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Inheritance of Morphologically Conditioned Floral Coloration in *Collinsia heterophylla*

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ABSTRACT

Violet band formation at the tip of the upper lip in flowers of *Collinsia heterophylla* Buist, 2N=14, (Scrophulariaceae) is controlled by multiple dominant alleles of the three recessive white flower (w_1, w_2, w_3) loci: W^v, narrow violet band; W^{kv} carmine-tinged violet band; and W^b, broad violet band extending to the base of the lip. The W^v, W^{kv}, and W^b alleles are recessive to the wild-type homologs W_1, W_2, and W_3. For the band to be expressed homozygosity at one band locus is required; presence of additional violet band alleles of the remaining two loci widens the band. Mutations at the keelless (*kl*) and superflower (*sf*) gene loci, occurring in the pre-reproductive or an early stage of floral ontogeny, condition the retrogressively altered flowers to express the violet (*W*), carmine (*Kn*) and other genes' phenotypes in floral parts where the wild-type genotype prevents their expression.

INTRODUCTION

The pentamerous, zygomorphic, sympetalous flowers of *Collinsia heterophylla* Buist (Scrophulariaceae), have a two-lobed upper lip and a three-lobed lower lip with two side lobes and a longitudinally folded middle lobe (keel) harboring four didynamous stamens and a two-carpellate pistil. The taxonomic literature describes the wild-type flowers of *C. heterophylla* as having a violet lower lip and a white upper lip (Abrams, 1951). According to Rasmuson (1920), two recessive genes (*a* and *b*) produce mutants having white flowers. Hiorth (1930,1933) confirmed their existence, labeled them white-1 (w_1) and white-2 (w_2), and added a third white flower gene (w_3) to the list. The w_1 is linked with the *F* locus (white vein and/or dots on cotyledons and/or leaves; Hiorth, 1930). Further genetic analyses (Goršič, 1957) agreed with Hiorth's interpretation of white flower inheritance in *C. heterophylla*, except regarding the existence of the third white flower gene (w_3) which has not been encountered. Hiorth's three-*W*-loci theory of violet flower inheritance acquired confirmation from another angle – namely the investigation of inheritance of violet band of flower's upper lip – reported below. Genetic analyses indicate that the violet band formation is controlled by specific dominant *W* alleles of the three white flower loci (w_1, w_2, w_3), and that the violet band is restricted to the tip of the upper lip of the morphologically wild-type, that is, zygomorphic flowers.

In the wild-type zygomorphic flowers the action of the *W* genes is restricted to the lower lip, so is the action of the gene *Ll*, producing colored midvein of the side lobes (Hiorth, 1930). Whereas, the action of the *Kn* gene, controlling carmine coloration, is restricted to the upper lip of the flower (Hiorth, 1930). In morphologically altered flowers of keelless (*kl*) and superflower (*sf*) mutants (Goršič, 1994) the phenotypic expressions of *W*, *Ll* and *Kn* genes appear to be shifted to other petals of the flower. This differential coloration of petals of upper versus lower lip in morphologically altered flowers has been investigated and the results are reported below.

Plants of this investigation were grown from seeds obtained from previous greenhouse cultures planted by the author. The cultivation and hybridization methods used in the investigative process have been described (Goršič, 1994). In calculating the fitness of segregation ratios (X^2), the Yates' correction factor has been applied for cultures with a population size below 50.

INHERITANCE OF VIOLET AND WHITE BAND OF THE UPPER LIP

The dominant *W* alleles of the three white flower loci w_1, w_2, and w_3, were assumed to have no effect on the coloration of flower's upper lip. Observation that in cultures segregating for violet (*W*) and white (*w*) flowering plants some individuals of the violet group had violet tipped upper lip (Fig. 1) and some members of the white flowered group had a white tipped upper lip (Fig. 2) suggested that this parallel behavior of violet and white bands in the upper lip may be explained by assuming that the specific *W* alleles produce the bands whose expression is inhibited in recessive flowers. The white band of the upper lip in white flowers may be white for the same reason the lower lip is white in *ww* genotype. The violet band gene has been labeled W^v, and its behavior in crosses was investigated.

