# ALLELIC NEGATIVE COMPLEMENTATION AT THE ABRUPTEX LOCUS OF *DROSOPHILA MELANOGASTER*

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Manuscript received September 3, 1974
Revised copy received February 24, 1975

ABSTRACT

The mutations of the Abruptex locus in *Drosophila melanogaster* fall into three categories. There are recessive lethal alleles and viable alleles. The latter can be divided into suppressors and nonsuppressors of Notch mutations. The recessive lethals are lethal in heterozygous combination with Notch. As a rule the recessive lethals are lethal also in heterozygous combination with the viable alleles. Heterozygous combinations of certain viable alleles are also lethal. In such heterozygotes, one heteroallele is a suppressor of Notch and the other is a nonsuppressor. Other heterozygous combinations of viable alleles are viable and have an Abruptex phenotype. The insertion of the wild allele of the Abruptex locus as an extra dose (carried by a duplication) into the chromosomal complement of the fly fully restores the viability of the otherwise lethal heterozygotes if two viable alleles are involved. The extra wild allele also restores the viability of heterozygotes in which a lethal and a suppressor allele are present. If, however, a lethal and a nonsuppressor are involved, the wild allele only partly restores the viability, and the effect of the wild allele is weakest if two lethal alleles are involved. It seems likely that of the viable alleles the suppressors of Notch are hypermorphic and the nonsuppressors are hypomorphic. The lethal alleles share properties of both types, and are possibly antimorphic mutations. It is suggested that the locus is responsible for a single function which, however, consists of two components. The hypermorphic mutations are defects of the one component and the hypomorphic mutations of the other. In heterozygotes their cumulative action leads to decreased viability. The lethal alleles are supposed to be defects of the function as a whole. The function controlled by the locus might be a regulative function.

GENETIC complementation is of fundamental importance in the definition of genes and the analysis of gene function. Complementation is defined as the complementary action (cooperation) of homologous sets of genetic material involving the interaction of mutant genes, or their products, in double mutants. Those combinations that result in marked improvement in the function under study or in the development of a character which cannot be realized by the individual action of single mutants are said to complement each other. (FINCHAM 1966; RIEGER, MICHAELIS and GREEN 1968). In particular the phenomenon of intracistronic complementation, i.e. complementation between heterozygous pairs of mutations which are on the basis of other criteria mutations within the same cistron, is a central phenomenon in the analysis of functioning of genes.

The opposite of complementation, i.e. the impairment of function due to interaction of homologous sets of genetic material, is called negative complementation. Negative complementation is the enhancement of mutant phenotype in a heterozygote beyond that exhibited by either homozygote. There are some examples of negative complementation in Neurospora (FINCHAM 1966; SUNDARAM and FINCHAM 1967), and the phenomenon seems to be rather common in yeast (ZIMMERMANN and GUNDELACH 1969). In higher eukaryotes the examples of negative complementation are scanty. Recent findings at the Abruptex locus of *Drosophila melanogaster* by FOSTER (1972) and PORTIN and RUOHONEN (1972), however, demonstrate a dramatic example of allelic negative complementation. Certain heterozygous combinations of homozygous viable Abruptex alleles are namely lethal. The present study demonstrates the basic properties and regularities of allelic negative complementation at the Abruptex locus of *Drosophila melanogaster*.

## MATERIALS AND METHODS

*Abruptex mutants*: The Abruptex (*Ax*, 1-3.0) locus is sex-linked and belongs to the Notch (*N*, 1-3.0) pseudoallelic series on the basis that Abruptex mutations map within the limits of the Notch gene (LINDSLEY and GRELL 1968; WELSHONS 1971; FOSTER, personal communication). Homozygous Abruptex flies have shortened L5 veins; usually also L4, and L2, and sometimes L3 are shortened. Wings are also shortened and arched. The mutations act as dominants, although they have a weaker expression in heterozygous condition (usually only 5th vein is shortened).

Abruptex mutations used in this study were classified as recessive lethals and viables. The lethal alleles were $Ax^{59b3.1}$ and $Ax^{59d5}$, subsequently designated $Ax^{59b}$ and $Ax^{59d}$ or simply *59b* and *59d* respectively. The mutants were obtained from PROF. W. J. WELSHONS (Ames, Iowa); they were induced by irradiation (WELSHONS 1971). In the stocks used they were coupled with white-apricot ($w^a$, 1-1.3) and balanced with *In(1)dl-49, y Hw* $m^2$. The viable alleles used were $Ax^{28}$, $Ax^{E2}$, $Ax^{16172}$, $Ax^{9B2}$, and $Ax^{71d}$ subsequently often designated *28*, *E2*, *16172*, *9B2*, and *71d* respectively. The $Ax^{28}$ stock was received from The Division of Biology, California Institute of Technology (Pasadena), $Ax^{E2}$, $Ax^{16172}$, and $Ax^{9B2}$ were courtesy of PROF. WELSHONS. $Ax^{28}$ is a spontaneous mutation, $Ax^{E2}$, $Ax^{16172}$, and $Ax^{9B2}$ are EMS induced (FOSTER 1972), and $Ax^{71d}$ was induced in our laboratory by X-rays (1000 r). $Ax^{9B2}$ is female sterile, the others are fertile in both sexes, although $Ax^{16172}$ is less so in females than males. $Ax^{9B2}$ was balanced with an attached-*X* chromosome, *y w f*, in all stocks used, and so was also $Ax^{16172}$ in some stocks. In certain experiments the viable *Ax*-alleles were coupled with white-eosin ($w^e$, 1-1.3).

