

THE MASKING OF SEPIA BY WHITE, TWO RECESSIVE
EYE-COLORS IN DROSOPHILA

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During the course of a discussion on the dilution effects produced by various eye-colors upon each other in *Drosophila*, there arose the question of the color of an individual homozygous for both White and Sepia. At the time, no definite answer could be given; although it was suggested that they would appear most likely as sepia-eyed flies, judging from the fact that homozygous Sepia-Peach individuals were known to approach so nearly the sepia color that it was extremely difficult to separate them into distinct classes.¹ Search in numerous publications failed to yield satisfactory information upon the subject. Consequently, as the best method for determining the solution to the problem, it was decided to produce a strain, homozygous for the characters involved, and then to observe the result of the combination.

A more exhaustive study of the available literature on parallel problems yielded the following data, much of which seemed to indicate that the flies in question would be of the Sepia phenotype: Homozygous Sepia-Scarlet individuals appear indistinguishable from the stock Sepias.¹ Pure Pink-Maroon flies appear maroon-colored.¹ These facts normally would lead one to believe that the dark eye-colors, located on the third chromosome, dominate those lighter ones found in the same group. On the other hand, the knowledge that homozygous White, Pink individuals appear no different from the pure Whites, indicates the possibility that these White, Sepias might have white eyes instead of sepia-colored eyes.¹

The factor which produces the white eye-color is located on the heterosome or sex-chromosome 1.5 units from the left end, while that which produces the sepia eyes is found on the third chromosome at locus 25.3. Since both of these characters are recessive, they must be present in the homozygous condition before they produce any effect. In the case of the White male, however, only one factor for White needs to be present to produce the character because he possesses only one sex-chromosome instead of two sex-chromosomes, as in the White females.

The first step in this project was the mating of a virgin Sepia to a White male. Figure 1 shows the Punnett square for this cross. In it and the subsequent squares, the following legend was used: Let *w* represent a gene for White eye-color, with *W* representing its dominant Wild allelomorph; then *s* represents a gene for Sepia eye-color with *S* representing its dominant Wild allelomorph; and *Y* represents the allosome. In the above cross, all of the offspring (the F_1 generation) were of the Wild or red-eyed type, being found in a ratio of 1 ♂ : 1 ♀.

¹ Bridges, C. B. and T. H. Morgan, '23. The Third-chromosome Group of Mutant Characters in *Drosophila*. Carnegie Inst. Wash., publ., 327.

The next step was to mate some Wild males from the F₁ generation with their virgin Wild sisters. Figure 2 shows the square depicting the expected results for the F₂ generation. From this it can be seen that the expected ratios are as follows: 3 Wild ♂♂ : 6 Wild ♀♀ : 3 White ♂♂ : 1 Sepia ♂ : 2 Sepia ♀♀ : 1 White, Sepia ♂.

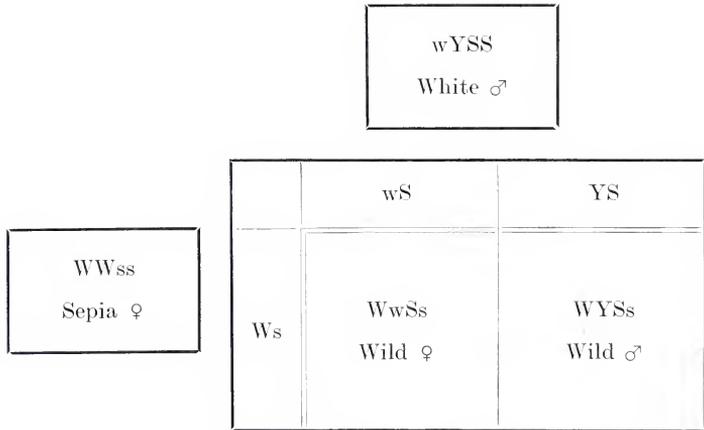


Fig. 1. The production of the F₁ generation by mating a normal White male with a pure Sepia virgin. All of the offspring are of the Wild or red-eyed type.