Crosses between plants with flowers exhibiting the violet band at the tip of the upper lip, and the wild-type plants exhibiting no such band produced progenies whose flowers were devoid of violet band indicating that the violet band is a recessive trait. The selfings and intercrosses of hybrid plants without violet band and selfings of some plants with the violet band produced progenies whose individuals varied from having no violet band to plants with only faint specks of violet to having 2-4mm wide violet band on the upper lip (Table 1, Fig. 1). The variation in width of violet band was so great that the two *W* loci (W_1^v and W_2^v) could not provide sufficient number of genotypes to accommodate all observed phenotypic classes. Therefore, the three-*W*-loci hypothesis (W_1, W_2, W_3) of violet flower inheritance in *C. heterophylla*, suggested by Hiorth, appeared agreeable. By hybridization and selfing it has been established that for the violet band to be expressed the W^v gene must be homozygous at least at one locus (W^vW^v). The presence of additional W^v alleles of the remaining two *W* loci adds to the width and intensity of the band.

Because the W^v alleles of all three loci behave in the same manner the numbering them is dispensed of in the following summary relating upper lip phenotypes of *W* plants to corresponding genotypes below.

Genotypes	Upper lip (UL) phenotype	*kn*	*Kn*	Fig
1. *WWWWWW*, and heterozygotes involving one, two, or all three *w* alleles	No violet band	UL^{o}	Kn^{o}	1, 14
2. Single (W^vW), double (W^vWW^vW), or triple ($W^vWW^vWW^vW$) heterozygotes involving W^v and *W* or *w* alleles	No violet band or trace of violet	UL^{o}	Kn^{o}	1, 14
3. Homozygotes for W^v at any one locus (W^vW^vWWWW, etc.)	Faint violet band or violet patches	$\mathrm{UL}^{(v)}$	$Kn^{(v)}$	1,3
4. Homozygous at any one locus (W^vW^v) plus heterozygous (W^vW) at one or both remaining *W* loci	Violet band 1-2mm wide	$\mathrm{UL}^{v\text{-}1}$	$Kn^{v\text{-}1}$	1,3
5. Homozygotes at any two W^v loci, including heterozygotes at the third locus ($W^vW^vW^vW^vW^vW$, etc.)	Violet band 2-3mm wide	$\mathrm{UL}^{v\text{-}2}$	$Kn^{v\text{-}2}$	1,2
6. Homozygotes at all three W^v loci ($W^vW^vW^vW^vW^vW^v$)	Violet band 3-4 mm wide	$\mathrm{UL}^{v\text{-}3}$	$Kn^{v\text{-}3}$	2,3

The widths in millimeters (mm) are approximations because the violet band often fades gradually at the lower margin. Other factors affecting the intensity and/or width of the violet bands of the upper lip: (1) intensifier gene (*In*; Goršič, 1957), (2) upper lip colored gene (*0*; Hiorth, 1931; Goršič, 1957), (3) heterozygosity involving *w* genes, and (4) size of the flower (upper lip).

Penetrance of W^v alleles of all three loci varies slightly so that some heterozygotes (W^vW, W^vWW^vW, $W^vWW^vWW^vW$) may exhibit a trace of violet at the tip of the upper lip (UL^{tr}, scored in UL^{o} class). For this reason it was not always possible to distinguish with certainty the W^vW^vWWWW ($\mathrm{UL}^{(v)}$) from $W^vW^vW^vWWW$ ($\mathrm{UL}^{v\text{-}1}$), or $W^vW^vW^vW^vW^vW$ ($\mathrm{UL}^{v\text{-}2}$), from $W^vW^vW^vW^vW^vW^v$ ($\mathrm{UL}^{v\text{-}3}$). The genotypes of these plants having overlapping phenotypes can be ascertained by examining progenies of their selfings exhibiting either uniformity or segregation for the violet band. The fact that e.g., in progenies of selfings of $\mathrm{UL}^{(v)}$ plants, no $\mathrm{UL}^{v\text{-}3}$ plants have ever been observed, or that $\mathrm{UL}^{v\text{-}2}$ and $\mathrm{UL}^{v\text{-}3}$ plants when selfed never produced a plant without a violet band on the upper lip (UL^{o}) give credence to this interpretation of inheritance of the upper lip band (Table 1, 2).

