The Journal of Heredity 78:126–128. 1987.

# Inheritance of resistance to clover yellow vein virus in *Pisum sativum*

**R. Provvidenti**

*ABSTRACT:* Most of the cultivars of *Pisum sativum* resistant to bean yellow mosaic virus (BYMV) were found to be resistant to clover yellow vein virus (CYVV). However, some accessions from Ethiopia (PI 193586 and PI 193835) and India (PI 347464, PI 347465, PI 347466, PI 347467, and PI 347492) were resistant to CYVV, but susceptible to BYMV. Conversely, others from China (PI 391630) and the USSR (PI 262818) were resistant to BYMV, but susceptible to CYVV, indicating that resistance to these two viruses is conferred by distinct genetic entities. In the BYMV+CYVV-resistant cultivar Bonneville, resistance to CYVV was found to be inherited monogenically recessive (*cyv*). This gene appears to be closely linked to that conferring resistance to BYMV (*mo*), which is located on the second chromosome. In the accessions from Ethiopia and India, resistance to CYVV is conditioned by a second recessive gene (*cyv*-2), that is situated in a different linkage group. In the lines from China and the USSR, resistance to BYMV is conferred by *mo*. The possible origin of two distinct genes for resistance to the same isolate of CYVV is discussed.

CLOVER yellow vein virus (CYVV) is the causal agent of devastating diseases occurring in the bean and pea in the northeast United States[10]. Susceptible pea cultivars respond to the prevalent strain of the virus (formerly known as the severe strain of bean yellow mosaic virus[5,10,13], with prominent veinal chlorosis, apical and stem necrosis, followed by premature death[5,8,10,11,13]). Resistance to CYVV has been associated with that to BYMV[2,5,8,13] suggesting that the *mo* gene[15] would confer resistance to both viruses. However, recently we have determined that some pea lines of foreign introductions are resistant to CYVV, but susceptible to BYMV and vice versa. These findings indicated distinct genetic factors for resistance to these two viruses. This study concerns the inheritance of resistance to CYVV in *Pisum sativum* L. and its relationship to BYMV.

## Materials and Methods

Seed of pea cultivars were obtained from several commercial sources. Accessions of foreign lines were secured from the USDA Northeast Plant Introduction Station, Geneva, New York. Genetic populations were derived from the following crosses: a) BYMV+CYVV-resistant Bonneville with the BYMV+CYVV-susceptible Ranger; b) Bonneville with BYMV-resistant/CYVV-susceptible PI 391630 and PI 269818; and c) Bonneville or Ranger with BYMV-susceptible/CYVV-resistant PI 347492. Cultures of BYMV and CYVV were the same as those used in previous studies[10–12]. Inocula were prepared from Ranger plants infected with either one of these viruses, by homogenizing infected leaves with 0.05 M phosphate buffer ($K^+$) (*p*H 8.5). Extracts were rubbed onto the first two leaves of each plant previously dusted with Carborundum. To avoid escapes among susceptible genotypes, test plants were reinoculated on the third leaf. In screening for resistance, 16 to 20 plants of each pea line were inoculated with BYMV or CYVV. All plants, regardless of their reaction, were assayed for viral infection using Ranger pea, or by enzyme-linked immunosorbent assays (ELISA). Tests were conducted in an insect-free greenhouse that was maintained at 25–30 C.

## Results

*Pea lines resistant to BYMV and CYVV.* Ace, Aurora, Bonneville, Bridger, Canjoy, Canner 1281, Canner King, Canner Prince, Champ, Cascade, Climax, Dark Skin Perfection, Davis Perfection, Early Frost, Early Harvester, Early Perfection, Early Perfection 3040, Early Sweet 20, Early Sweet A45, Ericson Perfection, Eureka, Famous, Frazer, Freezer, Freezer 626, Freezer 640, Freezer 5147, Freezer 68178, Freezer 73152, Galaxie, Greater Progress, Greenfeast, Hundredfold, Hylate, Improved Surprise, Ivy, Jade, Knight, Laxton Progress, Laxton Superb, Little Marvel, Maestro, Mars, Medalist, Melody, Midway, Mini, Morse's 55, Midfreezer, Neptune, Pacific Perfection, Perfected Freezer, Perfected Freezer 60, Perfected Freezer 70A, Perfection 25, Perfection 42, Perfection 400, Premium Gem, Pride, Progress No. 9, Rally, Resistant Early Perfection 326, Rondo, Shoshone, Signet, Small Sieve Perfection, Sirod, Sparkle, Surprise, Sybo, Target, Trojan, Trumpet, Venus, Viking, Wando, Wisconsin Perfection, and numerous breeding lines. The following plant introductions also were resistant to these two viruses: PI 140295, PI 174319, PI 174924, PI 180669, PI 236493, PI 347420, PI 347422, and PI 356851.