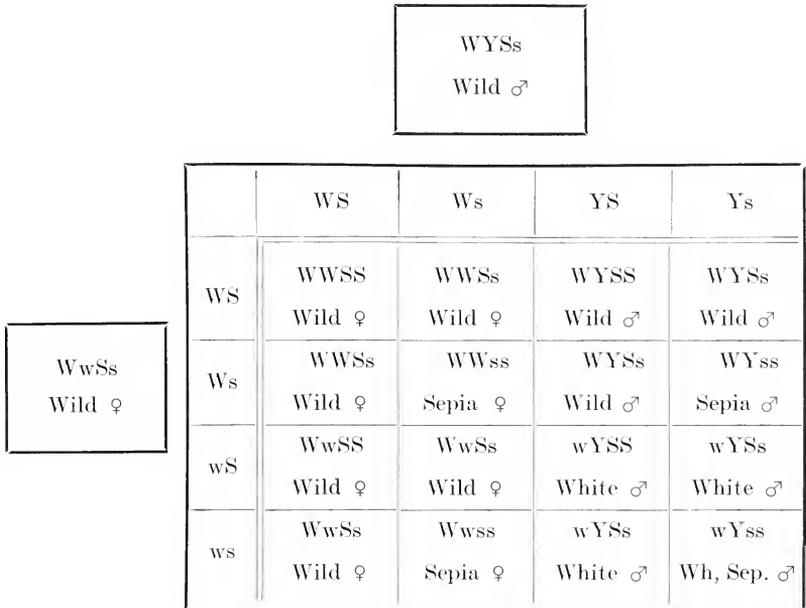


Fig. 2. A square showing the F₂ generation obtained by mating Wild F₁ males with Wild F₁ females. In this generation, the ratio of the offspring is 3 Wild ♂♂ : 6 Wild ♀♀ : 3 White ♂♂ : 1 Sepia ♂ : 2 Sepia ♀♀ : 1 White, Sepia ♂.

On the supposition that the White, Sepia flies fall in the Sepia phenotype, ten of the Sepia males, which appeared in the bottles, were mated to White virgins as soon as emergence had begun.

From figure 2, it is evident that only one combination, producing a true Sepia male, with the genotype WYss, is present in this genera-

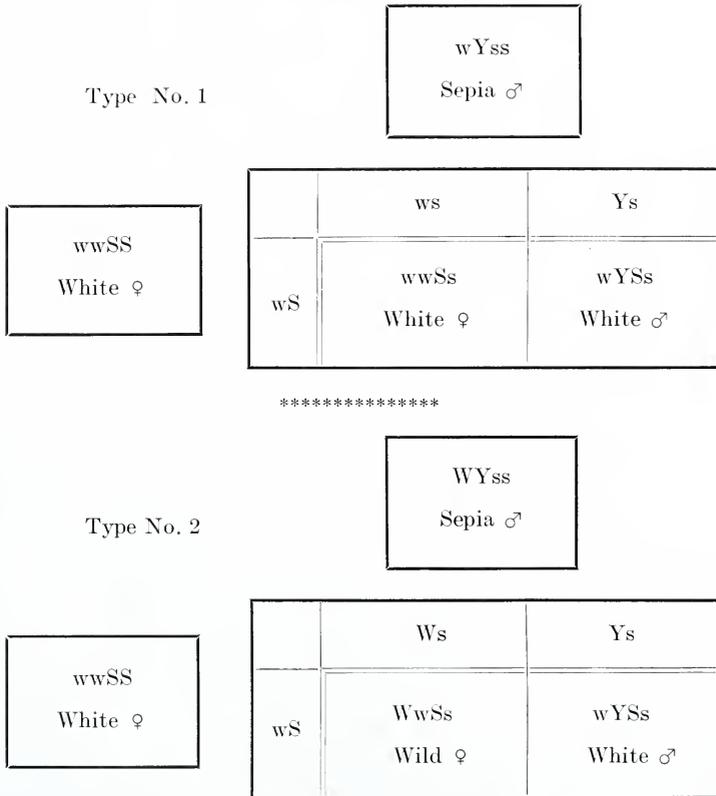


Fig. 3. Squares showing the results obtained when Sepia males of types No. 1 and No. 2 are mated with virgin Whites. In the case of the former type, all of the offspring are white-eyed, while the second type produces White males and Wild females.

tion. The White, Sepia male, having the genotype wYss, might also fall in the Sepia phenotype. Figure 3 shows the results that would be obtained when these two different types of males are mated to White virgins, in order to determine whether or not these sepia-eyed flies are carrying, in addition, the gene for White. It might be well to note, at this point, that all of the males tested throughout this experiment were removed from the mating bottles and saved separately until after their adult offspring had begun to appear. This was done in order to facilitate the production of a pure strain of White, Sepias, should an individual of that description appear—as determined by the kind of adult offspring

produced. Figure 3 clearly indicates how easily these two different types of flies may be differentiated from each other. Type No. 1, which is the kind of individual for which we are looking, would, when mated with the virgin White, give offspring all of which have white eyes. Type No. 2, the pure Sepia, will produce, under the same condition as above, offspring all the males of which have white eyes, and all the females of which are Wild, or red-eyed.

Only five of the ten Sepia males, mated as indicated above, produced offspring—the other five bottles being badly contaminated. In all five of these producing bottles, the offspring appeared as expected if the Sepia males were of the type No. 2 of figure 3. Since the ratio of the two types to each other was 1 : 1, it seemed that the first assumption about the eye-color of the combined characters (namely, that the White, Sepia male would fall in the Sepia phenotype) was at fault, unless the viability of the combination was so greatly reduced that they would not reproduce in the presence of a slight mold contamination.