*Experimental procedure*: To test the viability of the heterozygous combinations of viable Abruptex mutations, the mutant stocks were simply intercrossed, and the sex ratio of the progeny revealed the viability of the heterozygotes. When viables were tested against the lethals (*59b* and *59d*), females carrying the viable allele in coupling with $w^e$ were crossed to $w^a$ $Ax^{59}$; *Dp(1;2)51b* males. In the case of the female sterile mutation, *9B2*, the cross was $w^a$ $Ax^{59}$/ *In(1)dl-49, y Hw* $m^2$ × $w^e$ $Ax^{9B2}$; *Dp(1;2)51b*. The relative frequency of eosin-eyed females revealed the viability of the heterozygous females. When the interaction of lethal mutations was tested, the mating was $w^a$ $Ax^{59}$/*Basc* × $w^a$ $Ax^{59}$; *Dp(1;2)51b*, and the frequency of apricot, not-Bar females in the progeny revealed the viability of the heterozygotes.

Wild alleles of Abruptex, and white, and Notch are present in the duplication *Dp(1;2)51b* in which a short piece of the *X* chromosome is inserted into the 2nd chromosome. The *X* chromosome bands carried by the duplication are 3C1-2–3D6-7, with white$^+$ located in the 3C2 band and Abruptex$^+$ in the 3C7 band. If intercrosses of viable mutations indicated lethality of the heterozygous females, a second cross was made, in which the male parent carried the duplication. If the duplication can fully restore the viability of the females, a 1 : 2 ratio of

females and males is to be expected. Thus, again the sex ratio of the progeny directly revealed the viability of the heterozygotes. Also the effect of the duplication on the phenotype and viability of heterozygous females which were viable was tested. In these crosses the Abruptex mutations were coupled with white-eosin, and thus the comparison of the phenotypes and numbers of eosin-eyed and wild-eyed females in the progeny revealed the effect of the duplication. The effect of the duplication on the viability of females carrying a lethal allele in one *X* chromosome and a viable allele in the other was studied in the same crosses as the viability of the females without the duplication; the number of not-eosin females as compared to the number of eosin males directly revealed the effect of the duplication. In the case of *9B2*, however, an indirect conclusion was made on the basis of the frequency of not-eosin females. In the progeny of intercrosses of lethals, the duplication-carrying females were not-apricot, not-Bar in phenotype, and their viability was calculated by comparing their frequency with the frequency of *59/Basc* and *59/Basc; Dp* females.

According to FOSTER (1972) negatively complementing viable Abruptex alleles have different effects on the wing-nicking phenotype of the Notch mutants. The effect of the Abruptex mutations on the wing-nicking effect of two Notch mutations was studied. They were $Df(1)N^{8}$ and $N^{55e11}$. The former is associated with a deficiency of 18 bands but the latter is not associated with any visible deficiency. The effects of the viable Abruptex alleles on Notch were studied in the crosses $Df(1)N^{8}/In(1)dl\text{-}49$, *y Hw* $m^{2}$ × *Ax/Y* and $N^{55e11}/In(1)dl\text{-}49$, *y Hw* $m^{2}$ × *Ax/Y*. Control crosses using wild males were also made. The effects of lethal Abruptex alleles on Notch were studied in the crosses $w^{a}$ $Ax^{59}/Basc$ × $w^{a}$ $N^{55e11}$; *Dp(1;2)51b* and $w^{a}$ $Df(1)N^{8}/Basc$ × $w^{a}$ $Ax^{59}$; *Dp(1;2)51b* in which the phenotypes of apricot, not-Bar females revealed the effect of these Abruptex mutations on Notch. All crosses were made as single female cultures on the standard food medium (consisting of semolina, syrup, agar-agar and both dried and fresh yeast) at 25°. Virgin females were put with three males into 50 ml culture bottles for four days, then the parent flies were transferred to fresh bottles for two days, and then discarded. Thus, progeny from the first six days were collected.

## RESULTS

*Interaction of Abruptex and Notch mutations*: Two of the viable Abruptex alleles, namely *28* and *9B2*, suppressed the wing-nicking effect of both $Df(1)N^{8}$ and $N^{55e11}$. Two of them, namely *E2* and *16172*, enhance the expression of both Notch mutations, and one, namely *71d*, has a neutral effect on Notch (the same effect as the wild allele). The suppressors of Notch have a very weak phenotypic expression when heterozygous with Notch mutations. Of the non-suppressors *71d* and *16172* have a clear wing venation Abruptex phenotype when heterozygous with Notch but *E2* has a weak expression.

Lethal Abruptex alleles are lethal in heterozygous combination with both $Df(1)N^{8}$ and $N^{55e11}$ (cf. WELSHONS 1971).