*Pea lines resistant to CYVV, but susceptible to BYMV.* PI 193586 and PI 193835 (Ethiopia), PI 347464, PI 347465, PI 347466, PI 347467, and PI 347492 (India).

*Pea lines resistant to BYMV, but susceptible to CYVV.* PI 391630 (China) and PI 269818 (USSR).

*Inheritance studies.* Plants of the resistant parents Bonneville and PI 347492 remained free of local and systemic infection following inoculations with CYVV, thus they were considered highly resistant or immune to this virus. Conversely, those of the susceptible parents Ranger and PI 391630 developed a prominent foliar chlorosis that involved all veins and veinlets followed by stem streak, apical necrosis, and eventual death. Plants of $F_1$ (Bonneville × Ranger), (Bonneville × PI 391630), and (Ranger × PI 347492) reacted with systemic symptoms closely resembling those of the susceptible parents, indicating that resistance was inherited recessively. The reaction of $F_2$ populations of these crosses revealed a segregation close to the ratio of 3 susceptible plants to 1 resistant. Plants of the backcrosses (Bonneville × Ranger) × Ranger, (Bonneville × PI 391630) × PI 391630, and (Ranger × PI 347492) × Ranger were all susceptible, whereas those of (Bonneville × Ranger) × Bonneville, (Bonneville × PI 391630) × Bonneville, and (Ranger × PI 347492) × PI 347492 segregated approximately to the ratio 1 resistant to 1 susceptible. From the data presented in Table I, it is concluded that resistance to CYVV in Bonneville and PI 347492 is inherited monogenically recessive.

*Linkage between resistance factors.* Plants of each of 50 $F_3$ families deriving from (Bonneville × Ranger) $F_2$ were divided into groups of equal numbers and inoculated with BYMV or CYVV. The data in Table II show that 12 families were resistant to both viruses, 14 were susceptible, and 24 cosegregated for these two viruses. The absence of recombinations indicate that in Bonneville, the genes for resistance to BYMV and CYVV are closely linked.

*Evidence for two loci for CYVV resistance.* When $F_1$ plants resulting from the crosses between the CYVV-resistant Bonneville with CYVV-resistant PI 193586, PI 193835, PI 347464, PI 347465, PI 347466, PI 347467, or PI 347492 were inoculated with this virus, all plants were susceptible. In addition, plants of (Bonneville × PI 347492) $F_2$ segregated approximately in the ratio: 7 resistant to 9 susceptible. The data presented in Table III indicate the presence of two distinct loci for resistance to CYVV, one in Bonneville and another in PI 347492. The symbols *cyv* and *cyv*-2 (clover yellow virus) are assigned to these genes, respectively.

*Evidence for one locus for resistance to BYMV.* Plants of $F_1$, $F_2$, and reciprocal backcross populations of the crosses between BYMV+CYVV-resistant Bonneville and BYMV-resistant/CYVV-susceptible PI 269818 and PI 391630 were all resistant to BYMV. The data shown in Table IV demonstrate the existence in these lines of one locus for resistance to BYMV.

## Discussion

The identical reactions to BYMV and CYVV of a large number of domestic cultivars and several foreign introductions of *P. sativum* initially suggested a common gene for resistance to both viruses. This hypothesis was considered plausible, because these two viruses were, for years, believed to be strains of the same virus[5,10,13]. However, the finding of some lines resistant to CYVV, but susceptible to BYMV and vice versa, indicated the existence of two distinct genetic entities.

This study has shown that: a) resistance to CYVV in *P. sativum* is monogenically recessive; b) in Bonneville the gene *cyv* is closely

The author is professor in the Department of Plant Pathology, New York State Agricultural Experiment Station, Cornell University, Geneva, New York 14456.

linked to *mo*, known to be located on chromosome 2[6] and is linked to *Pgm-p*[14]; c) some lines from India (PI 347464, PI 347465, PI 347466, PI 347467, and PI 347492) and Ethiopia (PI 193586 and PI 193835) possess a second gene, *cyv*-2, that appears to be located in a different linkage group; and d) in BYMV-resistant/CYVV-Susceptible PI 269818 and PI 391630, and in Bonneville, resistance to BYMV is conditioned by the same gene (*mo*).

