The analysis of the pupae was carried out using the methods outlined by Wright (1963) for starch gel electrophoresis. The trays were run for four hours at 5-8 v/cm and stained using L-leucyl-beta-naphthylamide as the substrate and Black K salt as the dye (Beckman and Johnson, 1964). Crosses with the rucuca stock indicated that the Lap D gene is located between ro and ca. The relative frequency with which the Lap D alleles assorted with the outside markers ro and ca of the steroca chromosome, in crossovers in the ro-ca region, was used to determine the exact position of the Lap D gene. Twelve pupae were tested individually from the mating of each testcross male carrying a recombinant chromosome. If the male's genotype was Lap D^F/Lap D^S, two bands were observed in the D zone of approximately half the pupae so analysed. If the male's genotype was Lap D^F/Lap D^F, only one band was observed in the D zone of all the pupae. In this way, sixty-five recombinant chromosomes were tested. From the data obtained, the locus of the Lap D gene was found to be 98.3 ± 0.5 on Chromosome III.

DL-alanyl-beta-naphthylamide-HCl (AAP) was substituted for the Lap substrate (L-leucyl-beta-naphthylamide-HCl, or LAP). When the top half of a gel was stained with the AAP substrate and the bottom half with the LAP substrate, the banding apttern was the same for both. If the gels were placed one on top of the other, the bands were superimposable. Although the enzymes produced better bands with the LAP substrate, they also showed an observable amount of activity with the AAP substrate.

Although squashes of single adults gave little, if any, activity in starch gel, 10 lambda of a concentrated homogenate of a large number of adult flies when inserted into starch gel gave a heavy banding pattern of aminopeptidases, some of which are apparently different from those observed in the pupae.

References

Beckman, L. and F. M. Johnson, 1964, Hereditas 51: 221.; Wright, T. R. F., 1963, Genetics 48: 787.

Rowan, Sister M. Joan. University of Rochester. Is $Plum^K$ homologous to the mutants of the brown locus?

A mutant, Pm^K, which phenotypically resembles previously known Plum mutants, has been reported by Krivshenko (DIS 38:75) This mutant is associated with an inversion in 2R (proximal break at the beginning of 41 and distal break in 57 E-F). Slatis (Genetics 40:5, 1955) has described about 30 variegated position effects of the brown locus; all are due to chromosomal rearrangements involving 59D to 59F and the chromocentral heterochromatin of Y, 2, 3, or 4. However, the breakage point in the case of Pm^K is definitely in the region 57C - 57F. It was thus necessary to determine whether Pm^K is indeed homologous to the mutants of the brown locus or whether it is a separate mutant associated with the 57 region itself.

Initially it was found that the lethals associated with Pm^K and Pm (laboratory stock) are not homologous since Pm^K and Pm were viable in the homozygous condition as if they were associated with separate loci and were both present in the heterozygous condition.

If Pm^K is homologous to the bw locus, it should be possible to separate it from its associated inversion. (However, the fact that the presence of the inversion may or may not be necessary for the expressivity of the Plum character must also be considered). Slatis was not able to carry out such analyses because of the close proximity of the breakage points of his variegated bw mutants to the bw locus itself.

For genetical analysis, the Pasadena stock: px slt sp (#278, DIS, 1964) was used. The location of these markers on 2R is as follows; the bw locus is also indicated as well as the distal end of the inversion associated with Pm^K.

distal break	px	bw	slt	sp
99-100	100.5	104.5	106.3	107.0

Females Pm^K/px slt sp were test-crossed to px slt sp males, and among 3,548 progeny were found two plexus flies and one Pm^K slt sp fly (this latter female was crossed to

Canton stock males and a cytological analysis of the salivary chromosomes of her progeny revealed the presence of the Pm^K inversion). The px flies were readily accounted for by assuming that Pm^K is indeed associated with the distal end of the inversion (57E-F) and that a crossover occurred in the region between px and slt. It is still possible to speculate that Pm^K is allelic to bw but depends upon the inversion for expressivity. In this case the crossover just cited could occur, but the px flies could in actuality be px Pm^K with the Pm^K not being expressed in the absence of the inversion. However, this is highly improbable in light of the fact that considerable cytological analyses of larvae have never revealed the inversion's presence without also observing Pm^K flies in the progeny. Also, the Pm^K slt sp fly could have been the result of a crossover in the region to the left of the bw locus, in which case Pm^K could still be allelic to bw. However, further genetic and cytological analysis of crossover classes using a px bw sp stock from Le Mars, Iowa (#d15, DIS, 1964) has adequately justified the conclusion that Pm^K is not homologous to the mutants at the bw locus but it represents an analogous mutant, phenotypically similar to Pm mutants and located somewhere in section 57 of 2R.

Subsequent genetic analysis involving Pm^K and a Punch mutant, Pu^2 (Mutants of Drosophila Melanogaster; Bridges & Brehme; p 152) kindly supplied to us by Dr. D. Lindsley, has shown that Pm^K and Pu^2 are either allelic or else they have a lethal factor in common.

Meyer, Helen U. University of Wisconsin, Madison. Use of a dominant male sterile factor in second chromosome mutation studies.

In another attempt to avoid the need for virgin females in the F_2 generation of second chromosome breeding procedures, a dominant male sterilizing factor in chromosome 2 is now utilized. This is a modification of a scheme previously described by Abrahamson and Meyer (DIS 40:95, 1965) in which a Minute was used to delay the eclosion of the non-wanted class of F_2. Now, males of this class are made sterile by being heterozygous for a dominant male sterile factor. Both schemes are simple to use, and neither requires special marker or sterilizing genes in the chromosomes to be tested for mutations.

The search for such a dominant male sterile factor in chromosome 2 was initiated by a suggestion of Dr. H. J. Muller, who also advised that such factor could then be kept in stock by balancing it with a dominant female sterile factor. Two mutations of this kind were obtained from X-rayed stage 7 oocytes having wild type (Canton-S) second chromosomes. The male sterile (Ms) is located close to, and just right of, the cn locus. The location of the female sterile (Fs) is not known. Neither has any effect on the external phenotype of the flies carrying it, except that the males heterozygous for Fs are small, much reduced in number and late hatching.

The stock ("Ms") used in this scheme has the composition S Sp Ms bw^D/dp^{txI} Cy,Ins0 pr cn^2 sp and Fs/dp^{txI} Cy,Ins0 pr cn^2 sp. Only the brown-eyed, Ms/Cy females and the non-brown, Fs/Cy males are fertile. Since Fs males hatch late, one should transfer this stock not too early to fresh culture bottles to keep it going. On the other hand, this shortcoming has the advantage that often only virgin females are present during the first few days of hatching.

In its simplest form, the breeding procedure is as follows: Individual P_1 males are crossed to brown, curly virgin females from the "Ms" stock; P_1 parents should be removed. In F_1 one crosses the non-brown, curly males (a desired number from each P_1 culture) individually back to brown, curly females from the "Ms" stock; removal of parents is unnecessary. In F_2 one selects the non-brown, curly flies for brother-sister matings; parents may again be left in. The F_3 is scored in the usual manner for presence or absence of non-curly homozygotes.

Should it be necessary to treat P_1 females and not males, and in those cases where it is important to utilize all, not only half, of the F_1 males produced by a treated P_1, one can combine this stock with Muller's method of "criss-crossed lethals" (Muller, H. J. 1953, DIS 27:104-105). For this purpose the factors S, Sp, dp^{tx} and Cy are present. The procedure is the same as outlined in the above-mentioned note in DIS 40, with the only difference that the "Ms" stock is used instead of the "M" stock described there.