*Interaction of viable Abruptex alleles*: The results from the intercrosses of flies carrying different viable Abruptex alleles are given in Table 1. As shown in the table, *E2/71d*, *16172/71d*, and *28/9B2* heterozygous females are fully viable (as viable as males from the same cross). These females show an Abruptex phenotype. Thus, these alleles do not exhibit either complementation or negative complementation. Females of the *E2/28* genotype are semilethal; they have a viability of 61% as compared to the *28/Y* males, and of 50% as compared to the *E2/Y* males. These females have a strong Abruptex phenotype. Thus, this allele pair exhibits negative complementation as judged by the phenotype and the viability. It appears further from the table that *71d/28*, *71d/9B2*, and *28/16172*

TABLE 1

*Results of crosses between different viable Abruptex mutants*

| Cross | Progeny | | | $X^2$ (exp. 1:1) | Viability of females as compared to males |
|---|---|---|---|---|---|
| | females | males | Total | | |
| $Ax^{E2} \times Ax^{71d}$ | 918 | 987 | 1905 | 2.50 | 100% |
| $Ax^{71d} \times Ax^{16172}$ | 1107 | 1154 | 2261 | 0.98 | 100% |
| $Ax^{28} \times Ax^{9B2}$ | 191 | 165 | 356 | 1.91 | 100% |
| $Ax^{28} \times Ax^{E2}$ | 272 | 444 | 716 | 41.32* | 61% |
| $Ax^{E2} \times Ax^{28}$ | 345 | 691 | 1036 | 115.56* | 50% |
| $Ax^{71d} \times Ax^{28}$ | 3 | 438 | 441 | | 0.7% |
| $Ax^{71d} \times Ax^{9B2}$ | 0 | 367 | 367 | | 0% |
| $Ax^{28} \times Ax^{16172}$ | 0 | 155 | 155 | | 0% |

* significant at the 01% level.

females are lethal, i.e. these allele pairs exhibit a strong negative complementation. The lethal crisis is late pupal since several partly eclosed female pupae were found on the walls of the bottles. Sometimes these females succeed in eclosion but they die shortly after it. These dying females have a very strong Abruptex phenotype with practically no hairs and veins on the wings and no hairs on the thorax. FOSTER (1972) has shown that *E2/9B2* and *16172/9B2* females are also lethal, but that *E2/16172* females are viable.

It appears that the interaction of viable Abruptex alleles follows a certain rule: Those alleles which have similar effect on Notch neither complement each other nor exhibit negative complementation (the Abruptex alleles being classified as suppressors and nonsuppressors of Notch). On the other hand, those alleles which have opposite effects on Notch exhibit negative complementation. This principle is the same as that observed by FOSTER (1972). In details, however, the present results are somewhat different from those of FOSTER. FOSTER observed that all suppressor/enhancer combinations he studied are lethal, and on the other hand all lethal combinations are suppressor/enhancer combinations. In the present material, however, some lethal combinations, namely *28/71d* and *9B2/71d*, are suppressor/neutral-allele combinations, and one suppressor/enhancer combination, namely *E2/28*, is semilethal. Note, that *E2* is different from the other nonsuppressor alleles in the sense that it has only a weak expression in heterozygous combination with Notch.

The effect of *Dp(1;2)51b*—a duplication which carries the wild allele of the Abruptex locus—on the viability of otherwise-lethal females is shown in Table 2. It appears from the results that the duplication completely restores the viability of *28/71d*, *71d/9B2*, *28/16172*, and *E2/9B2* females, and that *16172/9B2; Dp* females have a viability of 82% as compared to *16172/Y* males. All these females show a clear Abruptex phenotype. Thus, a single dose of the wild allele of the Abruptex locus can usually fully eliminate the lethality caused by negative complementation of viable mutant Abruptex alleles. However, the wing-venation phenotype of the mutant alleles is strong despite the wild allele.

TABLE 2

*Effect of duplication,* Dp(1;2)51b, *on the viability of lethal Abruptex heterozygotes*

| Cross duplication in the male parent | Progeny | | | $X^2$ (exp. 1:2) | Viability of females with the duplication as compared to males |
|---|---|---|---|---|---|
| | females | males | Total | | |
| $Ax^{28} \times Ax^{71d}$ | 157 | 304 | 461 | 0.11 | 100% |
| $Ax^{71d} \times Ax^{9B2}$ | 450 | 937 | 1387 | 0.49 | 100% |
| $Ax^{28} \times Ax^{16172}$ | 120 | 291 | 411 | 3.16 | 100% |
| $Ax^{E2} \times Ax^{9B2}$ | 360 | 808 | 1168 | 3.45 | 100% |
| $Ax^{16172} \times Ax^{9B2}$ | 296 | 718 | 1014 | 8.10* | 82% |

Females from the two last crosses without the duplication have been found to be lethal by FOSTER (1972).

* significant at the 1% level.

The effect of the duplication on otherwise viable or semilethal homo-and heterozygous Abruptex-combinations is presented in Table 3. It appears that the duplication neither improves nor impairs the viability of these genotypes. The wing-venation phenotype of the females carrying two Abruptex mutations, which are suppressors of Notch, and the duplication, is weak Abruptex (at most only 5th vein shortened), whereas those females which carry two nonsuppressor mutations and the duplication have a clear Abruptex phenotype (3rd, 4th, and 5th veins shortened). Also the females with the *28/E2; Dp* genotype have a clear Abruptex phenotype.

*Interaction of viable and lethal Abruptex alleles*: The interaction of viable and recessive lethal Abruptex alleles regularly causes lethality (Table 4): All the viable/lethal heterozygotes are lethal except that *9B2/59b* and *9B2/59d* females are semilethal. Thus, as the result of the interaction between lethals and viables, the recessive lethality of the *59b* and *59d* alleles usually converts into dominant lethality.