Published reports also have associated resistance to BYMV with resistance to other potyviruses such as pea mosaic virus (PMV)[1], watermelon mosaic virus 2 (WMV-2)[12], the lentil strain of pea seed-borne mosaic virus (PSbMV-L)[4], and to the NL-8 strain of bean common mosaic virus (BCMV-NL8)[9]. Bonneville and many of the BYMV+CYVV-resistant cultivars also are resistant to these viruses[1,9,12]. Thus, it appears that in most of BYMV-resistant cultivars, there is a cluster of closely linked loci that during breeding are transferred as a unit. Recently, the name linkat has been proposed for a such unit[3].

Preliminary studies have suggested that additional loci for resistance to the same strains of BCMV-NL8, PMV, and PSbMV-L, are present in other linkage groups (unpublished data). Demarly[3] has speculated that linkats may be ancestral genes that, in the course of evolution, gave rise to duplicates that in some cases, moved to other loci. This hypothesis accounts for the observed linkage on chromosome 2 and may explain the presence of additional resistance genes for the same strains of CYVV, BCMV-NL8, PMV, and PSbMV. Conversely, it could be hypothesized that mutant loci for recessive resistance to BYMV, BCMV-NL-8, CYVV, PMV, and PSbMV-L may have originated as independent mutations in different linkage groups and through translocations may have converged in chromosome 2. However, both hypotheses assume repetition of genes for resistance. No duplicate genes have been reported for resistance to viruses, but there are several cases of specific genes conferring resistance to specific strains or pathotypes of the same virus. Hence, *cyv* and *cyv*-2 may have been the result of two independent mutations conferring resistance to two distinct pathotypes of CYVV, but they cannot be differentiated by the strain used in this study. For example, in the tomato, *Tm*-2 and *Tm*-2$^2$ for resistance to tobacco mosaic virus cannot be differentiated if strain 0 and 1 are used. However, plants with *Tm*-2 are infected by strains 1.2 and 2, and those with *Tm*-2$^2$ only by strain 2[27].

Recent data, however, seem to indicate that

**Table I. Segregation ratios of cross and backcross populations of *Pisum sativum* lines resistant and susceptible to clover yellow vein virus**

| Cultivars and lines | No. plants resistant | No. plants susceptible | Exp. ratio | Goodness-of-fit *P* |
|---|---|---|---|---|
| Bonneville | 33 | 0 | | |
| Ranger | 0 | 35 | | |
| (Bonneville × Ranger)$F_1$ | 0 | 15 | | |
| (Bonneville × Ranger)$F_2$ | 86 | 268 | 1:3 | 0.76 |
| (Bonneville × Ranger)$F_1$ × Bonneville | 32 | 38 | 1:1 | 0.48 |
| (Bonneville × Ranger)$F_1$ × Ranger | 0 | 77 | | |
| PI 391630 | 0 | 16 | | |
| (Bonneville × PI 391630)$F_1$ | 0 | 11 | | |
| (Bonneville × PI 391630)$F_2$ | 20 | 64 | 1:3 | 0.80 |
| (Bonneville × PI 391630)$F_2$ × Bonneville | 15 | 20 | 1:1 | 0.42 |
| (Bonneville × PI 391630)$F_1$ × PI 391630 | 0 | 41 | | |
| PI 347492 | 16 | 0 | | |
| (Ranger × PI 347492)$F_1$ | 0 | 10 | | |
| (Ranger × PI 347492)$F_2$ | 44 | 120 | 1:3 | 0.61 |
| (Ranger × PI 347492)$F_1$ × PI 347492 | 36 | 41 | 1:1 | 0.59 |
| (Ranger × PI 347492)$F_1$ × Ranger | 0 | 61 | | |

**Table II. Reaction to bean yellow mosaic virus (BYMV) and clover yellow vein virus (CYVV) of 50 $F_3$ families of the cross Bonneville × Ranger**

| No. families | Viruses | No. plants resistant | No. plants susceptible | Exp. ratio | Goodness-of-fit *P* |
|---|---|---|---|---|---|
| 12 | BYMV | 174 | 0 | | |
| | CYMV | 171 | 0 | | |
| 24 | BYMV | 68 | 240 | 1:3 | 0.24 |
| | CYVV | 70 | 244 | 1:3 | 0.27 |
| 14 | BYMV | 0 | 194 | | |
| | CYVV | 0 | 198 | | |

