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A Meiotic Effect of a Mutator Gene in *Drosophila melanogaster* *

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Summary. Two new parameters of the third chromosome mutator gene, *mu*, in *Drosophila melanogaster* are described. The mutator significantly alters both meiotic chromosome disjunction and crossing over in females, but not in males. In meiotic phenotype, the mutator resembles several recently described "meiotic" mutants of *D. melanogaster*. However, previous studies have demonstrated that the mutator also functions premeiotically. The mutator appears to be a unique mutant which functions exclusively in females during both mitotic and meiotic cell divisions.

Introduction

The fortuitous discovery of an experimentally tractable mutator gene (*mu*) in *D. melanogaster* (Green, 1970) provided an opportunity for the examination of the genetic properties of a higher eukaryote mutator (see Green, 1973). In the initial studies of the mutator (Green, 1970; Green and Lefevre, 1972), it was demonstrated that *mu*: 1) significantly increases spontaneous mutability, but only in females; 2) is semi-dominant and incapable of reverting its own induced mutations; 3) functions premeiotically; and 4) is localized to map position 57–58 on chromosome 3. Subsequently, Gold and Green (1973) showed that *mu* also increases spontaneous somatic mutability.

The close proximity of the mutator to the recessive meiotic mutant *c(3)G* (Green, 1970; Lindsley and Grell, 1968), and to the short third chromosome deficiency Df(3R)*sbd*^{105} (Green, 1970), coupled with the known "meiotic mutant" effect of the heterozygous *sbd*^{105} deficiency (Hinton, 1966; Lindsley *et al.*, 1968) prompted the question of possible mutator function during meiosis, despite the fact that both *c(3)G* and the heterozygous *sbd*^{105} deficiency are not mutators (Hall, 1971; Gold, unpublished data).

In the studies presented below, the mutator is shown to significantly alter both meiotic chromosome disjunction and crossing over, exclusively in females. The discovery of mutator meiotic function is discussed in terms of mutator function during both mitotic and meiotic cell divisions.

Methods and Materials

In Table 1, a synopsis of all gene mutants and special chromosomes used in this study is given. Symbols and map locations are taken from Lindsley and Grell (1968). Standard Droso-

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Table 1. Synopsis of mutants and special chromosomes used in text (symbols after Lindsley and Grell, 1968)

Symbols	Linkage	Phenotype
y	1– 0.0	yellow body, light bristles
y^2	1– 0.0	allele of *y*
y^{31d}	1– 0.0	allele of *y*
Hw	1– 0.0	Hairy wing
su(w^a)	1– 0.05	supressor of w^a
z	1– 1.0	♂ wild-type, ♀ zeste eye color
w^-	1– 1.5	♂ viable white deficiency
w^a	1– 1.5	white apricot eye color
spl	1– 3.0	rough small eye, split bristles
cv	1–13.7	crossveins absent or reduced
ct^6	1–20.0	cut wing
sn^3	1–21.0	singed bristles
lz^{50e}	1–27.7	reduced, almond shaped eye
v	1–33.0	vermilion eye color
g	1–44.0	garnet eye color
f^{3N}	1–56.7	forked bristles
odsy	1–59.2	reduced eye, outstretched wings
car	1–62.5	carnation eye color
C(1)RM	Compound reversed metacentric	attached-X homozygous $y^2 su(w^a) w^a$/O
$Y^s \cdot X \cdot Y^L$In(1)d149	attached-XY combination	homozygous *y v f* car
$B^s \cdot Y$	marked Y chromosome	Bar eye
Ly	3–40.5	Lyra wing
p^p	3–48.0	pink-peach eye color
mu^a	3–57–58	mutator gene
DF(3R)sbd^{105}	Df(3R)88F; 89B4-5	homozygous lethal

[a] Not listed in Lindsley and Grell (1968).

phila culture methods were employed throughout, and all experiments were carried out in a laboratory whose temperature was controlled to 22–23°C.