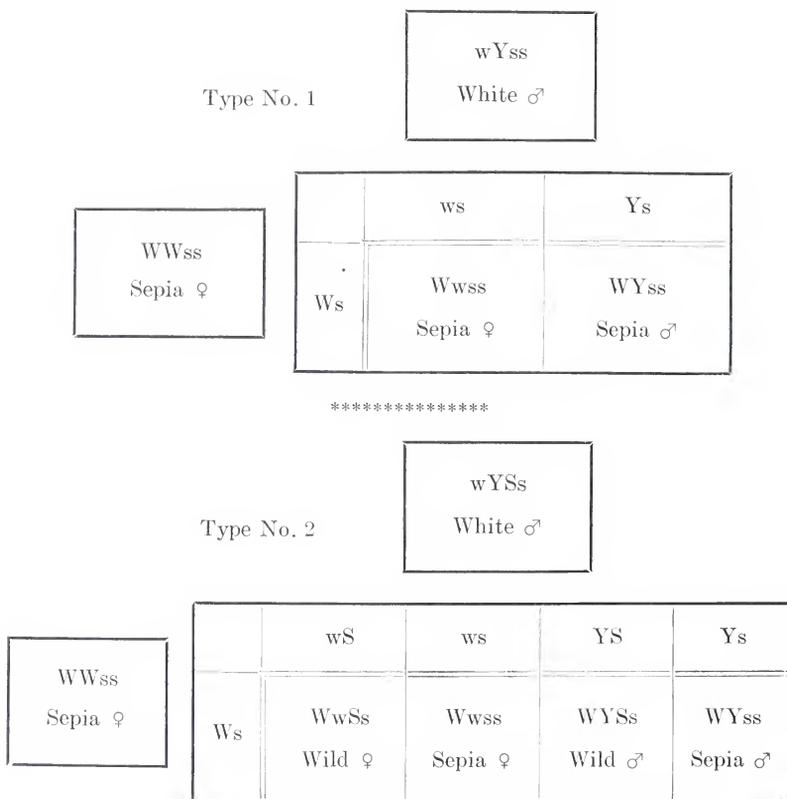


Fig. 4. Squares showing the results obtained when White males of types No. 1 and No. 2 are mated with virgin Sepias. In the case of the former type, all of the offspring are sepia-eyed, while the second type produces Wild males and females and also Sepia males and females.

Before it was time to discard the bottles which, theoretically at least, were producing White, *Sepia* individuals, it was noticed that the number of *Sepia* males produced was considerably below the expected number, while the number of White males was considerably higher than normal expectancy. Consequently, nineteen White males were removed from these bottles and mated to virgin *Sepias*.

In the case of the White males produced, there are two different possibilities with regard to genotypes. If it be assumed that the White, *Sepia*, individuals are White, there is one other possible combination. These different kinds of White males (see figures 1 and 4) will have the following genotypes: Type No. 1, the pure White, *Sepia*, wYss; type No. 2, the heterozygous White, *Sepia*, wYss; and type No. 3, a pure White with the same genotype as the original White male, wYSS. Figure 1 indicates the results obtained when the latter type is mated to a *Sepia* virgin; all of the offspring are Wild. The expected results, obtained when types No 1 and No. 2 are mated in the same way, are shown in figure 4. If the individual is a homozygous White, *Sepia*, all of his offspring will have *sepia* eyes. If, on the other hand, the White male is merely heterozygous for *Sepia*, Wild males and females and also *Sepia* males and females are produced in equal numbers. Again, it can be seen how easily the different genetic types of White males are determined, merely by mating them with virgin *Sepias* and observing their offspring. Here again, as was noted above, all of the males mated were saved until after their offspring had been observed long enough to determine the genetic constitution of the male parents.

Of the nineteen bottles in which the White males were mated with *Sepia* virgins, only four failed to produce offspring. Assuming that the White, *Sepia* flies fall in the White phenotype, it can be seen from figure 2 that the different genotypes of White males have the following ratio: 1 wYss : : 2 wYss : 1 wYSS. Therefore, a greater number of these were mated than in the case of the *sepia*-eyed males, because only one-fourth of the White males would be expected to be of the White, *Sepia* type. Of these fifteen males which produced offspring, thirteen proved to have the genotype wYss (type No. 2, figure 4), and one the genotype wYSS (male parent in figure 1); and only one proved to be a homozygous White, *Sepia* having the genotype wYss (type No. 1, figure 4). Since this last individual was the one for which search had been made, he was mated immediately to one of his virgin daughters, a *sepia*-eyed individual, for the purpose of producing a pure strain of flies of his type. Figure 5 depicts this latter cross, showing the offspring expected, White males and females and *Sepia* males and females in a ratio of approximately 1 : 1. The actual results agreed very closely with the expected ratio, as can be seen from the fact that, in this cross, there were 196 white-eyed individuals and 204 *sepia*-eyed individuals produced. Since all of these white-eyed flies were homozygous White, *Sepias*, some of the White males were mated with their virgin white-eyed sisters, thus establishing a strain which was homozygous for both White and *Sepia* eye-colors.

