ISOLATION AND CHARACTERIZATION OF *X*-LINKED MUTANTS OF *DROSOPHILA MELANOGASTER* WHICH ARE SENSITIVE TO MUTAGENS

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ABSTRACT

Thirteen *X*-linked mutants have been isolated in *Drosophila melanogaster* which render male and homozygous female larvae sensitive to the mutagen methyl methanesulfonate. Their characterization and preliminary assignment to functional groups is described. Four of these mutants are alleles of *mei-41* (BAKER and CARPENTER 1972). Like previously isolated alleles of this locus, these mutants reduce fertility and increase loss and nondisjunction of the *X*-chromosome in homozygous females. The remaining mutants have been tentatively assigned to six functional groups (two mutants to the *mus(1)101* locus, two to *mus(1)102*, two to *mus(1)103*, and one each to *mus(1)104*, *mus(1)-105*, and *mus(1)106*). Several of the complementation groups can be distinguished on the basis of nondisjunction and cross sensitivity to mutagens. Females homozygous for the *mei-41*, *mus(1)101* and *mus(1)102* mutants exhibit elevated levels of nondisjunction. Mutants belonging to complementation groups *mei-41*, *mus(1)101*, and *mus(1)104* are sensitive to nitrogen mustard (HN2) in addition to their MMS sensitivity. Among these mutants there is currently a direct correlation between sensitivity to HN2, sensitivity to 2-acetylaminofluorene and a deficiency in post-replication repair (BOYD and SETLOW 1976). Only the *mei-41* mutants are hypersensitive to UV radiation, although several of the mutants exhibit sensitivity to γ-rays. Semidominance is observed in female larvae of the *mei-41*, *mus(1)104*, and *mus(1)103* mutants after exposure to high concentrations of MMS. The properties of the mutants generally conform to a pattern which has been established for related mutants in yeast. Additional properties of these mutants are summarized in Table 9.

INTENSIVE studies of DNA repair in prokaryotes have revealed the existence of a complex array of mechanisms which are capable of reversing a variety of alterations in the genetic material (see review volumes edited by HANAWALT and SETLOW 1975). Because of the complexity and redundancy associated with these systems, the isolation of repair-deficient mutants has been of paramount importance in defining the various repair pathways and their interrelationships. Parallel genetic studies in fungi have extended this area into the realm of simple eukaryotes (see review by CATCHESIDE 1974). At the alternate end of the eukaryotic spectrum studies of human genetic disorders have provided definitive evidence for the existence of several different repair mechanisms in complex

[1] Studies of radiation sensitivity are taken from the Ph.D. thesis of T. D. NGUYEN.

eukaryotes (see review by CLEAVER and BOOTSMA 1975). However, restrictions on genetic manipulations in mammals are primarily responsible for the disparity between our knowledge of DNA repair in prokaryotes and higher eukaryotes. In an attempt to bridge this gap we have undertaken a study of DNA repair in *Drosophila melanogaster*, because among complex eukaryotes this organism is most accessible to genetic analysis. Drosophila has the further advantage that it has been extensively employed in studies of mutagenesis (see review by SOBELS 1974).

The first stage in a systematic effort to characterize the DNA repair mechanisms of Drosophila necessarily involves a search for repair-deficient mutants. Because a variety of microbial mutants belonging to this class are sensitive to methyl methanesulfonate (MMS), our initial effort has been concentrated in a search for *X*-linked mutants which render larvae sensitive to this mutagen. Our selection screens are a variation of the approach of SMITH (1973) and mimic replica plating in prokaryotes. This same rationale has previously been employed in the isolation of mutants which are sensitive to γ-ray treatment (NGUYEN, GREEN and BOYD 1976). Since several of the recovered mutants exhibit meiotic abnormalities, they may also contribute to studies of the relationship of DNA repair to genetic recombination in higher eukaryotes. In particular, the first mutagen-sensitive strain to be recovered by SMITH (1973), of which we report several new alleles, has proven to be an allele of a meiotic mutant that was originally isolated by BAKER and CARPENTER (1972).

The mutants recovered in our selection procedure have been tested for complementation with mutants of the same class that were recently isolated by SMITH (1976). On the basis of results obtained in both laboratories all mutants are tentatively assigned to 10 complementation groups. Although the relationship of these mutants to one another has not been thoroughly established, their extensive chemical analysis (BOYD and SETLOW 1976) necessitates a description of their preliminary genetic characterization at this time.

MATERIALS AND METHODS

Mutants

Mutants which define new genetic loci are designated *mus(1)a*^{D1,2,3} (*mus*—mutagen sensitive; *(1)*—chromosome designation; *a*—locus; *D*—Davis, site of isolation; ^{1}, superscript—allele). By agreement between the Davis and Atlanta groups, the numbers 101–199 are reserved for mutagen-sensitive loci on the *X*-chromosome, numbers 201–299 for loci on the second chromosome, 301–399 for loci on the third chromosome, and 401–499 for loci on the fourth chromosome. Alleles of the *mei-41* locus are so indicated.

The isolation and characterization of the following new *X*-linked mutants are described:

mei-41^{D2}, *mei-41*^{D3}, *mei-41*^{D4}, *mei-41*^{D5}
mus(1)101^{D1}, *mus(1)101*^{D2}
mus(1)102^{D1}, *mus(1)102*^{D2}
mus(1)103^{D1}, *mus(1)103*^{D2}
mus(1)104^{D1}
mus(1)105^{D1}
mus(1)106^{D1}

Additional mutants have been described elsewhere:

mei-41^{A1} (SMITH 1973; SMITH 1976)
mei-9, *mei-41*^{195}, *mei-218* (BAKER and CARPENTER 1972)
mei-41^{D1}, *mus(1)107*^{D1} (Nguyen, GREEN and BOYD 1976)

Mutant *X*-chromosomes isolated by BAKER and CARPENTER are marked with *y* and those reported here carry the *w* marker. For convenience, designations of phenotypic markers and the chromosome identifications are omitted from most references to the mutants in the text. Superscripts are omitted in cases where a complementation group is identified by a single mutation. The remaining mutants employed in this study are described in LINDSLEY and GRELL (1968).

Materials

Media containing cornmeal, yeast, agar, molasses, corn syrup and propionic acid was prepared in a pressure cooker and distributed to either 6 dram shell vials or ½-pint milk bottles placed on a flat surface. Plastic vials used in tests for carcinogen sensitivity were obtained from Carolina Biological Supply (Burlington, N.C.). Methyl methanesulfonate (MMS), nitrogen mustard (HN2) and 2-acetylaminofluorene (AAF) were obtained from Aldrich (San Leandro, California). Ethyl methanesulfonate (EMS) was purchased from ICN-K & K Labs (Irvine, California).

Selection procedures

Two independent selection screens have been employed to recover mutants sensitive to MMS (Figure 1). In both procedures mutagenized *X*-chromosomes were produced by feeding *w* males 0.005 M EMS as described by LEWIS and BACHER (1968). Cultures were maintained at 23°-25°.

