Grogan, and J. E. Welch. 1956. The effect of the percentage of seed transmission upon subsequent spread of lettuce mosaic virus. *Phytopathology* 46:662-664.

The Influence of Flower Removal on Growth and Seed Yield of *Phaseolus vulgaris* L.[1]

H. C. Wien[2], R. F. Sandsted, and D. H. Wallace

Abstract. Dry beans (*Phaseolus vulgaris* cvs. Red Kidney and Great Northern) were grown in the cool season in the lowland tropics at Los Banos, Philippines. Manual removal of flowers for 11 days from first bloom resulted in increased wt of vegetative parts and no change in rate of total dry wt gain. New branches, roots, and leaves provided efficient alternate sinks for assimilates, so that leaf area was increased and maintained longer, and more branches formed. Although temporary flower removal increased pod set compared to control plants, pod and seed abortion prevented a significant yield increase. This resulted in lower ratios of seed wt to total dry wt, and decreased pod wt production per unit leaf area.

Dry edible beans (*Phaseolus vulgaris* L.) can be grown during the cool months of November to February under lowland tropical conditions at Los Banos, Philippines. However, the period from emergence to maturity was only 59 days, compared to 99 days at Ithaca, New York. Seed yields were proportionately lower, being 1978 kg/ha at Los Banos and 3177 kg/ha at Ithaca, corresponding to production rates of about 33 kg/ha/day for both locations[3]. Several workers report delay in senescence of plants by removal of flowers or immature pods (9, 13, 16). Ojehomon (16) found when opening flowers of cowpea were removed for up to 12 days, seed yields were not significantly affected, or decreased only slightly. Lyons (11) showed that the gametocide Mendok (2,3-dichloroisobutyrate) temporarily prevented fruit set of tomatoes but increased the amount of fruit set per plant at a later date, increasing single-harvest yields.

This experiment was conducted to determine if early flower removal in dry beans would prolong growth, increase fruit set, and produce higher seed yields. By temporarily altering the major sink for assimilates, and determining effects on vegetative and reproductive growth by growth analysis and light interception measurements, we hoped to better understand source-sink relationships. Since bean cultivars differ greatly in days to first flower (3), the results may have implications in breeding for yield.

Materials and Methods

The experiment was conducted on a well-drained Lipa clay loam soil (1) fertilized by banding 65-28-54 kg/ha (N-P-K) in rows 73 cm apart and 15.2 cm deep. Granular nematocide Nemagon (dibromochloropropane) was mixed with the fertilizer at 10 kg/ha active ingredient. Preplant incorporation of 1 kg/ha of trifluralin provided good weed control. Seeds were hand planted Nov. 14, 1969, about 10 cm above and to the side of

[1]Received for publication June 15, 1972. Paper No. 638 of the Department of Vegetable Crops, Cornell University, Ithaca, New York.

[2]Present address: International Institute of Tropical Agriculture, PMB 5320 Oyo Road, Ibadan, Nigeria.

[3]Wien, H. C. 1971. The influence of spacing on light interception, growth and seed yield of *Phaseolus vulgaris* L. under temperate and tropical conditions. Ph.D. Thesis, Cornell University, Ithaca, New York.

[4]Martinez Rodas, R. 1971. Studies on cessation of pod growth and development in dry beans (*Phaseolus vulgaris* L.). M.S. Thesis, Cornell University, Ithaca, New York.