1. X chromosome disjunction in both females and males was followed in the chromosome 3 genotypes +/+, +/*mu*, *mu*/*mu*, +Df(3R)sbd^{105} and *mu*/Df(3R)sbd^{105}. In the male disjunction experiments, the sex chromosomes of the males were $y^2 w^- spl\ sn^3/B^s \cdot Y$, and of the females $y^2 v\ f^{3N}/y^2 v\ f^{3N}$. In some experiments, attached X females without a free Y, C(1)RM, $y^2 su(w^a)\ w^a$/O were used in place of homozygous $y^2 v\ f^{3N}$ females. In the female disjunction experiments, the X chromosomes of the females were several combinations of the genotypes $y^2 z\ spl$, "+" and $y^2 w^-\ spl\ sn^3$ and the males were of the genotype $y\ Hw\ w/B^s \cdot Y$.

Female or male virgins of appropriate X and third chromosome genotypes were collected 2 hrs following eclosion and crossed individually to 2–3 males or females. Parents were transferred to fresh medium every 5 days for a total of 15 days oviposition, after which all parents were discarded. F_1 progeny were scored for 17–18 days following the last day of mating at each transfer. In crosses of males to attached X females, an excess of C(1)RM females per mating was necessary since ca. one-half of the expected progeny are inviable, *i.e.*, O/$B \cdot Y$ die and C(1)RM, $y^2 su(w^a) w^a$/"X" metafemales are poorly viable.

In the female disjunction experiments, by marking the male X and Y chromosomes with the dominant markers *Hw* (Hairy wing) and *B* (Bar), respectively, all regular and exceptional X progeny are unambiguously identified, regardless of the parental origin of the exceptional X chromosome. Similarly, by marking the male Y chromosome with *B* in the male-disjunction experiments, all regular and exceptional X progeny are identified.

2. Meiotic crossing over on the X chromosome was examined in the same third chromosome genotypes as above, with the exception of the homozygous mutator genotype. During

Table 2. X non-disjunction[c] from crosses of ♀♀ of each chromosome 3 genotype to ♂♂ *y Hw w/B*^{s}·Y

Chromosome 3 genotype	Σ XXY ♀♀[a] / Σ ♀♀ scored	Mean freq.[b] diplo-X exceptions $\times 10^4$	Σ XO ♂♂ / Σ ♂♂ scored	Mean freq.[b] nullo-X exceptions $\times 10^4$
+/+	5/16485	3.03	11/16632	6.61
+/*mu*	5/16656	3.02	16/14035	11.40
+/Df(3R)*sbd*^{105}/	24/11971	20.05[d]	27/10656	25.34[d]
mu/*mu*	17/13799	12.32[d]	45/11212	40.14[d]
mu/Df(3R)*sbd*^{105}	72/24802	29.00[d]	155/19135	81.00[d]

[a] XXY ♀♀ = ♀♀ *Hw*$^{+}$ *B* (diplo-X exceptions from X non-disjunction in parental ♀♀). XO ♂♂ = ♂♂ *Hw B*$^{+}$ (nullo-X exceptions from X non-disjunction in parental ♀♀).
[b] Mean freq. diplo-X exceptions = Σ ♀♀ *Hw*$^{+}$ *B*/Σ ♀♀ scored. Mean freq. nullo-X exceptions = Σ ♂♂ *HwwB*$^{+}$/Σ ♂♂ scored.
[c] All metafemales are excluded from the data; and all presumptive XO ♂♂ were progeny tested to confirm expected sterility.
[d] Indicates significant difference ($P < 0.05$) from +/+ controls (after Stevens, 1942).

the crossover experiments, it was discovered that mutator activity was lost on a *p*^{p} *mu l* chromosome which had been used throughout both the non-disjunction and crossing over experiments. Since each chromosome (both X and 3) was balanced with crossover supressing chromosomes (Gold, 1973; Lindsley and Grell, 1968) to maintain as much homogeneity as possible among samples and between experiments, a new mutator chromosome was not synthesized for these experiments. The exclusion of the homozygous mutator genotype from the results did not, however, affect the conclusions drawn from the experiment.

Two types of crossing over experiments were performed. In one, crossing over was measured in the distal X chromosome map interval 0–20 using the marker genes *y*^{2}, *cv* and *ct*^{6}. In the second, the proximal interval 27–59 was examined using the marker genes *lz*^{50e}, *v*, *g* and *odsy*. The relatively short intervals, ca. 20–30 centiMorgans, were employed to obtain low but measurable frequencies of crossing over in both the distal and proximal regions of the X chromosome.