The duplication, *Dp(1;2)51b*, which carries the wild allele of the Abruptex locus, restores completely the viability of females which carry a lethal and a suppressor-of-Notch allele in their X chromosomes. (Table 4). The viability of the lethal/non-suppressor females reaches at most the level of subvitality with the aid of the duplication. The genotypes *59b/16172* and *59d/16172* reach a viability of 6.8% and 3.3% only when they carry the duplication (Table 4). The wing-venation phenotype of the females carrying a lethal allele and a viable allele, and the duplication is strong Abruptex.

*Interaction of lethal Abruptex alleles*: The recessive lethal Abruptex alleles are always lethal in heterozygous combination with each other (Table 5). The wild allele of the locus carried by the *Dp(1;2)51b* restores the viability of homo- and heterozygous $Ax^{59}$ females only to the level of semilethality (viabilities are between 12% and 34%) (Table 5). Because of the reduced viability of *Basc; Dp* males there was doubt whether the comparison of the number of *59/59; Dp* females to the number of *59/Basc* and *59/Basc; Dp* females gives a reliable estimate of the viability of the former females. Therefore, control crosses with

TABLE 3

*Effect of the duplication,* Dp(1;2)51b, *on the viability of otherwise viable or semilethal Abruptex homo- and heterozygotes*

| Cross (duplication in the male parent) | Progeny | | | | | | Comparison of the numbers of all the males and of females with the duplication (not-eosin females), and the viability of these females | |
|---|---|---|---|---|---|---|---|---|
| | females | | | males | | | | |
| | | eosin | not eosin | eosin | not eosin | | $X^2$ (exp. 2:1) | viability |
| $w^e\ Ax^{E2} \times w^e\ Ax^{23}$ | | 138 | 251 | 252 | 184 | | 3.22 | 100% |
| $w^e\ Ax^{E2} \times w^e\ Ax^{E2}$ | | 276 | 246 | 263 | 212 | | 0.20 | 100% |
| $w^e\ Ax^{16172} \times w^e\ Ax^{16172}$ | | 111 | 98 | 95 | 65 | | 2.51 | 100% |
| $w^e\ Ax^{71d} \times w^e\ Ax^{71d}$ | | 188 | 219 | 203 | 174 | | 3.12 | 100% |
| $w^e\ Ax^{E2} \times w^e\ Ax^{71d}$ | | 333 | 305 | 263 | 256 | | 5.00* | 100% |
| $w^e\ Ax^{16172} \times w^e\ Ax^{71d}$ | | 106 | 121 | 113 | 82 | | 3.49 | 100% |
| $w^e\ Ax^{E2} \times w^e\ Ax^{16172}$ | | 301 | 262 | 320 | 216 | | 0.09 | 100% |
| $w^e\ Ax^{23} \times w^e\ Ax^{23}$ | | 142 | 191 | 187 | 181 | | 0.18 | 100% |
| $w^e\ Ax^{23} \times w^e\ Ax^{9B2}$ | | 242 | 267 | 252 | 255 | | 0.47 | 100% |
| | Bar | (not-Bar) | | (not-Bar) | | Bar | | |
| $w^e\ Ax^{9B2}/Basc \times w^e\ Ax^{9B2}$ | 169 | 128 | 131 | 120 | 121 | 167 | 0.59† | 100% |

* significant at the 5% level.

† the comparison was made between all not-Bar males and not-Bar, not-eosin females.

TABLE 4

*Results of crosses indicating the viability of viable/lethal Abruptex heterozygotes and the effect of duplication,* Dp(1;2)51b, *on the viability of the heterozygotes*

| | Progeny | | | | | | Viability of the heterozygotes | |
|---|---|---|---|---|---|---|---|---|
| | females | | | males | | | | |
| | eosin *Ax* | not eosin | Total | eosin *Ax* | not eosin | Total | without dupl. | with the dupl. |
| $w^e\ Ax^{16172} \times w^a\ Ax^{59d}$; *Dp(1;2)51b* | 0 | 36 | 36 | 532 | 424 | 956 | 0% | 6.78% |
| $w^e\ Ax^{16172} \times w^a\ Ax^{59b}$; *Dp(1;2)51b* | 0 | 43 | 43 | 811 | 584 | 1395 | 0% | 3.33% |
| $w^e\ Ax^{E2} \times w^a\ Ax^{59d}$; *Dp(1;2)51b* | 0 | 507 | 507 | 799 | 624 | 1423 | 0% | 64.00% |
| $w^e\ Ax^{E2} \times w^a\ Ax^{59b}$; *Dp(1;2)51b* | 0 | 486 | 486 | 795 | 547 | 1342 | 0% | 61.00% |
| $w^e\ Ax^{71d} \times w^a\ Ax^{59d}$; *Dp(1;2)51b* | 0 | 327 | 327 | 552 | 445 | 997 | 0% | 59.00% |
| $w^e\ Ax^{71d} \times w^a\ Ax^{59b}$; *Dp(1;2)51b* | 0 | 278 | 278 | 459 | 391 | 850 | 0% | 61.00% |
| $w^e\ Ax^{28} \times w^a\ Ax^{59d}$; *Dp(1;2)51b* | 3 | 795 | 798 | 723 | 778 | 1501 | 0.4% | 110.00% |
| $w^e\ Ax^{28} \times w^a\ Ax^{59b}$; *Dp(1;2)51b* | 11 | 570 | 581 | 453 | 443 | 896 | 2.4% | 125.00% |
| | | | | *y, m* | *Ax* | | | |
| $w^a\ Ax^{59b}$/*dl-49, y Hw m* $\times$ $w^e\ Ax^{9B2}$; *Dp(1;2)51b* | 157 | 650 | 807 | 413 | 153 | 565 | 38.0% | 100.00%* |
| $w^a\ Ax^{59d}$/*dl-49, y Hw m* $\times$ $w^e\ Ax^{9B2}$; *Dp(1;2)51b* | 104 | 465 | 569 | 267 | 176 | 443 | 39.0% | 100.00%* |

* approximated values.