In addition to the W^v alleles two more violet band alleles, W^{kv} and W^b, have been identified. (1) W^{kv}, producing carmine-tinged violet band (Fig. 1), behaves in the same manner as the W^v alleles, and in combination with them, particularly when in the homozygous state ($W^{kv}W^{kv}$), imposes a distinct carmine-tinge upon the band. Its presence is most drastically demonstrated in flowers with carmine (*Kn*) colored upper lip (see below). (2) W^b, producing in homozygous state (W^bW^b) when associated with homozygous W^vW^v and/or $W^{kv}W^{kv}$ a 4-5mm wide violet band extending laterally to the base of upper lip (Fig. 4, 6). The presence of one W^b allele in double and triple homozygotes for violet band genes appreciably increases the width of the band ($\mathrm{UL}^{v\text{-}4}$, Fig. 4); in all other combinations the W^b behaves as a W^v allele. The rare W^b allele is a member of w_2 locus, and is in most cases associated with white fleck gene (*Wf*; Goršič, 1957) affecting the folding of petals of the lower lip.

In white flowers (*w*) the violet band is expressed as a white band at the tip of bluish upper lip (Fig. 2, 6; Table 1). Due to the fewer number of dominant W^v, W^{kv} and W^b alleles in white flowers' genotypes - for w_1w_1 and w_2w_2 plants the maximum being four, and for $w_1w_1w_2w_2$ plants two, in contrast to six for violet (*W*) flowers - the variability in width of the white band is lower than for the violet band of violet flowers. The white band of the $w_1w_1W_2^bW_2^bW_3^vW_3^v$ white flower (UL^{v-4}) is a mirror image of violet band of $W_1W_1W_2^bW_2^bW_3^vW_3^v$ violet flower (UL^{v-4}). In white flowering plants, exhibiting pure white upper lip, the white band cannot be discerned.

The violet and white bands on the upper lips are expressed in plants exhibiting carmine (*Kn*) colored upper lip as well. Intricate interactions between W^v, W^{kv} and W^b genes and *Kn* gene produced an array of phenotypes. The width and intensity of violet band in carmine colored flowers (*Kn*/--) depends on the number, type, and zygosity status of the W^v alleles of the three *W* loci (Table 2). Three distinct phenotypes unique for carmine flowers exhibiting a violet band at the tip of the upper lip have been observed: (1) $Kn^{-t} = Kn^{(v),v-1}$, carmine with diluted tip (Fig. 3). The $W_1^vW_1^vW_2W_2W_3W_3Kn$/-- and any other single locus homozygote for violet band (except $W_2^bW_2^b$) carrying one extra W^v or W^{kv} apparently cannot produce sufficient concentration of pigment to be expressed as a distinct violet band – in old flowers some violet may appear in the diluted margin. (2) Kn^{vz}, the violet band separated from carmine by a white zigzag (z) line (Fig. 2, 3, 4). Depending on the number of W^v and/or W^b alleles in the genotype, the width of the band varies from 1-5mm: e.g. $Kn^{v-2z} = W_1^vW_1^vW_2^vW_2^vW_3W_3Kn$/--, $Kn^{v-3z} = W_1^vW_1^vW_2^vW_2^bW_3W_3Kn$/--, $Kn^{v-3z} = W_1^vW_1^vW_2^vW_2^vW_3^vW_3^v$, $Kn^{v-4z} = W_1^vW_1^vW_2^vW_2^bW_3^vW_3^v$, or $W_1^vW_1^vW_2^bW_2^bW_3^vW_3^v$. (3) Kn^{vn}, the violet band confluent with carmine (n, no zigzag separation line, Fig. 3, 4, 5): e.g. $Kn^{v-2n} = W_1^vW_1^vW_2^{kv}W_2^{kv}W_3W_3Kn$/--, $Kn^{v-3n} = W_1^vW_1^vW_2^vW_2^vW_3^{kv}W_3^{kv}Kn$/--, $Kn^{v-4n} = W_1^vW_1^vW_2^vW_2^bW_3^{kv}W_3^{kv}Kn$/--. Plants with these genotypes carrying intensifier gene (*In*) exhibited a particularly exaggerated confluent line (Fig. 3). In homozygous $W^{kv}W^{kv}$ individuals, the blending of the bands is complete, in heterozygotes, $W^{kv}W$, $W^{kv}W^v$, $W^{kv}W^b$, including heterozygotes involving *w* alleles, the blending is not complete and a weak demarcation line is discernible. The somewhat turbid separation line between violet and carmine, observed in flowers of some plants carrying no W^{kv} gene, is probably due to the coupling association of *Kn* and *0* (colored upper lip gene); *0Kn* combination produces violet-impregnated carmine upper lip (instead of pure carmine) giving the weak demarcation line a pinkish tinge. In some *Kn* plants carrying two or more W^{kv} genes the terminal part of the upper lip is diluted or devoid of all pigmentation (Kn^{v-2n-t}, Fig. 5), possibly due to a negative interaction between *Kn* and W^{kv} genes all of which enhance carmine pigment formation leading to overproduction and chemical imbalance (Straus, 1999).