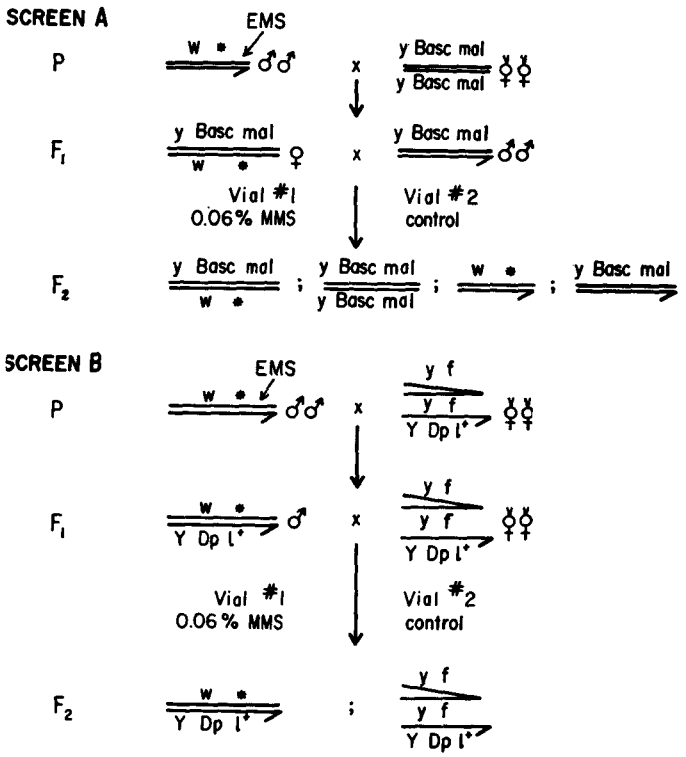

FIGURE 1.—Mating scheme employed in selection.

The first screen (Figure 1, Screen A) was initiated by mass mating mutagen-treated males to homozygous *y Basc mal* virgin females. Single F_1 females were mated to their brothers and allowed to deposit eggs for two consecutive days in each of two separate vials containing 10 ml of medium each. One vial of each pair was treated with 0.25 ml of 0.06% (v/v) MMS immediately after the flies were transferred. The absence of *w* F_2 males in the vial to which MMS had been added and their presence in the rescue vial means that a presumptive MMS-sensitive mutant had been recovered. The presumptive mutants were retested for MMS sensitivity. By crossing virgin heterozygous F_2 females to *w* F_2 males it was possible to monitor the sensitivity of both sexes together with female fertility in the F_3 generation.

The second screen (Figure 1, Screen B) for selecting mutagen-sensitive strains eliminated recessive lethals and included improvements for selecting virgin females. In this scheme mutagenized *w* males were mass mated to virgin compound-*X* females (*C(1)DX, y f/y*^{2}·Y). (These females were produced in large numbers by a self-selection system in which *C(1)DX, y f/Y* females were crossed to males *l/y*^{2}·*Y*. Such males carrying a lethal *l*, are viable only because the *y*^{2}·*Y* carries a tiny duplication bearing *l*+. Thus, the cross yields *C(1)DX, y f/y*^{2}·*Y* females and no males.) Surviving F_1 males were separately mated to two virgin females of the above genotype. After 48 hr all flies were transferred to a fresh vial (the rescue vial!). F_2 larvae were tested for MMS sensitivity as described previously. Eighty percent of the F_1 males produced testable cultures.

Determination of MMS sensitivity

Stocks of each mutant were established in which the male carried the mutant *X*-chromosome and the females were compound *X*. Flies were collected 0-3 days after emergence and fed on a heavy yeast paste for three days. Fifteen pairs were then placed in ½-pint milk bottles containing 50 ml of level medium, dry yeast and no paper. Two days later the flies were removed and on the following day each bottle was placed on a level surface and treated with 1.25 ml of the appropriate MMS solution. One week from the time the flies had been added to the test bottles, tissue paper was placed in the bottles, and the sex ratios of the progeny were determined through day 15 after initiation of the cultures. MMS has not been observed to significantly extend the emergence times of the flies. Cultures were maintained at 23°-25°.

Determination of HN2 and AAF sensitivity

The double *X* stocks employed in these studies were produced by two generations of outcrossing mutant males to virgin compound *X* females. Embryos were collected from population cages of yeast-fed flies over a 12-hr period, suspended in water and distributed to culture media by pipetting. For tests of AAF sensitivity 400–500 embryos were placed on 20 ml of media contained in disposable plastic vials. The number of vials per determination varied from 4 to 10 depending upon the mutagen concentration. In tests for sensitivity to HN2, 800–1,000 embryos were pipetted into 50 ml of media in ½-pint milk bottles. Larger numbers of cultures were also established for tests of the higher HN2 concentrations. 60 hr after egg addition, larvae were treated with either HN2 in water or AAF in 70% ethanol. One-half ml of AAF solution was added to each vial or 1.25 ml of HN2 solution to each bottle. Papers were added to HN2-treated cultures on day 7. Cultures were maintained at 25° and 70% relative humidity and were scored regularly until 18 days after the egg collection. The length of the experiment was extended, because HN2 extends the emergence time of the flies.

Determination of γ-ray sensitivity

The same outcrossed stocks described in the previous section were analyzed for radiation sensitivity. Fifteen pairs were placed in bottles containing 50 ml of medium and no paper. Flies were removed on day 3 and bottles were irradiated with γ-rays on day 5. Irradiation was performed with a ^{60}Co source at dose rates ranging from 3.96 rads/sec. to 4.34 rads/sec. Tissue paper was added to the bottles on day 8. The sex ratio of eclosing adults was determined on days 12 and 14.

Determination of UV sensitivity

Stocks and collection procedures were those described for testing sensitivity to AAF and HN2. Embryos were collected for 6 hr, washed and dechorionated with 50% Clorox (2.25% NaOCl). Dechorionated eggs were maintained on 1.5% agar containing 1% sucrose for 24 hr at 25°. Young larvae were isolated with the procedure of BOYD and PRESLEY (1974) and quickly distributed in a monolayer on damp filter paper. Irradiation was performed in the dark with a G.E. germicidal lamp (fluence rate: 1.3 J/m²/sec). Several hundred irradiated larvae were transferred to each bottle containing 50 ml of medium (day 1). Paper was added to the bottles one week later and the sex ratio was determined on days 11 and 13. As in all sensitivity studies precautions were taken to assure complete eclosion.

Complementation of MMS sensitivity

Males and virgin females were placed together for 2-3 days in the presence of a thick yeast paste. Fifteen pairs were then placed in single bottles containing 50 ml of medium to which dried yeast but no tissue was added. After two days the flies were removed. The bottles were held at 23° for one day prior to the addition of 1.25 ml of water containing one of the following concentrations of MMS: 0.15, 0.10, 0.08, 0.06, 0.04, 0.02, 0.016 or 0.0%. The progeny were scored through day 18 after MMS addition. Most bottles contained well over 100 flies, but any containing less than 60 have been disregarded.

RESULTS

Mutant selection

The new mutants described in this report were recovered in two separate selection experiments (Figure 1). We have previously employed a similar approach to recover two mutants sensitive to γ-rays (NGUYEN, GREEN and BOYD 1976). The first screen, described in MATERIALS AND METHODS, produced 5 sensitive mutants (*mei-41*^{D2}, *mus101*^{D1}, *mus101*^{D2}, *mus102*^{D1}, and *mus103*^{D1}) from 5,200 tested *X*-chromosomes. The recessive lethal frequency in this experiment was 17%, and 92% of the F_1 crosses were fertile. The remaining 8 mutants were isolated in a second screen from 1,650 treated chromosomes. The high recovery obtained with the second procedure is in part due to the elimination of *X*-chromosomes bearing recessive lethals.

Relative sensitivity to chemical mutagens

The relative sensitivity of the mutants to chemical mutagens was determined by varying the concentration of mutagen applied to larval stocks consisting of compound *X* females (*C(1)DX*, *y f*) and males carrying a mutant *X*-chromosome. Females provide an internal standard with which to compare the relative killing of the males. A measure of the male/female ratio as a function of mutagen concentration, therefore, provides an estimate of the relative sensitivity of the mutants.

The relative sensitivity of mutant males to MMS is indicated in Figure 2. The four mutants assigned to the *mei-41* complementation group are consistently the most sensitive (Figure 2e and f). For comparison the sensitivity of the *mei-41*^{A1} allele isolated by SMITH (1973) is also presented. The mutant *mus104* exhibits nearly as strong a sensitivity as the *mei-41* alleles. Mutants assigned to the *mus101* and *mus102* loci are somewhat less sensitive to MMS (Figure 2a and b).