TABLE 5

*Results of crosses made for the estimation of the viability of* $Ax^{59}/Ax^{59}$ *and* $Ax^{59}/Ax^{59}$; Dp(1;2)51b $Ax^{+}$ *females*

| Experimental crosses | Females | | | | Males | | | | | Viability of |
|---|---|---|---|---|---|---|---|---|---|---|
| | $w^a$ *Ax* | $w^+$ *Ax* | $w^a$ *B* | $w^+$ *B* | $w^a$ *Ax* | $w^+$ *Ax* | *Basc* | Bar | Total | *Ax/Ax; Dp females* |
| $w^a$ $Ax^{59d}$/*Basc* × $w^a$ $Ax^{59b}$; *Dp(1;2)51b*/+ | 0 | 39 | 328 | 245 | 0 | 181 | 280 | 115 | 1188 | 13.6%* |
| $w^a$ $Ax^{59b}$/*Basc* × $w^a$ $Ax^{59d}$; *Dp(1;2)51b*/+ | 0 | 18 | 160 | 145 | 0 | 101 | 107 | 69 | 600 | 11.8% |
| $w^a$ $Ax^{59b}$/*Basc* × $w^a$ $Ax^{59b}$; *Dp(1;2)51b*/+ | 0 | 49 | 206 | 168 | 0 | 161 | 77 | 70 | 731 | 26.8% |
| $w^a$ $Ax^{59d}$/*Basc* × $w^a$ $Ax^{59d}$; *Dp(1;2)51b*/+ | 0 | 81 | 262 | 218 | 0 | 220 | 166 | 97 | 1044 | 33.8% |
| Control crosses | $w^e$ *Ax* | $w^+$ *Ax* | $w^e$ *B* | $w^+$ *B* | $w^e$ *Ax* | $w^+$ *Ax* | *Basc* | Bar | Total | |
| $w^e$ $Ax^{28}$/*Basc* × $w^a$ $Ax^{59b}$; *Dp(1;2)51b*/+ | 3 | 321 | 295 | 299 | 296 | 290 | 285 | 244 | 2033 | 108.1% |
| $w^e$ $Ax^{28}$/*Basc* × $w^a$ $Ax^{59d}$; *Dp(1;2)51b*/+ | 0 | 321 | 334 | 299 | 302 | 318 | 293 | 218 | 2085 | 101.4% |
| $w^e$ $Ax^{71d}$/*Basc* × $w^a$ $Ax^{59b}$; *Dp(1;2)51b*/+ | 0 | 121 | 256 | 217 | 274 | 246 | 256 | 152 | 1522 | 51.2% |
| $w^e$ $Ax^{71d}$/*Basc* × $w^a$ $Ax^{59d}$; *Dp(1;2)51b*/+ | 0 | 41 | 248 | 210 | 199 | 205 | 215 | 162 | 1280 | 17.9% |

* When estimating the viabilities the number of $w^+$ *Ax* females was compared with the number of heterozygous Bar females. In the control crosses the frequencies of heterozygous Bar females and $w^e$ *Ax* males did not differ significantly from the expected 2:1 ratio chi-squares being 0.45, 0.002, 2.74, and 3.77 for the four crosses respectively. On the other hand, there is no heterogeneity in the numbers of heterozygous Bar females between all the crosses ($X^2=7.71$; d.f.$=7$). Thus, the viability estimates are comparisons to $w^e$ $Ax^{28}$ and $w^e$ $Ax^{71d}$ males also.

*28/Basc* and *71d/Basc* females were also made (Table 5). The results show that the method of the estimation of the viability of *59/59; Dp* females was a reliable one. The values of viabilities given in Table 5 also measure the viability of these females in relation to *28/Y* and *71d/Y* males (see footnote of Table 5). The females with the genotype *59/59; Dp* have a very strong Abruptex phenotype the wings being arched, the venation of the wings very weak, and the hair pattern of wings and thorax sparse.

*Summary of the results*: The results are summarized in Figures 1, 2, and 3. Figure 1 indicates the type of interaction of the Abruptex alleles. The viabilities of different homozygous and heterozygous allele combinations are presented with appropriate symbols and so is the effect of the Abruptex mutations on Notch. In Figure 2 the effect of the duplication, *Dp(1;2)51b* $Ax^+$, on the viability of different genotypes is presented. Figure 3 presents a simple complementation map of the Abruptex locus. The alleles fall into three complementation units. The recessive lethal alleles, *59b* and *59d*, constitute one unit, the second consists of *9B2* and *28*, i.e. the suppressors of Notch, and the third unit consists of *E2*, *71d*, and *16172*, i.e. the nonsuppressors of Notch. Alleles belonging to the same