Coloration of the upper lip in *C. heterophylla* and *C. tinctoria* Hartw. flowers are strikingly similar (Figs. 7, 8). It is likely that the intensely colored lower band of *tinctoria* flowers is a condensed carmine band (*Kn*), and the upper (wider band) corresponds to the violet band (W^v) of *heterophylla* flowers.

In white flowering plants with the carmine upper lip (w_1w_1Kn/--, w_2w_2Kn/--, and $w_1w_1w_2w_2Kn$/--) the width of the terminal white band (designated $Kn^{-t} = Kn^{(v),\ v-1,\ v-2,\ v-3,\ v-4}$) is comparable to the width of the violet band of violet flowering (*W*) plants having an equivalent gene content and zygosity status regarding W^v, W^{kv}, and/or W^b alleles (Fig. 2, 6; Table 2).

DISPLACEMENT OF PHENOTYPE IN MORPHOLOGICALLY ALTERED FLOWERS

The specific dominant alleles W^v, W^{kv}, and W^b producing violet bands in flowers of *C. heterophylla* act in the realm of the lower and upper lip in contrast to their wild-type counterparts W_1, W_2, and W_3 whose action is restricted exclusively to the lower lip of the flower. Similarly, the carmine (*Kn*) gene, controlling carmine pigmentation, is restricted to act only in the upper lip of bilaterally symmetric (zygomorphic) wild-type flowers (Fig. 2, 3, 5, 6, 14). These restrictions were apparently lifted in flowers of two recessive mutants, keelless (*kl*, kl^p) and superflower (*sf*) reported by Goršič (1994).

Dominant *Kl* gene controls folding of the middle lobe (keel) of the lower lip. The folded keel has no colored midvein (*ll*), is white and has a triangular yellow-tinged violet patch at the tip (in Fig. 9, keel manually opened). There are two recessive alleles of the *Kl* locus, *kl* and kl^p. In *kl*/*kl* plants the open keel assumes the appearance of side lobes of lower lip in shape and coloration (Fig. 10). In the *kl*-type of keelless flowers all three lobes of the lower lip exhibited the phenotype of the side lobes of non-modified flowers. In the folded keel (*Kl*/--), evidently the *Ll* gene producing colored midvein (Hiorth, 1930) and *W* genes producing violet coloration of side lobes of lower lip have been suppressed, but in the open keel of keelless mutants (*kl*/*kl*), the suppression of *Ll* and *W* genes has been eliminated (Fig. 10).

In keelless flowers of the kl^p mutants (p, pattern), the change in phenotype went a step further; in kl^p/kl^p and kl^p/kl mutants all three lobes of the lower lip exhibited the basic coloration pattern of the upper lip: e.g., flowers of $W_1^vW_1^vW_2^vW_2W_3W_3Kn/\text{--}kl^p/kl$ genotype exhibited carmine pigmentation and terminal violet band with a white demarcation line on all lobes of the modified lower lip (Fig. 10), the $W_1W_1W_2W_2W_3W_3kn/knkl^p/kl$ genotype produced flowers with bluish (nearly white) upper and lower lip (Fig. 11), and $W_1^vW_1^vW_2^vW_2^vW_3^vW_3Kn/Knkl^p/kl$ genotype produced flowers in which upper and lower lip lobes exhibited carmine and violet band separated by a white demarcation line (Fig. 12). In all mutants of the kl^p-type the altered lower lip was conditioned physiologically to allow carmine (*Kn*) and some other genes normally expressed in the upper lip only to be expressed in the lower lip as well (Fig. 10, 11, 12).