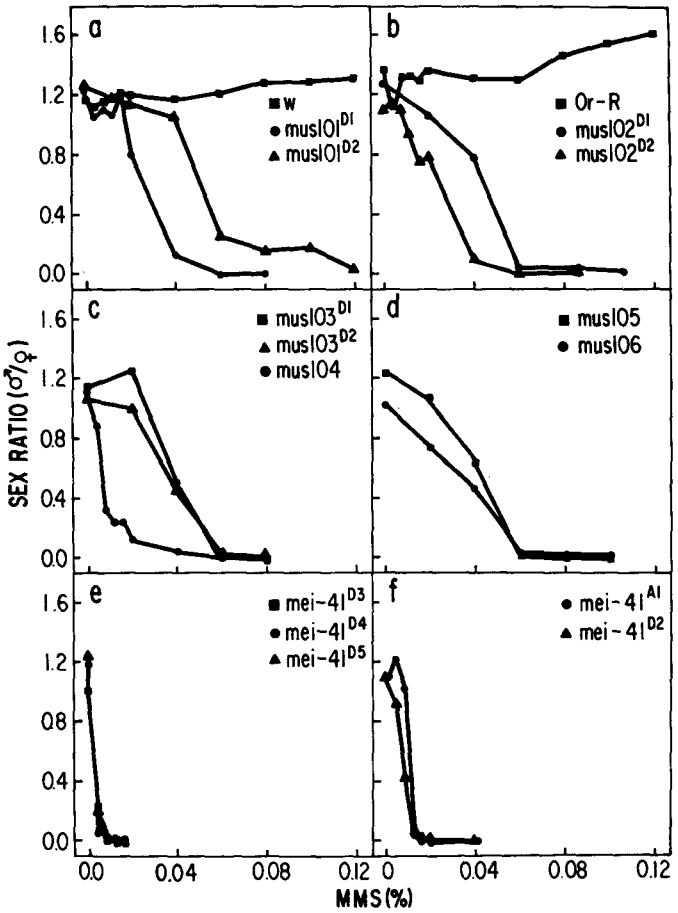

FIGURE 2.—Survival of mutant males relative to control females as a function of MMS concentration. The sex ratio of mutant males to compound *X* females was determined after exposure of larvae to MMS as described in MATERIALS AND METHODS and the text. Abscissa values refer to the concentration of the MMS solution added to the cultures. Each determination is derived from an analysis of 800–1,500 viable embryos. Within the range of 0.0 to 0.1% (v/v) MMS the average number of flies per determination in the control population (*w*) decreased steadily from 1,500 to 1,000. Thus, the controls with which the mutants are compared experience about 30% mortality within the concentration range employed.

The extended plateau observed with *mus101*^{D2} above 0.06% MMS is not attributable to the recovery of a mosaic F_1 male in the original selection, because subsequent stocks generated from single males also exhibit the highly resistant phase of that curve (unpublished observations). Curves obtained with the *mus101* and *mus103* mutants display a shoulder at the lower MMS concentrations (Figure 2a and c), but no significant shoulder is produced by the *mus102* mutants (Figure 2b). It should be noted that these data reflect only the sensitivity that these mutants impart to males. The relative sensitivity of homozygous females carrying the same mutations differs in a few cases (see discussion of complementation analysis).

Analogous experiments have been performed to determine the relative sensitivity of these mutants to nitrogen mustard (HN2) (Figure 3). This mutagen was selected for study because of its capacity to act as a bifunctional crosslinking agent (BROOKES and LAWLEY 1961). Mutants previously selected for sensitivity to γ-rays (NGUYEN, GREEN and BOYD 1976) are also included in this analysis. The controls, which were performed with both a wild type and *w* stock, reveal little difference between the sensitivity of males and females to HN2 (Figure 3a and b). The mutant studies demonstrate that sensitivity to HN2 represents a useful criterion for distinguishing two classes of MMS-sensitive mutants. Those assigned to the complementation groups *mei-41*, *mus101*, *mus104*, and *mus107*

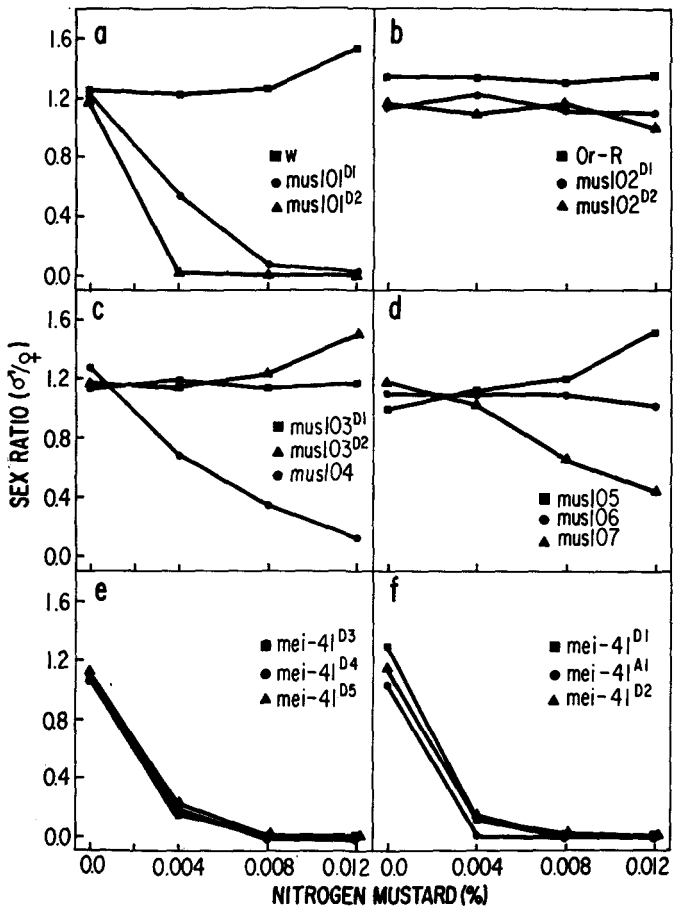

FIGURE 3.—Survival of mutant males relative to control females as a function of nitrogen mustard concentration. The sex ratio of mutant male flies to compound *X* female flies was determined after exposure of larvae to nitrogen mustard as described in MATERIALS AND METHODS and the text. Abscissa values refer to the concentration of the nitrogen mustard solution added to the culture. The number of double *X* females per determination varied from 500 to 1,500. The survival of double *X* females averaged for all stocks was 75% at 0.008% (w/v) HN2 and 50% at 0.012% HN2. Thus, the controls with which the mutants are compared experience about 50% mortality within the concentration range employed.