**Table III. Reaction of $F_1$ and $F_2$ populations of the cross between clover yellow mosaic virus-resistant Bonneville with CYVV-resistant plant introductions from Ethiopia and India**

| Cultivars and PIs | No. plants resistant | No. plants susceptible | Exp. ratio | Goodness-of-fit *P* |
|---|---|---|---|---|
| (Bonneville × PI 193586)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 193835)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 347464)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 347465)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 347466)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 347467)$F_1$ | 0 | 6 | | |
| (Bonneville × PI 347492)$F_1$ | 0 | 12 | | |
| (Bonneville × PI 347492)$F_2$ | 63 | 81 | 7:9 | 0.14 |

**Table IV. Reaction of $F_1$ and $F_2$ populations from crosses between bean yellow mosaic virus-resistant lines**

| Cultivars and PIs | No. plants res. | No. plants susc. |
|---|---|---|
| Bonneville | 15 | 0 |
| PI 269818 | 16 | 0 |
| (Bonneville × PI 269818)$F_1$ | 12 | 0 |
| (Bonneville × PI 269818)$F_2$ | 104 | 0 |
| PI 391630 | 12 | 0 |
| (Bonneville × PI 391630)$F_1$ | 14 | 0 |
| (Bonneville × PI 391630)$F_2$ | 98 | 0 |
| Ranger | 0 | 16 |

resistance to BYMV, CYVV, and PMV is not strain- or pathotype-specific. Barnett (pers. comm.) demonstrated that the pea cultivar Greenfeast is resistant to a number of isolates of BYMV, CYVV, and PMV from Canada, Australia, and the USA although these isolates exhibited divergence by molecular hybridization (cDNA) and serology.

Work is in progress to elucidate the inheritance of resistance to BCMV-NL8, PMV, and PSbMV-L as well as several other points raised by this investigation.

### References


1. BARTON, D. W., W. T. SCHROEDER, R. PROVVIDENTI, and W. MISHANEC. Clones from segregating progenies of garden pea demonstrate that resistance to BV2 and PV2 is conditioned by the same genotype. *Plant Dis. Reptr.* 48:353–355. 1964.
2. BOS, L., K. LINDSTEN, and D. Z. MAAT. Similarity of clover yellow vein virus and pea necrosis virus. *Neth. J. Plant. Path.* 83:104–108 1977.
3. DEMARLY, Y. The concept of linkat. *Proc. Conf. Broadening Genet. Base Crops. Pudoc. Wageningen.* p. 257–265. 1979.
4. GOODELL, J. J. and R. O. HAMPTON. Interaction of *Pisum* genes *sbm* and *mo* with pea seedborne mosaic virus (PSbMV). Sympton expression and immunity to three variant strains. *Phytopathology* 73:789 (Abst.). 1983.
5. GROGAN, R. G. and J. C. WALKER. A pod-distorting strain of the yellow mosaic virus of bean *J Agr. Res.* 77:301–314. 1948.
6. MARX, G. A. and R. PROVVIDENTI. Linkage relations of *mo. Pisum Newsl.* 11:28–29. 1979.
7. PELHAM, J. Strain-genotype interaction of tobacco mosaic virus in tomato. *Ann. Appl. Biol.* 65:293–297. 1970.
8. PRATT, M. J. Clover yellow vein virus in North America. *Plant Dis. Reptr.* 53.201–212. 1969.
9. PROVVIDENTI, R., M. J. SILBERNAGEL, and W. Y. WANG. Local epidemic of NL-8 strain of bean common mosaic virus in bean fields of western New York. *Plant Disease* 68:1092–1094. 1984.
10. —— and W. T. SCHROEDER. Resistance in *Phaseolus vulgaris* to the severe strain of bean yellow mosaic virus. *Phytopathology* 63:196–197. 1973.
11. SCHROEDER, W. T. and R. PROVVIDENTI. Evaluating *Pisum sativum* for resistance to pea mosaic. *N.Y.S. Agr. Exp Sta. Bull. 806*, 10 pp. 1964.
12. —— and ——. Resistance to watermelon mosaic virus 2 in *Pisum sativum* conditioned by the gene for resistance to bean yellow mosaic virus. *Phytopathology* 60:1312–1313. 1970.
13. THOMAS, H. R. and W. J. ZAUMEYER. A strain of yellow bean mosaic virus producing local lesions on tobacco. *Phytopathology* 43:11–15. 1953.
14. WEEDEN, N. F., R. PROVVIDENTI, and G. A. MARX. An isozyme marker for resistance to bean yellow mosaic virus in *Pisum sativum. J. Hered.* 75:411–412. 1984.
15. YEN, D. E. and P. R. FRY. The inheritance of immunity to pea mosaic virus *Austr. J. Agr. Res.* 7:272–281. 1956.