The most drastic departure from the zygomorphic pattern of flower formation was observed in the superflower (*sf*/*sf*) mutants. The terminal flowers of these mutants are composed of five or more keel-like petals exhibiting typical keel coloration (Fig. 14). In the keelless superflower double mutants (*sf*/*sf kl*/*kl*) all keels of the terminal flowers were modified into side lobes in shape and coloration, demonstrating a shift in phenotypic expression conditioned by morphological alteration of flower.

The highest degree of transformation from zygomorphy to actinomorphy exhibited flowers of keelless superflower double mutants of the kl^p-type. The pentamerous actinomorphic terminal flowers of keelless superflower double mutants ($W_1W_1W_2W_2W_3W_3kn/kn$ $kl^p/kl^psf/sf$) were composed of 5 sepals, 5 identical petals exhibiting the coloration pattern of the upper lip of non-modified lateral flowers, 5 stamens producing viable pollen, and a two-carpellate pistil (Fig. 13).

DISCUSSION

Alleles of the three *W* loci control the flower coloration in *C. heterophylla* as follows: the wild-type W_1, W_2, and W_3 alleles produce violet coloration of the lower lip, the dominant W^v, W^{kv}, and W^b alleles in specific combinations control the formation of violet lower lip and violet band on the tip of the upper lip, and the recessive alleles w_1, w_2, and w_3 suppress the formation of violet pigmentation in lower as well as upper lip.

The violet-mouth-corners trait (Fig. 3, 5, 10, 12) characterized by violet patches at the site of divergence of upper and lower lip is most likely controlled by the *W* genes as evidenced by the fact that in the white flowers (*w*) it is suppressed as all other violet traits controlled by the *W* genes are (Fig. 2, 6). In some cultures the violet patches of the upper and the lower lip mouth corner are associated, and in most cultures the mouth corner violet coloration appears together with the violet band of the upper lip, suggesting that the violet mouth corners and the violet band on the upper lip may be controlled by the same *W* genes. On the basis of genetic analyses to date, it may be stated that the w_1 and w_3 loci have three dominant alleles each: W_1, W_1^v, W_1^{kv}, and W_3, W_3^v, W_3^{kv} respectively, and the w_2 locus has four: W_2, W_2^v, W_2^{kv}, and W_2^b. Further genetic tests may either add additional *W* alleles controlling mouth corner (corolla tube) coloration (e.g., W^u and W^l - for upper vs. lower mouth corner) or rename the existing ones (e.g., W^{uv}, W^{ukv} – for violet mouth corner and upper lip violet band, and upper lip mouth corner and carmine-tinged upper lip violet band respectively).

Many species of flowering plants with drastically zygomorphic flowers have petals exhibiting differential pigmentation. In *C. heterophylla* wild-type flowers the expression of several genes (*W*, *Kn*, *Ll*, and others) is strictly reserved for specific petals. Gene mutations that alter the morphology of floral parts may lift these restrictions as exemplified by keelless (*kl*) and superflower (*sf*) mutants of *C. heterophylla*. These two mutations seem to demonstrate the "retrogressive evolution" from strict zygomorphy to less drastic zygomorphy to nearly actinomorphic configuration of flowers.

In *kl*-type keelless flowers (*kl*/*kl*) the opening of the keel, although equalizing morphologically the petals of the lower lip, did not suffice to revert their physiology to the level at which they would behave as members of an actinomorphic regime. In the keelless mutation brought about by a more potent kl^p gene, the alteration appears to have reached the level at which all petals of the lower and the upper lip behaved physiologically as being members of an actinomorphic flower, permitting a common phenotypic expression in all petals (Fig. 11, 12).

Flowers of keelless mutants (*kl*/*kl* and kl^p/kl^p) were morphologically zygomorphic but terminal flowers of the keelless superflower double mutants (kl^p/kl^p *sf*/*sf*) seem to have reverted fully to the actinomorphic flower configuration (Fig. 13). A *C. heterophylla* plant with this type of pentamerous flower in the lateral position would be a worthy representative model of ancient presumably actinomorphic *Protocollinsia.*

Whether the mechanisms controlling various expressions of numerous *W* alleles and other genes, and mechanisms restricting gene actions in floral development are transcriptional involving cytokinins, methylation types and/or transposable elements (Kunze et al., 1997), or epigenetic, that is, posttranscriptional involving various RNA silencing pathways, remains to be answered (Voinnet, 2002; Zamore, 2002).