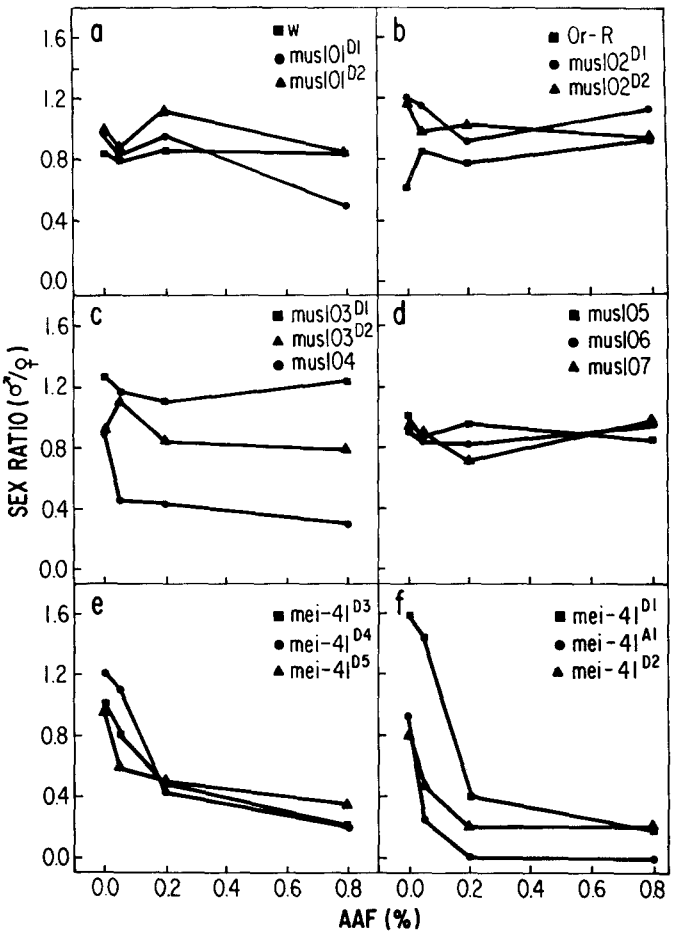

FIGURE 4.—Survival of mutant males relative to control females as a function of AAF concentration. The sex ratio of mutant male flies to compound *X* female flies was determined after exposure of larvae to AAF as described in MATERIALS AND METHODS and in the text. Abscissa values refer to the concentration of the AAF (w/v) in 70% ethanol added to the culture. The number of double *X* females per determination varied from 420 to 1,060. The survival of double *X* females averaged for all stocks dropped to 78% after treatment with an AAF concentration of 0.4% and to 50% at 0.8% AAF.

are sensitive to HN2. The remaining MMS-sensitive mutants are insensitive to this mutagen.

The data in Figure 4 indicate the relative sensitivity of the mutants to the pro-carcinogen 2-acetylaminofluorene (AAF). This compound is of interest because some eukaryotic systems respond to UV and AAF treatment in similar ways (SETLOW and REGAN 1972). Technical difficulties involved in testing this water-insoluble compound make these data less precise than those obtained with the soluble mutagens. The data suggest a parallel between the patterns of sensitivity obtained with HN2 and AAF. Thus, mutants which are strongly sensitive

to HN2 are moderately sensitive to AAF, and mutants with a low sensitivity to HN2 exhibit a slight or negligible sensitivity to AAF. Mutants assigned to both the *mei-41* and *mus104* groups are clearly sensitive to both these agents, whereas it is not clear if the *mus101* mutants are sensitive. The sensitivity of mutagen-sensitive strains to AAF supports previous suggestions that Drosophila possesses the metabolic capacity to activate this pro-carcinogen (FAHMY and FAHMY 1972).

Relative sensitivity to radiation

The selection procedure potentially recovers mutations that alter metabolism and transport of the compound being tested. In an attempt to distinguished mutations which alter these phenomena from mutations in DNA repair mechanisms, we have studied the sensitivity of the mutants to γ-rays and UV radiation. Data presented in Figure 5 indicate the sensitivity of representative mutants from each complementation group to γ-radiation. Data for one of the mutants (*mus107*) previously selected for sensitivity to γ-rays is shown for comparison (Figure 5a). This mutant as well as *mus101*^{D1} and *mei-41*^{D5} exhibit a high sensitivity. Of the remaining mutants tested, three (*mus103*^{D1}, *mus103*^{D2}, and *mus105*^{D1}) are

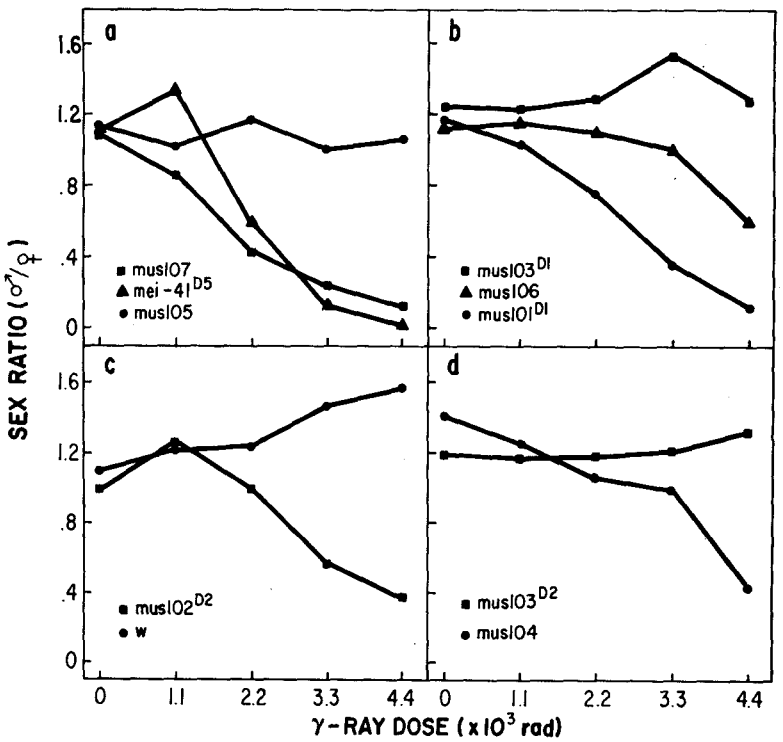

FIGURE 5.—Survival of mutant males relative to control females as a function of γ-ray dose. The sex ratio of mutant male flies to compound *X* female flies was determined after exposure of larvae to γ-rays from a ^{60}Co source. The number of double *X* females per determination varied from 350 to 900. The survival of double *X* females averaged for all stocks was 95% at 2.2 × 10^3 rads and 55% at 4.4 × 10^3 rads.

insensitive and three (*mus102*^{D2}, *mus104*, and *mus106*) exhibit intermediate sensitivity.

Only those mutants belonging to the *mei-41* group exhibit a significant sensitivity to UV radiation (Table 1). The remaining mutants are either very weakly sensitive or insensitive in spite of the fact that the dose employed is well over the half lethal dose for wild-type larvae of this age (DONINI 1972).

Dominance of MMS sensitivity

Interpretation of complementation analyses requires a knowledge of the dominance-recessive relationship of the mutants. This information has been obtained by comparing the MMS sensitivity of heterozygous females relative to control males. Females which are heterozygous for all *mei-41* alleles exhibit wild-type sensitivity to 0.04% MMS and about a 20% reduction in relative survival at 0.06% MMS. At 0.08% MMS this value varies between 50 and 80% relative survival depending upon the allele tested. Thus, at high MMS concentration the most sensitive mutants express a semidominant effect. In order to define clearly those mutants that express semidominance, we have tested heterozygous females at high MMS concentration (Table 2). Under these conditions all the *mei-41* alleles in addition to *mus104* exhibit a strong MMS sensitivity. Mutants of the *mus103* group are moderately sensitive as heterozygotes and the remainder exhibit no significant effect. The MMS sensitivity of heterozygous females

TABLE 1

Sex ratios of adults emerging from UV-irradiated larval cultures in which males carried a mutant X *chromosome and females carried compound* X *chromosomes*

Mutation carried by males	Sex ratios (♀/♂) Unirradiated	Irradiated	Adjusted sex ratio*
w	1.08 (480)	1.12 (400)	1.04
mei-41^{A1}	1.00 (507)	0.27 (258)	0.27
mei-41^{D1}	1.19 (624)	0.36 (284)	0.15
mei-41^{D2}	1.17 (379)	0.40 (750)	0.23
mei-41^{D3}	1.05 (387)	0.28 (484)	0.23
mei-41^{D4}	1.24 (584)	0.55 (528)	0.31
mei-41^{D5}	1.10 (1,227)	0.52 (653)	0.42
mus101^{D1}	1.21 (639)	0.96 (206)	0.75
mus101^{D2}	0.98 (531)	0.82 (396)	0.89
mus102^{D1}	1.38 (854)	0.93 (1,005)	0.55
mus102^{D2}	1.22 (392)	1.05 (535)	0.83
mus103^{D1}	1.19 (470)	1.25 (490)	1.06
mus103^{D2}	1.06 (634)	0.97 (490)	0.91
mus104^{D1}	1.09 (543)	0.91 (433)	0.82
mus105^{D1}	1.17 (603)	0.89 (574)	0.72
mus106^{D1}	1.13 (638)	0.82 (297)	0.69
mus107^{D1}	1.14 (492)	0.97 (273)	0.83

The total number of flies scored in each determination is included in parentheses.
Young larvae were irradiated with 25J/m^2 as described in MATERIALS AND METHODS.
The data for the unirradiated samples are taken from 2–3 cultures of each mutant and the data from irradiated cultures from 4–5 bottles.
* Adjusted ratio is calculated as: Irradiated ratio—(Control ratio—1).