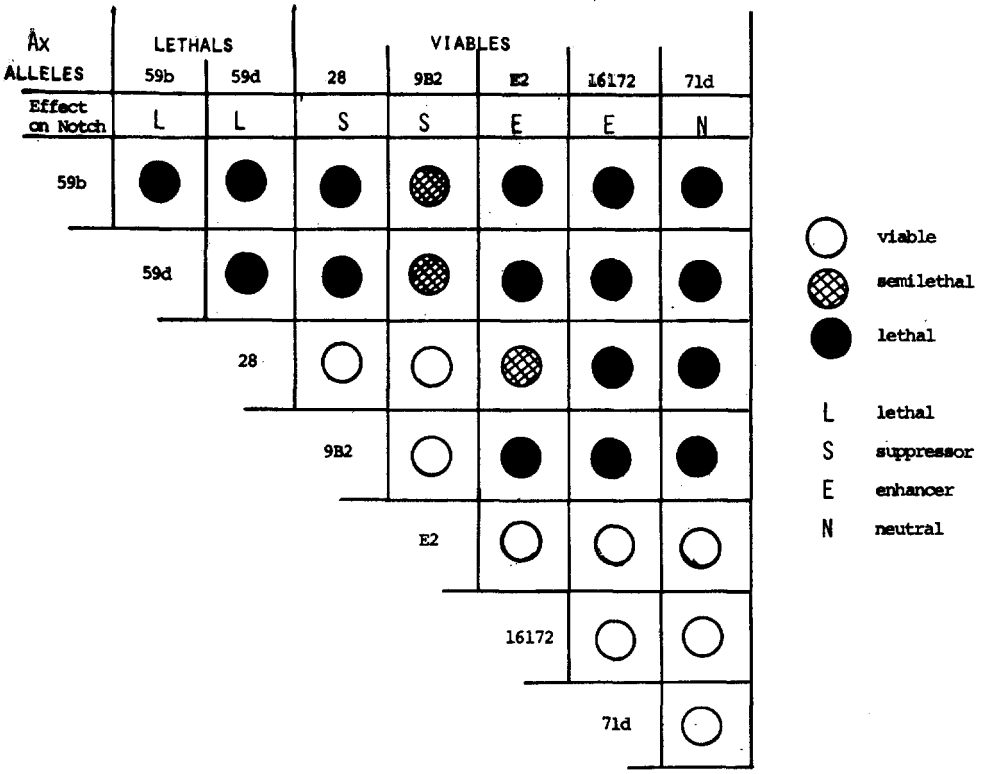


FIGURE 1.—Complementation grid of Abruptex alleles. In each square viability of the respective genotype is presented. A decreased viability of the heterozygote as compared to either homozygote indicates negative complementation. At the top of the grid the type of interaction of Abruptex alleles and Notch mutations is presented.

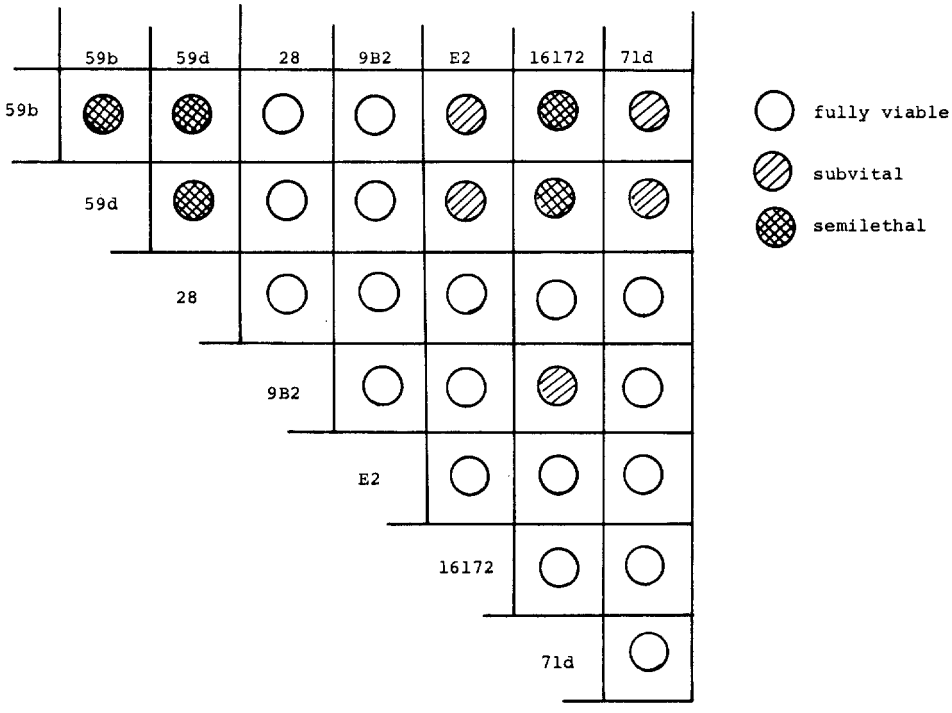


FIGURE 2.—The effect of the wild allele of the Abruptex locus carried by the duplication, *Dp(1;2)51b*, on the viabilities of different combinations of the Abruptex alleles. Compare with Figure 1.

complementation unit neither complement each other nor show negative complementation. Overlapping of dotted lines in the map indicates subvitality or semilethality as the result of the interaction. Heterozygous combinations of alleles falling into nonoverlapping complementation bars are lethal.