Virgin females of appropriate genotype were collected within 2 hrs after eclosion and immediately singly mated to *sn*^{3} males. Females were allowed to oviposit for two consecutive 7-day intervals. All F_1 males were scored for recombinant and non-recombinant chromosomes, and for nullo-X exceptions (*sn*^{3}/O) resultant from X non-disjunction in parental females. This latter class served as the internal control for the presence of the chromosome 3 genotype tested.

Results

The data from the female disjunction experiments are presented in Table 2 along with the female X non-disjunction frequencies for each chromosome 3 genotype. It should be noted that observed X exception frequencies (both diplo-X and nullo-X) represent recovered X exceptions only, and hence reflect one-half of the actual non-disjunction frequencies in each genotype. That is, nullo-X ova fertilized by Y-bearing sperm produce inviable zygotes, and diplo-X ova fertilized by X-bearing sperm produce metafemales which are excluded from the data.

As shown in Table 2, the frequency of both diplo-X and nullo-X non-disjunctions is significantly increased in mutator homozygotes. The increase in X non-disjunction when compared to the +/+ control, however, is relatively small,

ca. a 4-fold increase in diplo-X non-disjunctions and an 8-fold increase in nullo-X non-disjunctions. A much greater deviation (>10-fold) from wild-type is found in mutator-deficiency (*mu*/Df(3R)*sbd*^{105}) heterozygotes. Although this result could partially reflect a mutator dosage effect, the more probable explanation is a synergism with the effect on disjunction of the heterozygous deficiency (cf. Table 2), an effect previously demonstrated (Hinton, 1966; Lindsley *et al.*, 1968). The increase in nullo-X non-disjunctions in mutator heterozygotes is of borderline significance ($0.10 > P > 0.05$), and in all probability reflects a partial or semi-dominant effect of the mutator as was shown for mutator-induced reversion (Green, 1970).

The increased X non-disjunction found in +/Df(3R)*sbd*^{105} heterozygotes (relative to the +/+ control) merits further attention. Hinton (1966) reported 48 X non-disjunctions in 1,273 progeny recovered from +/Df(3R)*sbd*^{105} mothers (ca. 4% recovered X non-disjunctions), which is roughly a 20-fold increase in X non-disjunctions over that observed in these experiments, *i.e.*, 51 X non-disjunctions in 22627 progeny (ca. 0.2% recovered X non-disjunctions). Genetic tests of the limits of Df(3R)*sbd*^{105} (see Lindsley and Grell, 1968) were performed by crossing ♂♂ Df(3R)*sbd*^{105} separately to: 1) ♀♀ heterozygous for *sbd*^{1} and ♀♀ heterozygous for *Sb* (both *sbl*^{1} and *Sb* are recessive lethal mutations uncovered by Df(3R)*sbd*^{105}); and 2) ♀♀ heterozygous for the extreme meiotic mutant *c(3)G* (Gowen and Gowen, 1922; Hall, 1972), which is also uncovered by Df(3R)*sbd*^{105} (Lewis, 1949). The heterozygous compounds Df(3R)*sbd*^{105}/*sbd*^{1} and Df(3R)*sbd*/*Sb* were both lethal; and heterozygous *c(3)G*/Df(3R)*sbd*^{105} ♀♀ exhibited a high frequency of X non-disjunction (80 X non-disjunctions in 260 F_1 progeny from the cross ♀♀ *c(3)G*/Df(3R)*sbd*^{105} X ♂♂ *y Hw w*/*B*^{s} · *Y*) characteristic of *c(3)G* homozygotes. These results confirm genetically that the Df(3R)*sbd*^{105} deficiency used here uncovers the wild-type alleles for *sbd*^{1}, *Sb* and *c(3)G*, and hence is inseparable genetically from the original *sbd*^{105} deficiency defined by Lewis (1949). Thus, no clear explanation for the differences between Hinton's results and those described here is evident. It is important to note, however, that the deficiency used in these experiments, originally obtained from Pasadena, has been used in this laboratory for almost 4 years. In that time, little variability from ca. 0.2% X non-disjunctions has been observed on over 50000 progeny scored from +/Df (3R)*sbd*^{105} females.