TABLE 2

Test for dominance: Sex ratio of emerging heterozygous females to control males after MMS treatment of larvae

X chromosomal mutant	Sex ratio ♀/♂ Without MMS	With MMS
Oregon-R	1.06 (285)	0.80 (296)
w	0.97 (363)	0.85 (282)
mei-41^{195}	0.90 (201)	0.11 (273)
mei-41^{A1}	1.07 (366)	0.07 (254)
mei-41^{D1}	0.96 (277)	0.07 (230)
mei-41^{D2}	1.18 (359)	0.04 (397)
mei-41^{D3}	1.01 (488)	0.05 (275)
mei-41^{D4}	1.06 (431)	0.03 (438)
mei-41^{D5}	1.01 (411)	0.04 (330)
mus101^{D1}	1.22 (349)	1.00 (345)
mus101^{D2}	0.99 (352)	0.83 (326)
mus102^{D1}	1.01 (402)	0.82 (328)
mus102^{D2}	1.10 (477)	0.84 (318)
mus103^{D1}	1.14 (524)	0.38 (222)
mus103^{D2}	1.10 (309)	0.51 (224)
mus104^{D1}	0.90 (462)	0.10 (284)
mus105^{D1}	0.91 (557)	0.93 (303)
mus106^{D1}	1.02 (418)	0.88 (345)
mus107^{D1}	0.92 (470)	0.78 (264)

Homozygous *Oregon-R* virgin females were mated to males carrying the indicated *X* chromosome. The F_1 progeny larvae were treated with 0.1% MMS as described in MATERIALS AND METHODS. The sex ratio of the eclosing F_1 adults is presented and the total number of males recovered is indicated in parentheses.

reported in Table 2 is considerably higher than that normally detected, but these extreme conditions define more clearly those mutants which potentially express a semidominant effect.

Complementation by MMS sensitivity

Three extensive series of complementation analyses, which are too lengthy to enumerate in detail, were performed as described in MATERIALS AND METHODS (Table 3). All combinations of mutants were generated and tested according to the scheme in Figure 6 at several MMS concentrations. The presence of white-

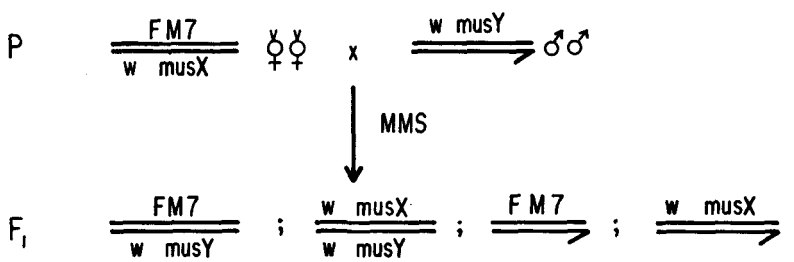

FIGURE 6.—Mating scheme employed in complementation analysis.

eyed females in the F_1 generation indicates that the two mutations leading to sensitivity are complementary and affect separate functions. The number of FM7 males present is an indication of the fertility of the culture, and the absence of sensitive males demonstrates the effectiveness of the MMS. In most cases criteria outlined in Table 3 have permitted a clear decision. With the exception of the mutants *mus103*^{D1}, *mus103*^{D2}, and *mus104* this analysis permits a con-

TABLE 3

Complementation of MMS sensitivity

	mei-41							*mus(1)*—										*mei-9*
	195	A1	D1	D2	D3	D4	D5	101^{D1}	101^{D2}	102^{D1}	102^{D2}	103^{D1}	103^{D2}	104^{D1}	105^{D1}	106^{D1}	107^{D1}	
mei-41^{195}	—	—	—	—	—	—	—	+			+	±	±	±				
mei-41^{A1}		—	—	—	—	—	—					+	±	±				
mei-41^{D1}			—	—	—	—	—	+	+	+	+	±	±	±	+	+	+	+
mei-41^{D2}				—	—	—	—	+	+	+	+	±	±	±	+	+	+	+
mei-41^{D3}					—	—	—	+	+	+	+	±	±	±	+	+	+	+
mei-41^{D4}						—	—	+	+	+	+	±	±	±	+	+	+	+
mei-41^{D5}							—	+	+	+	+	±	±	±	+	+	+	+
mus101^{D1}								—	—	+	+	+	+	+	+	+	+	+
mus101^{D2}									—	+	+	+	+	+	+	+	+	+
mus102^{D1}										—	—*	+	+	+	+	+	+	+
mus102^{D2}											[—]	+	+	+	+	+	+	+
mus103^{D1}												—	±	±	+	+	+	+
mus103^{D2}													[—]	±	+	+	+	+
mus104^{D1}														—	+	+	+	+
mus105^{D1}															[—]	+	+	+
mus106^{D1}																[—]	+	+
mus107^{D1}																	—	+
mei-9																		[—]

Males carrying one mutant *X* chromosome were crossed with females which were heterozygous for the alternate mutant chromosome and an FM7 balancer chromosome as described in MATERIALS AND METHODS. The F_1 larvae were treated with MMS and the eclosing flies were scored. The following criteria were applied in analyzing the data.

— negative complementation

1) *mus/mus* ♀♀ $\lesssim$ *mus* ♂♂ (The minimum MMS dose was used which kills almost all of the *most* sensitive males.)
2) *mus/mus* ♀♀ $\lesssim$ 20% of *mus*/FM7 ♀♀.

\+ positive complementation

1) *mus/mus* ♀♀ $\gtrsim 2 \times$ *mus* ♂♂ (The minimum MMS dose was used which kills almost all of the *least* sensitive males.)
2) *mus/mus* ♀♀ $\gtrsim$ 30% of mus/FM7 ♀♀.

± ambiguous complementation

1) One of the criteria is weak; or
2) The test fails to meet criteria due to intermediate interactions of two mutants; or
3) The test is complicated by extreme differences in the sensitivity of the two mutants to MMS.

[—] Denotes those cases in which the homozygous ♀♀ carrying identical alleles are significantly more resistant to MMS than are ♂♂ carrying that mutant.

* Complementation between *mus102*^{D2} and *mus102*^{D1} is clearly negative, although the test does not rigorously meet the above criteria. This failure is probably due to the insensitivity of homozygous *mus102*^{D2} females relative to *mus102*^{D2} males.

sistent assignment of the MMS-sensitive mutants to 6 complementation groups. Four of the new mutants are seen to be allelic to *mei-41*^{195}. The three mutants which exhibit ambiguous complementation results have been tentatively assigned to functional groups on the basis of related genetic and chemical properties (BOYD and SETLOW 1976). The conflict between SMITH's (1976) results and ours concerning these three mutants may arise from our use of more MMS concentrations. Nevertheless, the assignment of these three mutants remains tentative. Complementation analyses by MMS sensitivity are further complicated by problems of semidominance, density dependent sensitivity of cultures, selective male sensitivity, synergism, divergent MMS sensitivity of the mutants, and intragenic complementation. Additional criteria are therefore required to assign mutants firmly to functional groups. This approach does, however, provide a quick and valuable preliminary analysis.