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Table 1. Phenotypic ratios of progenies of selfings segregating for violet band (W^v, W^{kv}, W^b) and white band of the upper lip (UL) of flowers of *Collinsia heterophylla*. UL^o = no violet band; UL$^{(v)}$ = faint violet band; UL^{v-1}, UL^{v-2}, UL^{v-3}, UL^{v-4} = 1, 2, 3, 4 mm wide violet band in violet flowers (*W*) or white band in white flowers (*w*). Single, double, and triple heterozygotes may exhibit a trace of violet (UL^{tr} included in UL^o). Expected number of individuals are in parentheses.

Genotype	Phen	UL^o		UL$^{(v)}$		UL^{v-1}		UL^{v-2}		UL^{v-3}		UL^{v-4}		Total	X^2	P
		W	*w*	*W*	*w*	*W*	*w*	*W*	*w*	*W*	*w*	*W*	*w*			
$W_1^vW_1W_2W_2W_3W_3$	UL^o	24(27)		12(9)										36(36)	0.925	.3
$W_1^vW_1W_2^vW_2^vW_3W_3$	UL^{v-1}			21(19.25)		56(57.75)								77(77)	0.212	.5
$W_1^vW_1W_2^vW_2W_3W_3$	UL^o	27(28.69)		8(6.37)		13(12.75)		3(3.19)						51(51)	0.531	.9
$W_1W_1W_2^vW_2^vW_3^vW_3^v$	UL^{v-2}							81(81)						81(81)	---	--
$W_1^vW_1W_2^vW_2W_3^vW_3^v$	UL^{tr}	38(51.89)		8(5.76)		53(46.13)		5(5.76)		19(13.45)				123(123)	8.006	.1
$W_1W_1W_2^vW_2W_3^{kv}W_3^{kv}$	UL^{v-1}			23(19.25)		39(38.5)		15(19.25)						77(77)	1.674	.7
$W_1^vW_1W_2^bW_2W_3W_3$	UL^o	24(20.81)		2(2.31)				4(6.94)				7(6.94)		37(37)	1.204	.7
$W_1^vW_1W_2^vW_2W_3^vW_3$	UL^o	14(15.61)		4(1.73)		15(13.88)		2(1.73)		2(4.05)				37(37)	2.508	.5
$W_1^vW_1^vW_2^bW_2W_3^{kv}W_3$	UL^{v-2}			3(2.81)		5(5.63)		7(8.44)		15(16.87)		15(11.25)		45(45)	1.156	.8
$W_1^vW_1W_2^bW_2W_3^{kv}W_3^{kv}$	UL^{v-1}			5(2.94)		8(5.88)		8(8.81)		12(11.75)		14(17.62)		47(47)	1.835	.7
$W_1^vw_1W_2^bw_2W_3W_3$	UL^o	16(12)	18(15)		1(3)			7(6)				4(9)	2(3)	48(48)	4.560	.5
$W_1^vw_1W_2^{kv}W_2W_3W_3$	UL^{tr}	14(16.12)	9(8.06)	4(2.69)	2(2.69)	9(10.75)		5(2.69)						43(43)	1.804	.8
$W_1^vw_1W_2^bw_2W_3^{kv}W_3$	UL^o	17(14.25)	24(17.81)				4(10.69)	10(7.13)		7(9.5)		8(11.87)	6(4.75)	76(76)	10.268	.2

Table 2. Phenotypic ratios of progenies for carmine (*Kn*), non-carmine (*kn*), and violet band (W^v, W^{kv}, W^b) or white band of the upper lip (UL) of flowers of *Collinsia heterophylla*. Kn^o, UL^o = no violet band; $Kn^{(v)}$, $UL^{(v)}$ = faint violet band; Kn^{v-1}, Kn^{v-2}, Kn^{v-3}, Kn^{v-4} = 1, 2, 3, 4 mm wide violet band in violet flowers (*W*) or white band in white flowers (*w*). Kn^{tr} = trace of violet, $Kn^{(v)}$ appears a Kn^{-t}. Expected number of individuals are in parentheses.