The Journal of Heredity 78:128–129. 1987

# Brown and rust mutants of the Syrian hamster are *p* and *b* genes of mammalian coat colors

**Roy Robinson, C. V. Beechey, and A. G. Searle**

*ABSTRACT*: The mutant genes of the Syrian hamster, which were originally designated as brown (*b*) and rust (*r*), are shown by morphological and phenotypic criteria, as well as by linkage studies in the case of brown, to be homologous with pink-eyed dilution (*p*) and brown (*b*), respectively, two well established loci in the genetics of mammalian pigmentation. It is proposed that the two mutants be appropriately redesignated.

THE SYRIAN HAMSTER (*Mesocricetus auratus*) is a light-bellied agouti more auburn in color than the mouse or rat. The dorsum is overlaid with black tipped guard hairs while the under parts of the body are pale cream or off-white. The skin of the ears are black, and the eyes have dark brown irises and black pupils.

Nineteen mutant genes are known to affect color and pattern in the hamster, two of which have similar phenotypes. These are the recessive genes designated as brown (*b*)[3,4] and rust (*r*)[12]. The brown gene produces a bright orange-brown coat, overlaid with light colored guard hairs. The skin of the ears is light brown while the eyes have pale brown irises and red pupils; the overall effect is that of a red eye that darkens with age. The rust gene produces a similarly colored coat but of a duller tone. The skin of the ear is medium brown while the eyes have light brown irises and a dark pupil that glows dull red under bright illumination. Again, the eye color darkens with age.

## Materials and Methods

Hair samples were obtained from wild-type, rust, and brown golden hamsters, and, for comparison, from wild-type, brown (*b*) and pink-eyed dilute (*p*) mice. Hairs were mounted by a method based on that of Grüneberg[1]: 1) placed on slides thinly smeared with albumin; 2) covered with O.P.74 (95 percent ethyl alcohol) that was then allowed to dry on a hot-plate; 3) dried in an oven overnight and then passed through O.P.74 alcohol, 1:1 O.P.74 alcohol + xylene, xylene for at least 2, 1, and 1 days, respectively, in a vacuum; and 4) mounted in Euparal. They were then examined microscopically under oil immersion.

## Results

Figure 1 shows that there were marked differences in granule size and shape between brown in the hamster and brown in the mouse, although wild-type granules in the two species were a similar ovoid shape. Instead, brown granules in the hamster resembled those of pink-eyed dilution in the mouse (Figure 1*F*) which are very small and irregular in shape, with a tendency to form flocculent clumps and with reduced numbers in cortical cells[9]. The largest ones seemed to be only about 0.3 $\mu$m in diameter, compared with 1.2 $\mu$m for the long axis of the hamster wild-type granules. On the other hand, granules of the hamster rust mutant closely resembled those of the brown mouse (Figure 1*E*) in their spherical shape[9] but variable size, with an average diameter of about 0.8 $\mu$m.

The first indication that the brown gene may be misnamed was the discovery that it was linked to albino[7]. This finding invited comparison with the known linkages of albino (*c*) and pink-eyed dilution (*p*) genes in the deermouse, house mouse, and Norway rat[6]. The implication is that brown could be pink-eyed dilution. The red eye color is typical of this type of coat mutant although the coat color is brighter than that shown by pink-eyed dilution phenotypes in the other three species. However, this could be accounted for by the rich color of the wild-type agouti in the hamster. On the other hand, the crossover value for the hamster genes is 30.9 ± 1.8, which differs from the remarkably similar crossover values of 17.4 ± 2.5 (deermouse), 15.1 ± 0.3 (house mouse), and 18.4 ± 0.4 (Norway rat) of the other species[6], but agrees with the recently discovered linkage of 30 ± 5 for the *c* and *p* loci of the Mongolian gerbil (B. D. Leiper and R. Robinson, in press).

Linkage between brown and rust in the hamster also has been tested for, by intercrossing $F_1$ progeny of crosses between them. Phenotypes of $F_2$ offspring were 122 ++, 37 +*r* and 54 *b*+ or *b r*, with *b* epistatic over *r*; total 203. This is a very

The first author's address is: St. Stephens Road Nursery, Ealing, London W 13 8HB; the second and third authors' address is: Medical Research Council Radiobiology Unit, Chilton, Didcot, Oxon OX11 0RD England. They thank Dr. Mary F. Lyon for her critical comments and Dr. Dennis A. Stephenson for allowing them to quote his unpublished information.