Complementation of female fertility among the mei-41 *alleles*

Since each of the *mei-41* alleles we have tested is associated with reduced female fertility, we have measured complementation of female fertility as a further test of allelism between these mutants (Table 4). In this test females heterozygous for two *mei-41* alleles were mated with males carrying the FM7 balancer. The viability of embryos produced in each cross was measured as the percent of eggs developing to adult flies. Those alleles which are associated with the greatest fertility are seen partially to complement one another, but total viability never exceeds 25%. These results imply that a single function responsible for low female fertility is associated with each of these alleles. This conclusion strengthens the assignment of these mutants to a single gene and confirms that the *mei-41*$^{+}$ function is associated with female fertility as well as genetic recombination and mutagen resistance (BAKER and CARPENTER 1972).

Generation of all combinations of the *mei-41* alleles with *mus103*^{D1}, *mus103*^{D2}, and *mus104* in heterozygous females has also served to separate these latter

TABLE 4

Complementation analysis of egg viability (in percent) among mei-41 *alleles*

	mei-41^{195}	*mei-41*^{A1}	*mei-41*^{D1}	*mei-41*^{D2}	*mei-41*^{D3}	*mei-41*^{D4}	*mei-41*^{D5}
mei-41^{195}	2	2	1	16	1	5	25
mei-41^{A1}		1	0	3	0	4	11
mei-41^{D1}			0	0.2	0	0	1
mei-41^{D2}				0	0.2	1	3
mei-41^{D3}					0	0	0
mei-41^{D4}						0	0.3
mei-41^{D5}							9

Males carrying one *mei-41* allele were crossed with virgin females which possessed a *mei-41* allele balanced with FM7. Ten to fifteen females bearing the two *mei-41* alleles were crossed to FM7 males. Viability was determined as the percentage of eggs producing adult flies. Reciprocal crosses were made in all cases involving different alleles. A minimum of 200 eggs was counted for each determination involving the same allele and 500 eggs for the data derived from heterozygous females. In control experiments egg viability approached 100%.

mutants from the *mei-41* locus. Females homozygous for each of these three mutants are fully fertile and there is no indication of interaction between them and any of the *mei-41* alleles with respect to female fertility (unpublished observations).

Nondisjunction and loss of X*-chromosomes in female meiosis*

As a result of the frequent correlation between deficiencies in DNA repair and genetic recombination in microbial mutants, we have initiated a study of the meiotic process in the MMS-sensitive mutants. In Drosophila recombination-defective mutants there is an inverse relationship between the frequency of meiotic recombination and nondisjunction of chromosomes in female meiosis (CARPENTER and BAKER 1974). Thus, analysis of the frequency of exceptional gametes produced by females provides a convenient first approach for probing genetic recombination in these mutants. This analysis has been performed for representative mutants of most of the identified complementation groups (Table 5). In this experiment females which were homozygous for the mutagen-sensitive mutations were crossed to $y\ Hw\ w/y^{31d}\cdot Y\cdot B^{s}$ males. All parental females were derived from established stocks in which the males carried a $Y\cdot B^{s}$. This permitted immediate identification of *XX* (not *B*) and *XXY* (*B*) females. Only the former were tested. Use of these tester males permits the separate identification of exceptional progeny derived from either diplo-*X* or nullo-*X* gametes produced by females or males. The frequencies of exceptional gametes of female origin are presented for each sex. Female exceptions are presumed to be derived exclusively from chromosome nondisjunction, whereas the male exceptions represent the combined effects of chromosome loss and nondisjunction. A comparison of the frequencies of exceptional gametes in mutant and control females

TABLE 5

Analysis of spontaneous nondisjunction and chromosome loss in homozygous mutant females

Female parent	F_1 progeny $w/w/B^{s}$ ♀♀	$y\ Hw\ w/O$ ♂♂	Total females	Total males	Frequency of exceptions: Exceptional females × 10^4	Exceptional males × 10^4	Sex ratio (♂/♀)
w	1	2	7,879	7,237	1	3	0.92
$y^2\ sn^3$	7	31	27,999	25,490	3	12	0.91
$w\ mei\text{-}41^{D2}$	7	57	760	487	92	1,170	0.64
$w\ mei\text{-}41^{D5}$	37	174	5,461	3,741	68	465	0.69
$w\ mus101^{D1}$	4	61	3,655	2,455	11	248	0.67
$w\ mus101^{D2}$	2	29	2,046	710	10	408	0.35
$w\ mus102^{D1}$	20	18	7,256	7,010	28	26	0.97
$w\ mus102^{D2}$	9	15	6,211	5,619	14	27	0.90
$w\ mus103^{D1}$	0	6	10,874	9,708	0	6	0.89
$w\ mus103^{D2}$	3	2	4,210	4,010	7	5	0.95
$w\ mus104^{D1}$	3	7	9,334	8,135	3	9	0.87
$w\ mus105^{D1}$	7	3	7,530	4,458	9	7	0.59

Homozygous mutant females were crossed with $y\ Hw\ w/y^{31d}\cdot Y\cdot B^{s}$ males. Nondisjunction and loss of *X* chromosomes is recorded as the percentage of exceptions for a given sex divided by the total for that sex. Additional details are provided in the text.

($w, y^2 sn^3$) permits a division of the mutants into three classes. The strong meiotic mutants belong to either the *mei-41* or *mus101* complementation groups. The *mus102* mutants exhibit a weak but significant effect on chromosomal loss and nondisjunction. The *mus101* and *mus102* loci therefore appear also to function in meiosis. The remaining four mutants tested have no significant effect on this process. Thus, the striking differences associated with these mutants provide a valuable criterion for subdividing the mutagen-sensitive mutants.

Similar tests have also been made for X-ray-induced loss and nondisjunction of *X*-chromosomes in selected mutants (Table 6). Females homozygous for the mutants were irradiated within 8 hr after emergence and mated to $y\ Hw\ w/y^{31d}.Y.B^s$ males. Two mutants which are either normal or weak meiotic mutants (*mus104* and $mus102^{D2}$) exhibit a stimulated increase in male exceptions which is comparable to that of the control (*w*). The stronger meiotic mutants ($mei\text{-}41^{D5}$ and $mus101^{D1}$) likewise exhibit an increase, but the relative increase in these two mutants is much less than that exhibited by the controls. The significance of the difference between the two groups of mutants is in question because of the high spontaneous frequency exhibited by the one class. The influence of X-rays on the production of female exceptions follows a similar pattern, although the strength of the effect is much less striking.

TABLE 6

X-ray-induced nondisjunction and loss of X *chromosomes—percent by sex*

Female parent	X-ray dose in roentgens		
	Zero R	2,000R*	3,000R*
Male exceptions			
w	0.03% (7,237)	1.45% (7,792)	2.57% (7,087)
$mei\text{-}41^{D5}$	4.65% (3,741)	4.96% (1,989)	
$mus101^{D1}$	2.48% (2,455)	4.36% (2,635)	
$mus102^{D2}$	0.27% (5,619)		4.38% (3,335)
$mus104^{D1}$	0.09% (8,135)		2.90% (5,130)
Female exceptions			
w	0.01% (7,879)	0.10% (8,325)	0.11% (8,229)
$mei\text{-}41^{D5}$	0.68% (5,461)	0.08% (2,626)	
$mus101^{D1}$	0.11% (3,655)	0% (4,691)	
$mus102^{D2}$	0.14% (6,211)		0.39% (3,753)
$mus104^{D1}$	0.03% (9,334)		0.04% (5,870)
Sex ratios			
w	0.92	0.93	0.86
$mei\text{-}41^{D5}$	0.69	0.76	
$mus101^{D1}$	0.67	0.56	
$mus102^{D2}$	0.90		0.89
$mus104^{D1}$	0.87		0.87

* Values obtained without irradiation are subtracted from values obtained with irradiation.