## DISCUSSION

Abruptex mutations are members of the Notch pseudoallelic series. The viable Abruptex alleles fall into two groups one containing suppressors of Notch

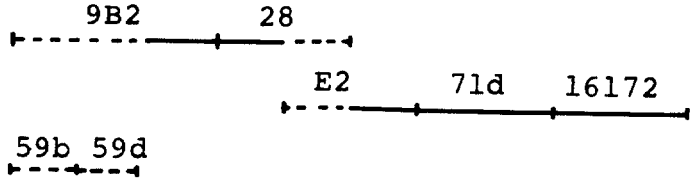


FIGURE 3.—Complementation map of the Abruptex locus. Alleles falling into the same complementation unit neither complement each other nor show negative complementation. Hetrozygous combinations of nonoverlapping alleles are lethal. Overlapping of dotted lines indicates reduced viability.

(alleles *28* and *9B2*) and the other containing alleles which have an neutral effect on Notch (*71d*) or enhance the wing-nicking phenotype of Notch (*E2* and *16172*). Subsequently the suppressor group will be designated $Ax^{SoN}$ (SoN for suppressor of Notch) and the other group for the sake of simplicity will be designated $Ax^{EoN}$ (*EoN* for enhancer of Notch). Homo- and heterozygous $Ax^{SoN}/Ax^{SoN}$ and $Ax^{EoN}/Ax^{EoN}$ females are viable and have an Abruptex phenotype, whereas $Ax^{SoN}/Ax^{EoN}$ heterozygotes always have a decreased viability in relation to either homozygote, and are usually lethal. Thus, alleles falling into the same group neither complement nor show negative complementation, but alleles falling into separate groups exhibit strong negative complementation.

In addition to viable Abruptex alleles there are recessive lethal alleles (*59b* and *59d*). They are lethal in heterozygous combination with Notch mutations, and they do not complement each other. The lethal allele group is subsequently designated as $Ax^{L}$ (*L* for lethal). $Ax^{L}/Ax^{SoN}$ and $Ax^{L}/Ax^{EoN}$ heterozygotes are lethal except that *59b/9B2* and *59d/9B2* are semilethal.

The suppressor-of-Notch alleles are very likely hypermorphic mutations on the basis of following criteria: in $Ax^{SoN}/N$ heterozygotes both Notch and Abruptex phenotypes are suppressed, and since Notch mutations are typically amorphic mutations (WRIGHT 1970), $Ax^{SoN}$ are hypermorphic. When MULLER (1932) defined the concept of hypermorphism he used $Ax^{28}$ as an example of a hypermorphic mutation.

The enhancer-of-Notch (nonsuppressors) on the contrary seem to be hypomorphic mutations since the Notch phenotype is enhanced in $Ax^{EoN}/N$ heterozygotes, and the Abruptex phenotype usually is clearly expressed. The phenotypes of $Ax^{SoN}/Ax^{SoN}/Ax^{+}$ and $Ax^{EoN}/Ax^{EoN}/Ax^{+}$ females also support the conclusion of opposite morphism of $Ax^{SoN}$ and $Ax^{EoN}$ alleles.

The lethal Abruptex alleles seem to share properties of both groups of the viables. Firstly, $Ax^{L}/Ax^{9B2}$ heterozygotes are not completely lethal as are the other heterozygotes in which the lethal alleles are involved. Secondly, $Ax^{L}/Ax^{SoN}/Ax^{+}$ genotypes are fully viable while $Ax^{L}/Ax^{EoN}/Ax^{+}$ genotypes are subvital or nearly lethal, and their viability seems to be very sensitive to external factors (compare the viabilities of *59d/71d; Dp* in Tables 4 and 5). These results suggest that the lethals belong to the same group with the $Ax^{SoN}$ alleles. On the other hand the lethality of $Ax^{L}/N$ females suggest that the $Ax^{L}$ alleles are strong enhancers of Notch, and anyway they seem not to be hypermorphic mutations. It seems likely, therefore, that $Ax^{L}$ mutations are antimorphic alleles. This suggestion is favored by the result that effect of the extra wild allele on the viability of the heterozygotes is weaker if two lethals are involved than if a lethal and a viable are involved. The lethal alleles are antagonists of the wild allele, whereas the two groups of the viable alleles seem to be antagonists of each other but not antagonists of the wild allele.

The opposite morphism of the two groups of viable alleles offers at least a formal explanation for their negative interaction. In the $Ax^{SoN}/Ax^{EoN}$ heterozygotes the alleles with opposite morphism nullify the effect of each other, and the function under control of the locus will be destroyed. A more concrete

explanation of the negative complementation at the Abruptex locus might be as follows: The locus is responsible for a single function which, however, consists of two interdependent components. The $Ax^{SoN}$ alleles might be defects of the one component and the $Ax^{EoN}$ alleles of the other component. They both have the same phenotypic effect as homozygotes because the same function as a whole is disturbed in both of them. In heterozygotes, however, the cumulative action of antagonistic mutations leads to negative complementation, and decreased viability. The recessive lethal alleles ($Ax^{L}$) might be defects of the function as a whole. Therefore, they are usually lethal with both $Ax^{SoN}$ and $Ax^{EoN}$ alleles, and, therefore they are also lethal with the amorphic Notch mutations.