Genotype	Phen		Kn^oUL^o		$Kn^{(v)}UL^{(v)}$		$Kn^{v-1}UL^{v-1}$		$Kn^{v-2}UL^{v-2}$		$Kn^{v-3}UL^{v-3}$		$Kn^{v-4}UL^{v-4}$		Total	X^2	P
			W	*w*	*W*	*w*	*W*	*w*	*W*	*w*	*W*	*w*	*W*	*w*			
$W_1W_1W_2^vW_2^vW_3^vW_3^vKn/Kn$	Kn^{v-2}								25(25)						25(25)	---	--
$W_1W_1W_2^{kv}W_2W_3W_3Kn/Kn$	Kn^o		41(42.75)		16(14.25)										57(57)	0.285	.7
$W_1^vW_1^vW_2^{kv}W_2W_3^{kv}W_3Kn/Kn$	Kn^{v-1}				6(2.56)		21(20.5)		13(15.38)		1(2.56)				41(41)	4.792	.3
$W_1^vW_1^vW_2^vW_2W_3^{kv}W_3Kn/Kn$	Kn^{v-1}				3(2.44)		21(19.5)		13(14.62)		2(2.44)				39(39)	0.137	.98
$W_1^vW_1^vW_2^vW_2^vW_3^{kv}W_3^{kv}Kn/Kn$	Kn^{v-3}										18(18)				18(18)	---	--
$W_1^vw_1W_2^vW_2^vW_3^{kv}W_3Kn/Kn$	Kn^{v-1}					0(1.69)	14(10.12)	2(3.38)	4(5.06)	1(1.69)	6(5.06)				27(27)	2.314	.8
$W_1^vW_1W_2^vw_2W_3^{kv}W_3^{kv}Kn/Kn$	Kn^{v-1}					0(1.44)	11(8.63)	4(2.87)	6(7.18)	1(1.44)	1(1.44)				23(23)	1.220	.95
$W_1^vw_1W_2^vW_2^vW_3^{kv}W_3Kn/Kn$	Kn^{v-1}					0(1.31)	12(7.88)	5(2.63)	3(6.56)	1(1.31)	0(1.31)				21(21)	5.418	.5
$W_1^vw_1W_2^vW_2^vW_3^{kv}W_3Kn/Kn$	Kn^{v-1}					0(1.81)	15(10.87)	2(3.63)	9(9.07)	2(1.81)	1(1.81)				29(29)	2.564	.8
$W_1^vW_1^vW_2^vW_2W_3^{kv}W_3Kn/kn$	Kn^{v-1}	*Kn*			2(2.90)		12(11.63)		24(29.06)		4(2.91)				62(62)	11.250	.1
		kn			1(0.97)		9(7.75)		6(5.81)		4(0.97)						
$W_1^{kv}w_1W_2^vW_2W_3W_3Kn/Kn$	Kn^o	*Kn*	9(7.6)	4(3.79)	3(1.27)	0(1.26)	5(5.06)		1(1.27)						27(27)	3.065	.95
		kn	4(2.53)			0(1.26)	1(1.69)		0(1.27)								
$W_1^vw_1W_2^{kv}w_2W_3W_3Kn/kn$	Kn^o	*Kn*	9(6.37)	10(7.97)		1(3.19)	6(6.38)		3(1.59)						34(34)	7.201	.5
		kn		3(2.66)		0(1.06)	2(4.25)		0(0.53)								
$W_1^vW_1W_2^bW_2W_3^{kv}W_3Kn/kn$	Kn^{tr}	*Kn*	7(9.18)		2(0.68)		4(2.72)		4(2.04)		2(2.04)		4(5.09)		29(29)	4.071	.95
		kn	1(1.02)		1(0.45)		2(2.27)		2(0.91)		0(0.79)		0(1.81)				
$W_1^vw_1W_2^bW_2^vW_3^{kv}W_3^{kv}Kn/kn$	Kn^{v-2}	*Kn*			0(1.5)	0(1.5)	5(3)	1(3)	5(6)	1(1.5)	4(3)		4(6)		32(32)	6.372	.9
		kn			4(1.5)	1(0.5)		2(1)	3(2)		1(1)		0(1.5)	1(0.5)			

Figures 1-8. Phenotypes and genotypes of the upper lip (UP) coloration of *Collinsia*. Figs. 1-6 *C. heterophylla*. Figs. 7-8 *C. tinctoria*.