The next problem was to prove definitely whether the White, *Sepia* strain was homozygous for *Sepia* as well as for White. If it were, this

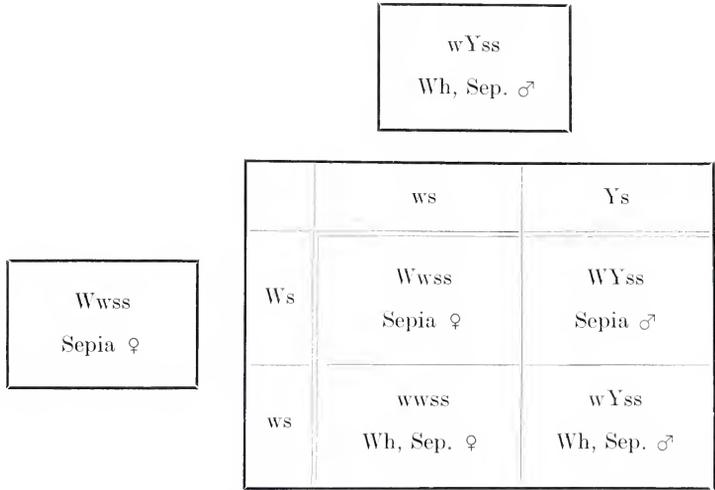


Fig. 5. The cross between the isolated White, Sepia male and one of his virgin daughters in the production of a pure strain of White, Sepias. All of the white-eyed flies produced in this generation are pure for both White and Sepia.

would indicate that the Sepia eye-color was completely masked by the White eye-color. Accordingly, two sets of matings were made. In the first, White, Sepia males were mated with virgin Sepias in pairs. The offspring were observed, and all were found to have sepia eyes. This conforms very nicely with the expected result for this cross and proves that the White, Sepia stock is homozygous for the Sepia factor. In the other set, White, Sepia virgins were mated with pure White males. All of the offspring, in this case, were white-eyed, thus proving that the above combination was also homozygous for White.

The accompanying table shows the actual results obtained in the F₂ generation: and, in addition, whether in the various ratios the White Sepia males are counted as a separate class, in the Sepia phenotype, or

TABLE 1. Actual Results and Expected Numbers Obtained in the F₂ Generation

		Wild		White	Sepia		Wh, Sep.
		♂	♀	♂	♂	♀	♂
Actual results.....		117	187	124	22	60	?
Exp.	1. 3:6:3:1:2:1.....	96	192	96	32	64	32
	2. 3:6:3:2:2:0.....	96	192	96	64	64	0
	3. 3:6:4:1:2:0.....	96	192	128	32	64	0

1. $X^2 = 13.266$
P = 0.010069

2. $X^2 = 40.701$
P = 0.000018

3. $X^2 = 8.223$
P = 0.087

in the White phenotype. If they are counted as a separate class, the ratio is 3:6:3:1:2:1; if in the Sepia phenotype it is 3:6:3:2:2:0. If, however, one counts them in the White phenotype, the ratio is 3:6:4:1:2:0. The X^2 method for determining the goodness of fit, when applied to the results expected from the above ratios, gave the following probabilities: 0.010069 for the 3:6:3:1:2:1 ratio, 0.000018 for the 3:6:3:2:2:0 ratio, and 0.087 for the 3:6:4:1:2:0 ratio. Since probabilities are considered acceptable when they fall within the arbitrarily set limits of 0.02 and 0.98, it is seen at once that the 3:6:4:1:2:0 ratio gives the best result, although even it is a little too low for an extremely good fit. The first two results, in fact, are below the lower limit within which a probability is acceptable. This then is a further indication that the White, Sepias had been counted in the class with the White males.

There are three lines of evidence all of which indicate that the White, Sepia individuals are white-eyed instead of sepia-eyed, and that Sepia is completely masked by White. They are: First, the evidence obtained from the matings used in the isolation of the White, Sepia male; second, the evidence from confirmatory matings of flies from the new White, Sepia strain with White and Sepia individuals obtained from the respective stock strains; and third, the evidence from the use of the X^2 method for determining the goodness of fit when applied to the combined data obtained from mating two of the heterozygous Wild offspring, resulting from the original cross of the White male with the Sepia female.

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