Loss and nondisjunction of maternal *X* chromosomes were analyzed as described in Table 5 with the exception that the homozygous female parents were irradiated as virgins at 0–8 hr after eclosion. For each sex the total number of flies scored is indicated in parentheses. Irradiation was performed with a modified General Electric dental machine which delivers 90 KV X-rays at 430R/min.

Complementation of nondisjunction and chromosome loss

Selected pairs of mutants have been tested for their effect on nondisjunction and chromosome loss in heterozygous females (Table 7). This is a more reliable test for assigning mutants to functional groups than is the criterion of MMS sensitivity, because meiotic mutants generally express little or no semidominance with respect to disjunction (CARPENTER and SANDLER 1974). This experiment was identical to that described in Table 5 with the exception that the parental females were heterozygous for two different mutagen-sensitive mutations. The data reveal that three mutants assigned to the *mei-41* locus fail to complement. This failure strengthens their assignment to the same locus. In addition, *mus101^{D1}* and *mus101^{D2}* are noncomplementary, thereby supporting their assignment as alleles. The data reported also provide strong genetic evidence that *mus101^{D1}*, *mus101^{D2}*, *mus103^{D1}*, *mus103^{D2}* and *mus104* are not alleles of the *mei-41* locus. The disjunction data are consistent with the interpretation that in compound with a *mei-41* allele, each of the five mutants is complementary. Added support for the conclusion of complementation comes by comparing the disjunction data

TABLE 7

Complementation of nondisjunction and chromosome loss between mutagen-sensitive mutants

Female parent	F_1 progeny		Total females	Total males	Frequency of exceptions		Sex ratio (♂/♀)
	w/w/B^s ♀♀	*y Hw w/O*♂♂			Exceptional females × 10^4	Exceptional males × 10^4	
w mei-41^{D4}/ w mei-41^{D5}	37	190	3,740	2,608	99	729	0.70
w mei-41^{D3}/ w mei-41^{D5}	24	135	2,950	1,908	81	708	0.65
w mei-41^{D5}/ w mus101^{D1}	3	2	3,167	2,885	9	7	0.91
w mei-41/ w mus101^{D2}*	1	1	3,602	2,930	3	3	0.81
w mei-41^{D2}/ w mus103^{D1}	0	3	11,343	10,022	0	3	0.88
w mei-41^{D5}/ w mus103^{D2}	3	6	8,504	7,126	4	8	0.84
w mei-41^{D2}/ w mus104^{D1}	2	11	11,251	9,948	2	11	0.88
w mus101^{D1}/ w mus101^{D2}	5	72	5,492	3,406	9	211	0.62
y mei-218/ w mus101^{D1}	1	1	3,779	3,389	3	3	0.90
y mei-218/ w mus101^{D2}	1	4	4,830	4,333	2	9	0.90
y mei-218/ w mus106^{D1}	2	3	4,799	4,077	4	7	0.84

Spontaneous loss and nondisjunction of maternal *X* chromosomes were analyzed as described in Table 5 with the exception that the female parent was heterozygous for two different sensitive mutants.

* Data compiled from tests with *mei-41^{D2}*, *mei-41^{D4}*, and *mei-41^{D5}*.

with those obtained from females carrying an MMS-insensitive meiotic mutant (*mei-218*) and MMS-sensitive mutants (*mus101^{D1}*, *mus101^{D2}* and *mus106*). *A priori* complementation is expected and the data given in Table 7 bear out this expectation. Finally, the disjunction tests with the *mei-41* alleles confirm the absence of significant semidominance among these mutants with respect to meiotic disjunction (BAKER and CARPENTER 1972).

Mapping

Preliminary mapping studies have permitted localization of most mutants to restricted regions of the *X*-chromosome (see DISCUSSION and Table 9). In these experiments females heterozygous for a mutagen-sensitive mutant and a multiply marked *X*-chromosome were mated to wild-type males. Progeny larvae were treated with the appropriate amount of MMS to kill most males carrying the mutagen-sensitive marker. Surviving progeny were scored for the presence of recombinant males. These data were compared with data obtained from parallel cultures which had not been exposed to MMS. This procedure has permitted the precise localization of one mutant and the approximate location of several others. In the former case *w mus102^{D2}/y^{2} sn^{3} lz^{50e30}* females produced equal numbers of male progeny recombinant for the *y^{2} w* region in untreated cultures. In treated cultures 11 *y^{2} w* males to 4 *sn^{3} lz^{50e30}* males were produced. Thus, the *mus102^{D2}* mutant is located at about 0.4 map units on the *X*-chromosome; this observation is in agreement with SMITH's (1976) positioning of another allele (*mus102^{A1}*) of this mutant at 0.5. Of those mutants which are less precisely mapped *mus105^{D1}* maps between *ec* and *ct* (5.5–20.0) which agrees with the more exact localization of 14.8 by SMITH (1976) for an allele (*mus105^{A1}*) of this mutant. Three mutants (*mus101^{D1}*, *mus103^{D1}* and *mus104*) map close to *f* and *mus106^{D1}* is less clearly assigned to this region. The four new *mei-41* alleles reported here have each been localized near *f*. The likelihood that any of the recovered *X*-chromosomes carries more than one mutagen-sensitive mutation is reduced by the localization of this phenotype to restricted chromosomal loci.

Relationship to other mutagen-sensitive mutants

During the final stages of this work we have coordinated our efforts with those of other laboratories in order to rationalize the nomenclature for the mutagen-sensitive mutants. The *mei-41* alleles were identified with that locus as a result of SMITH's observation (personal communication) that *mei-41^{A1}* (formerly *mut^{s}*, SMITH 1976) is an allele of *mei-41*. Complementation analyses performed between mutants recently recovered in Davis and Atlanta are presented in Table 8. These data suggest that both laboratories have recovered alleles of two loci (*mus102* and *mus105*). Mapping studies have further confirmed this relationship (Table 9). Although weak complementation of MMS sensitivity was observed between *mus107* and *mus109^{A1}*, allelism between these mutants has been excluded by mapping studies (NGUYEN, GREEN and BOYD 1976; SMITH 1976). In this context the *mus107* mutant has also been observed to interact weakly with *mei-41* alleles in complementation analyses. The Davis collection

TABLE 8

Complementation by MMS sensitivity of Davis mutants with Atlanta mutants

	0.06% MMS		0.1% MMS	
	mus102^{A1}	*mus105*^{A1}	*mus109*^{A1}	*mus110*^{A1}
mus101^{D1}	+ (1.12)	+ (1.23)	+ (1.27)	+ (1.28)
mus102^{D2}	— (0.00)	+ (0.87)	+ (1.24)	+ (0.96)
mus103^{D1}	+ (0.88)	+ (1.17)	+ (0.71)	+ (1.16)
mus104^{D1}	+ (1.19)	+ (0.65)	± (0.31)	+ (0.79)
mus105^{D1}	+ (1.14)	— (0.13)*	+ (1.26)	+ (1.31)
mus107^{D1}	+ (1.32)	+ (1.17)	± (0.13)	+ (1.13)

Males carrying the Atlanta-derived *X* chromosome were crossed with females heterozygous for FM7 and the Davis-derived chromosome. Embryos were collected for two days and the cultures were held for one day prior to the addition of the stated amount of MMS. Combinations of the six most sensitive mutants were treated with a lower MMS dose in order to reduce complications arising from semidominance. Killing of males was nearly complete with the exception of those carrying the *mus105*^{D1} chromosome. + Positive complementation. — Negative complementation. Ratios of females carrying two MMS sensitive chromosomes to females heterozygous for the FM7 balancer are given in parentheses.