Fig 1. Left to right: $\mathrm{UL}^0 = W_1^vW_1W_2^vW_2W_3W_3$, $\mathrm{UL}^{(\mathrm{v})} = W_1^vW_1^vW_2W_2W_3W_3$, $\mathrm{UL}^{\mathrm{v}\text{-}1} = W_1^vW_1^vW_2^vW_2W_3^vW_3$, $\mathrm{UL}^{\mathrm{v}\text{-}2} = W_1^vW_1^vW_2W_2W_3^{kv}W_3^{kv}$.

Fig 2. $Kn^{\mathrm{v}\text{-}2} = w_1w_1W_2^vW_2^vW_3^vW_3^vKn$-(left), $Kn^{\mathrm{v}\text{-}3\mathrm{z}} = W_1^vW_1^vW_2^vW_2^vW_3^vW_{3v}Kn$- (right).

Fig 3. $Kn^{\mathrm{v}\text{-}1} = W_1^vW_1^vW_2^vW_2W_3^vW_3Kn$- (left), $Kn^{\mathrm{v}\text{-}3\mathrm{n}} = W_1^vW_1^vW_2^vW_2^vW_3^{kv}W_3^{kv}Kn\text{-}InIn$ (middle), $Kn^{\text{-}\mathrm{t}}(Kn^{(\mathrm{v})}) = W_1^vW_1^vW_2W_2W_3W_3Kn$- (right).

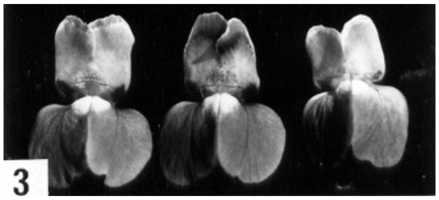

Fig 4. $Kn^{\mathrm{v}\text{-}4\mathrm{z}} = W_1^vW_1^vW_2^bW_2^vW_3^vW_3^vKn$- (left), $\mathrm{UL}^{\mathrm{v}\text{-}5} = W_1^vW_1^vW_2^bW_2^bW_3^vW_3^vknkn$ (middle), $Kn^{\mathrm{v}\text{-}4\mathrm{n}} = W_1^vW_1^vW_2^vW_2^bW_3^{kv}W_3^{kv}Kn$- (right).

Fig 5. $Kn^{\mathrm{v}\text{-}2\mathrm{n}\text{-}\mathrm{t}} = W_1^vW_1^vW_2W_2^{kv}W_3^{kv}W_3^{kv}Kn$-.

Fig 6. $Kn^{\mathrm{v}\text{-}4} = w_1w_1W_2^bW_2^bW_3^vW_3^vKn$-.

Fig 7. Upper lip with carmine-red and violet band.

Fig 8. Upper lip with carmine-red and white band.

Figures 9-14. Keelless *(kl,kl^p)* and superflower (*sf*) mutants of *Collinsia heterophylla*.

Fig 9. Flower with manually opened keel.

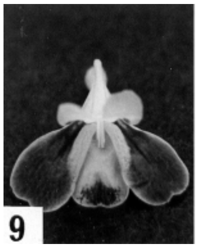

Fig 10. $UL^{(v)} = W_1^vW_1^vW_2W_2W_3W_3knkn$ *klkl* (left), $Kn^{v-1} = W_1^vW_1^vW_2^vW_2W_3W_3$ $KnKnkl^pkl^p$ (right).

Fig 11. $UL^o = W_1W_1$ $W_2W_2W_3W_3knkn$ kl^pkl.

Fig 12. $Kn^{v-2} = W_1^vW_1^vW_2^vW_2^vW_3^vW_3KnKn$ kl^pkl.

Fig 13. Pentamerous, actinomorphic terminal flower of a keelless superflower double mutant: $UL^o = W_1W_1W_2W_2W_3W_3$ $knknkl^pkl^psfsf$.

Fig 14. Superflower $W_1^vW_1W_2W_2W_3KnKn$ *sfsf* (Kn^0) mutant with terminal actinomorphic flower made of keels only, and lateral zygomorphic flowers.