During the scoring of F_1 progeny derived from homozygous mutator and heterozygous mutator/deficiency females, a number of instances of multiple exceptions per female were recovered from singly mated P_1 mothers. Since clustering of both mutator-induced reversions and sex-linked lethals was found by Green (1970) and Green and Lefevre (1972), it was necessary to evaluate the origin, premeiotic or meiotic, of the X chromosomes in each genotype tested. In *D. melanogaster* females, three of four haploid nuclei are lost as polar body nuclei during meiasis. Thus, if X non-disjunction is of meiotic origin, the distribution of X exceptions should be random (each recovered X exception represents a single non-disjunctional event). Conversely, if X non-disjunction is of premeiotic origin where a "cluster" may derive from a single non-disjunctional event, the distribution of X exceptions should be non-random.

The observed distribution of X exceptions in each genotype was compared to the expected random distribution of infrequent events based on the Poisson

Table 3. Observed and expected (from the Poisson series) distribution of X non-disjunctions from single tester females of each chromosome 3 genotype

Chromosome 3 genotype		Summed XXY and XO exceptions/tested female				Σ ♀♀	df	χ^2	P[a]
		0	1	2	3				
+/+	Obs.	185	14	1	—	200	1	0.31	>0.05
	Exp.	184.6	14.8	0.6					
mu/+	Obs.	180	19	1	—	200	1	10^{-3}	>0.05
	Exp.	180.1	18.9	1.0					
+/Df(3R)*sbd*^{105}	Obs.	230	40	4	1	275	2	0.15	>0.05
	Exp.	228.4	42.4	3.9	0.3				
mu/*mu*	Obs.	148	44	6	2	200	2	0.23	>0.05
	Exp.	146.7	45.5	7.1	0.7				
mu/Df(3R)*sbd*^{105}	Obs.	606	165	25	4	800	2	1.31	>0.05
	Exp.	602.4	170.9	24.3	2.4				

[a] Non-significant chi-square ($P>0.05$) indicates random distribution of observed X non-disjunctions.

series of a multinomial distribution. A chi square test for goodness-of-fit was carried out between the observed and expected number of X exceptions, and the results are shown in Table 3. For each chromosome 3 genotype, no difference ($P>0.05$) was found between the observed and expected number of X exceptions per female. The conclusion follows that X non-disjunction in both mutator homozygotes and mutator/deficiency heterozygotes is of meiotic origin, and hence that the mutator gene functions in females during meiosis.

The finding that the mutator is active during meiosis in females led to the question of whether the mutator is also active during meiosis in males. Each of the 5 third chromosome genotypes was tested for its influence on XY non-disjunction in males. As noted in "Methods", males were crossed to both normal and attached-X females. The purpose of the additional cross to attached-X ♀♀ was to determine whether any diplo-X sperm (products of non-disjunction at the equational division) could be recovered.

The results of these crosses are given in Tables 4a and 4b. The frequency of X exceptional sperm appears to be the same irrespective of third chromosome genotype. Statistically there is no difference from one genotype to the next. However, a substantial amount of variability in recovered X exceptional sperm was noted among all samples from each third chromosome genotype. A similar experience is reported from other studies of *D. melanogaster* male non-disjunction (Hall, 1970), and may reflect an inherent property of male sex chromosome disjunction. Clearly, the mutator gene has no effect on male X chromosome disjunction.

As the above results establish that the mutator gene does not affect male chromosome disjunction, the previous interpretation (Green, 1970; Green and Lefevre, 1972) that the mutator gene functions exclusively in females is reinforced. The mutator, thus, appears to be a unique mutant which functions in females during both meiotic and mitotic cell divisions.

Table 4a. Male X non-disjunction and frequencies (including 95% confidence limits)[a] from crosses of ♂♂ of each chromosome 3 genotype to ♀♀ homozygous $y^2v\ f^{3N}$

Chromosome 3 genotype	Recovered ♂♂ gametes[c]				Frequency recovered exceptional sperm/10^3 recovered sperm	
	X (♀♀)	Y (♂♂)	XY (♀♀)	O (♂♂)	XY	O
+/+	7422	6512	12	28	0.86 (0.44–1.52)[b]	2.00 (1.33–2.94)
+/Df(3R)*sbd*^{105}	7667	6372	15	23	1.07 (0.60–1.76)	1.63 1.01–2.45
mu/+	3214	3079	3	6	0.48 (0.10–1.39)	0.95 (0.35–2.07)
mu/*mu*	3921	3612	3	8	0.40 (0.08–1.16)	1.06 (0.46–2.10)

[a] 95% confidence limits are computed from the standard error following the method and corrections of Stevens (1942).
[b] Parentheses below observed frequency indicate computed 95% confidence limits.
[c] Parentheses after recovered ♂♂ gamete indicate the sex of F_1 progeny.