The situation described above might arise for example in the following cases:

1) The Abruptex gene is functional at two times during the development of the fly. $Ax^{SoN}$ mutations are defects of the first functioning time and $Ax^{EoN}$ mutations are defects of the second functioning time. In heterozygotes the disorder accumulates and leads to negative complementation. This idea is supported by the fact that different mutations of the Notch gene are functional at different periods during the development. For example, $N^{g11}/N^{g11}$; *Dp(1;2)51b* genotypic flies have a temperature-sensitive period for lethality at the embryonic stage whereas $Ax^{16172}/N^{40}$ flies have a temperature-sensitive period for lethality at the second-instar larval stage (FOSTER 1973b).

2) The Abruptex locus might be a tandem-repeat coding for a single polypeptide which consists of two more-or-less identical subunits. $Ax^{SoN}$ mutations might be mutations of the first subunit and $Ax^{EoN}$ mutations of the second subunit. In the heterozygote the hybridization of two different mutant polypeptides in the formation of the functional enzyme might decrease the activity of the enzyme below a critical level. $Ax^{L}$ mutations are such in which both subunits are somehow altered. These mutations map as points (WELSHONS 1971), but despite this, the effect of the mutation might spread in the polypeptide so that both subunits become defective. On the basis of the comparison of complementation and recombination maps of the Notch locus FOSTER (1973a) has also presented the idea that the Notch might possibly be a tandem repeat.

3) Perhaps the most tempting alternative for the explanation of the allelic interactions at the Abruptex locus is to suppose that the locus is responsible for regulative function. Considering structural genes (or producer genes to use the terminology of BRITTEN and DAVIDSON (1969)) it would seem likely that a *trans* heterozygote of hypo- and hypermorphic alleles would be more or less wild-type. But considering genes with regulative functions it is conceivable that in this kind of heterozygote a serious imbalance in the developmental homeostasis would arise, and the end-result could be lethality.

The three alternatives presented above are not mutually exclusive. On the contrary, they may complement each other. It should be noted that BRITTEN and DAVIDSON (1969) presented the Notch gene as a possible example of an integrator gene. Indeed, it seems that Notch locus has several characteristics of an integrator gene which are corollaries of the BRITTEN and DAVIDSON-model. Firstly, the locus is pleiotropic, having a variety of mutant forms from em-

bryonic lethals to recessive eye and wing mutations. Secondly, the locus is functional at several times during the development (FOSTER 1973b). Thirdly, the locus is possibly a repetitive locus as suggested by the comparison of the recombination and complementation maps (FOSTER 1973a) and the sequence of the recessive visible mutations which is as follows: *fa–fa$^{no}$–spl–nd* (WELSHONS 1965), i.e. there is a repetition of an "eye mutant-wing mutant" sequence. Fourthly, the negative complementation of the Abruptex mutations suggests a regulative role for the locus.

Financial support for this work has been received from the National Research Council of Sciences of Finland. The skillful technical assistance of MARIITTA RUOHONEN-SIREN (B. Sc.) and the critical reading of the manuscript by DR. ALOHA ALAVA are gratefully acknowledged. The correspondence with DR. G. G. FOSTER has been most fruitful.

## LITERATURE CITED

BRITTEN, R. J. and E. H. DAVIDSON, 1969 Gene regulation for higher cells: A Theory. Science **165**: 349–357.

FINCHAM, J. R. S., 1966 *Genetic Complementation*. W. A. Benjamin Inc., New York, Amsterdam.

FOSTER, G. G., 1972 Genetic studies of the Notch locus of *Drosophila melanogaster*. Genetics **71** suppl. s. 18. ——, 1973a Is the Notch locus of *Drosophila melanogaster* a tandem repeat? Correlation of the genetic map and complementation pattern of selected mutations. Genetics **73**: 435–438. ——, 1973b Temperature-sensitive mutations in *Drosophila melanogaster*. XIII Temperature sensitive periods of the lethal and morphological phenotypes of selected combinations of Notch-locus mutations. Devel. Biol. **32**: 282–296.

LINDSLEY, D. L. and E. H. GRELL, 1968 *Genetic Variations of Drosophila melanogaster*. Carnegie Institution of Washington Publication **627**.

MULLER, H. J., 1932 Further studies on the nature and causes of gene mutations. Proc. 6th Int. Congr. Genet. **1**: 213–255.

PORTIN, P. and M. RUOHONEN, 1972 A new type of allelic interaction at the *Ax* locus of *Drosophila melanogaster*. Drosophila Inform. Serv. **49**: 70.

RIEGER, R., A. MICHAELIS and M. M. GREEN, 1968 *A Glossary of Genetics and Cytogenetics*. Springer Verlag, Berlin, Heidelberg, New York.

SUNDARAM, T. K. and J. R. S. FINCHAM, 1967 Hybridization between wild-type and mutant *Neurospora* glutamate dehydrogenase in vivo and in vitro. J. Mol. Biol. **29**: 433–439.

WELSHONS, W. J., 1965 Analysis of a gene in *Drosophila*. Science **150**: 1122–1129, ——, 1971 Genetic basis for two types of recessive lethality at the Notch locus of Drosophila. Genetics **68**: 259–268.

WRIGHT, T. R. F., 1970 The genetics of embryogenesis in *Drosophila*. Advan. Genetics **15**: 261–395.

ZIMMERMANN, F. K. and E. GUNDELACH, 1969 Intragenic complementation, hybrid enzyme formation and dominance in diploid cells of *Saccharomyces cerevisiae*. Molec. Gen. Genetics **103**: 348–362.

Corresponding editor: B. H. JUDD