* This ratio is artificially high, because the MMS concentration used failed to kill about 17% of the mutant males.

TABLE 9

Summary of mutant properties

Mutant	Map position	Nondisjunction and chromosome loss	Semidominance	Fertility of homozygous cultures	Exhibits sensitivity to: MMS	HN2	AAF	γ-rays	UV
mei-41^{A1}	54.2†		+++	—	+++	+++	+++		++
mei-41^{D1}			+++*	—*	+++*	+++	+++	+++*	++
mei-41^{D2}	near *f*	+++	+++	—	+++	+++	+++		++
mei-41^{D3}	near *f*		+++	—	+++	+++	+++		++
mei-41^{D4}	near *f*		+++	—	+++	+++	+++		++
mei-41^{D5}	near *f*	+++	+++	+	+++	+++	+++	+++	+
mus101^{D1}	near *f*	++	—	++	++	++	+	+++	—
mus101^{D2}		+++	—	—	+	+++	+		—
mus102^{D1}	0.5†	+	—	+++	+	—	—		—
mus102^{D2}	0.4	+	—	+++	++	—	—	++	—
mus103^{D1}	near *f*	—	+	+++	+	—	—	—	—
mus103^{D2}		—	+	+++	+	—	—	—	—
mus104^{D1}	near *f*	—	+++	+++	+++	++	++	+	—
mus105^{D1}	14.8†	—	—	+++	+	—	—	—	—
mus106^{D1}	near *f*		—	—	+	—	—	+	—
mus107^{D1}	27.7–33*		—*	—*	++*	+	—	+++*	—
w				+++	—	—	—	—	—

* From data of NGUYEN, GREEN and BOYD 1976.

† From data of SMITH (1976) for that mutant or an allele of that mutant. See MATERIALS AND METHODS for details.

+++ Exhibits this characteristic strongly.

++ Exhibits this characteristic moderately.

\+ Exhibits this characteristic weakly.

— Does not exhibit this characteristic.

Blank. Not tested.

probably does not include alleles of either *mus109*^{A1} or *mus110*^{A1}, although two of the Davis mutants (*mus106* and *mus103*^{D2}) have not been successfully tested against the Atlanta mutants.

DISCUSSION

Properties of individual complementation groups. A qualitative summary of the properties of the new mutagen-sensitive mutants is presented in Table 9. The *mei-41*^{A1}, *mei-41*^{D1} and *mus107* alleles, which have been described previously (SMITH 1973; NGUYEN, GREEN and BOYD 1976), are included for comparison. The mutants are assigned to 8 functional groups on the basis of available genetic and biochemical information (BOYD and SETLOW 1976). Because of limitations in each of these forms of analyses, they are being extended to solidify the assignments. In general, the groupings of the stronger mutants are more secure than those of the weak mutants. Of particular interest is the apparent localization of several functionally different mutants near *f*. The interrelationship of these mutants will hopefully yield to detailed mapping studies. In comparing the relative sensitivity of the mutants to different mutagenic agents it should be noted that each of these agents was administered to larval populations of different ages. The relative sensitivity of different larval stages is known for UV treatment (DONINI and BOYD 1976) but not for the other mutagens.

Complementation analyses by MMS sensitivity, female sterility, and nondisjunction clearly place all *mei-41* mutants in a single functional group. The properties of the *mei-41* alleles are remarkably uniform with the exception of *mei-41*^{D5} which is somewhat more fertile and less UV-sensitive than the others. That mutant is also seen to exhibit a lower nondisjunction frequency than the *mei-41*^{D2} allele. The *mei-41*^{D3}, *mei-41*^{D4} and *mei-41*^{D5} alleles were all recovered in the same selection experiment and are thus potentially derived from the same mutagenic event in a premeiotic cell. However, since the *mei-41*^{D5} allele exhibits significantly different properties, it probably had an independent origin. The broad sensitivity of these mutants to a variety of mutagenic agents suggests that the *mei-41*$^{+}$ function is vital to the repair of many types of DNA damage. The *mei-41*^{D5} allele is deficient in a form of postreplication repair (CAS) which is postulated to involve a recombination function (BOYD and SETLOW 1976).

Mutants of the *mus101* group which, like the *mei-41* alleles, map near *f*, exhibit similar properties to the *mei-41* mutants except that they do not exhibit UV sensitivity or semidominance to MMS sensitivity. The fertility of homozygous females of *mus101*^{D1} indicates that female sterility is not necessarily associated with this locus. The *mus101*^{D1} mutant is deficient in postreplication repair (BOYD and SETLOW 1976).

The *mus102*^{D2} mutant has been clearly separated from other mutagen-sensitive strains by mapping studies. Alleles of this locus display a relatively weak sensitivity to MMS and γ-rays and are insensitive to both HN2 and AAF. They are also weak meiotic mutants which exhibit no semidominance or sterility. *mus102*^{D2} appears to have a much stronger influence on the MMS sensitivity of

males than it does on females. No chemical defect in DNA repair has yet been associated with these mutants.

Of the remaining mutants, two (*mus104* and *mus107*) are also highly sensitive to MMS. Both of these mutants also interact to some extent with the *mei-41* alleles. Due to the normal chromosome disjunction observed in the repair-defective *mus104* mutant, this locus has been postulated to function in a crossover independent form of postreplication repair (BOYD and SETLOW 1976). No defect in DNA repair has been associated with the four weaker mutants and they otherwise possess properties expected of mutants that alter the metabolism or transport of MMS.

Relation of Drosophila mutants to those in other organisms. The cross sensitivity of the Drosophila mutants to mutagenic agents is similar to the pattern of sensitivity observed with mutagen-sensitive yeast mutants. That pattern permits the assignment of yeast strains to one of three groups: (I) mutants sensitive to HN2 and UV, (II) mutants sensitive to MMS and X-rays, and (III) mutants which are sensitive to all four agents (BRENDEL and HAYNES 1973). The summary presented in Table 9 reveals that Drosophila mutants which are strongly sensitive to γ-rays are also very sensitive to MMS. A reasonable correlation is also found between the mutants that are sensitive to HN2 and those sensitive to AAF. If we equate sensitivity to UV, HN2 and AAF for purposes of this discussion, mutants in the complementation groups *mei-41*, *mus101*, *mus104* and *mus107* satisfy criteria established for Class III yeast mutants. The loci *mus102* and *mus106* result in Class II mutants which are only sensitive to MMS and X-rays. Since we selected for MMS-sensitive mutants, we could not have recovered any mutants corresponding to Class I. The remaining 3 mutants (*mus103*^{D1}, *mus103*^{D2}, and *mus105*^{D1}) may be weak Class II mutants.

Relationship to previous genetic studies. Several lines of evidence suggest the existence of a functional relationship between the processes of meiotic recombination and DNA repair in Drosophila. Young oocytes containing synaptinemal complexes are particularly resistant to irradiation (KING 1970). Since enzymes that mediate recombination are presumably present at high concentration at this time, these same enzymes are postulated to play a role in DNA repair. WATSON (1972) has implicated the meiotic mutant *c(3)G* in the repair of X-ray and alkylation damage. The two loci *mei-9* and *mei-41*, which were initially identified as affecting meiotic processes, have been shown to impart MMS sensitivity to somatic cells (BOYD, GOLINO and SETLOW 1976; SMITH 1976). This relationship has been further strengthened by this report in which several new MMS-sensitive mutants are shown to disrupt meiotic processes (Table 5). Thus, the repair processes which are presumably disrupted in these Drosophila mutants may involve recombination, although this supposition remains to be proven biochemically. An involvement of recombinational mechanisms in bacterial repair has been extensively documented, and potentially related mechanisms are currently being investigated in mammalian systems (see volumes edited by HANAWALT and SETLOW 1975).

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