Table 4b. Male non-disjunction and frequencies (including 95% confidence limits)[a] from crosses of ♂♂ of each chromosome 3 genotype to ♀♀ C(1)RM, $y^2su(w^a)w^a$/O

Chromosome 3 genotype	Recovered ♂♂ gametes[c]					Frequency recovered exceptional sperm/10^3 recovered sperm	
	X(♂♂)	Y(♀♀)	XY(♂♂)	O(♀♀)	XX(♀♀)[d]	XY	O
+/+	692	741	0	0	0	—	—
+/Df(3R)*sbd*^{105}	6027	4650	6	24	0	0.56 (0.21–1.22)[b]	2.24 (1.44–3.34)
mu/*mu*	5371	3982	5	14	0	0.53 (0.17–1.25)	1.49 (0.82–2.51)
mu/Df(3R)*sbd*^{105}	4970	2952	3	10	0	0.38 (0.08–1.11)	1.26 (0.61–2.32)

[a] 95% confidence limits are computed from the standard error following the method and corrections of Stevens (1942).
[b] Parentheses below frequency indicate computed 95% confidence limits.
[c] Parentheses after recovered ♂♂ gamete indicate the sex of F_1 progeny.
[d] Diplo-X gametes derived from non-disjunction at the equational division (cf. Text).

Since meiotic crossing over is also restricted to female *D. melanogaster*, experiments were undertaken to further assess the effect of the mutator on crossing over. Earlier it was found (Green, unpublished data) that meiotic crossing over is severely reduced at the X chromosome tip (map interval 0–21) in mutator homozygotes and mutator/deficiency heterozygotes. Studies of *D. melanogaster* "meiotic mutants" (Sandler *et al.*, 1968; Sandler and Lindsley, 1974) have shown

that different mutants exhibit variable "polarity" effects on crossing over in different chromosome regions. Thus, crossing over was examined in both proximal and distal regions of the X chromosome to detect possible polarity in crossing over in mutator females. The empirical results of all experiments are given in Tables 5a and 5b and the crossover frequencies per chromosome 3 genotype per interval are statistically evaluated in Tables 6a and 6b.

As noted in "Methods", the crossing over data for mutator homozygotes are not shown. An internal control for the presence of the mutator, *i.e.*, frequency of recovered nullo-X exceptions (sn^3/O ♂♂—cf. Table 2), indicated that mutator activity was lost. That is, the frequency of sn^3/O ♂♂ was reduced from expected (ca. 1×10^{-3} *vs.* 4×10^{-3}—cf. Table 2). The homozygous mutator females were obtained as *Ly mu*/p^p *mu 1* (where 1 = a recessive lethal to the right of *mu*), and both mutator chromosomes were synthesized independently from the original y^2w^- *spl* sn^3; *mu* stock isolated by Green in 1969. As the samples of *mu*/Df(3R) sbd^{105} heterozygotes included both *Ly mu*/Df(3R)sbd^{105} and p^p *mu 1*/Df(3R)sbd^{105} genotypes, a comparison of sn^3/O (X/O) exceptional male frequencies between the two confirmed that only the p^p *mu 1* chromosome had lost mutator activity. This was further confirmed when the two chromosomes were independently tested for y^2 reversions in females homozygous y^2w^-*spl* sn^3 (see Green, 1970 for expected frequencies of y^2 reversions in mutator homozygotes).

The loss of mutator activity in the p^p *mu 1* chromosome was disappointing but not unexpected given the transient nature of previously isolated *D. melanogaster* mutator genes (Green, 1973). Nonetheless, a comparison of crossing over frequencies between *mu*/Df(3R)sbd^{105} heterozygotes and +/Df(3R)sbd^{105} heterozygotes (cf. Tables 6a and 6b) strongly indicates that the mutator significantly reduces crossing over along the entire X chromosome, and that the reductions in both proximal and distal regions are approximately equivalent. A polarity effect is not indicated. The absence of a polarity effect in crossing over in mutator/deficiency heterozygotes conflicts with the results of Green (unpublished data). He found the reduction in crossing over in homozygous mutator females more extreme at the chromosome tip (region y^2-*w*-*spl*, map units 0–3) than in more proximal regions (*spl*-sn^3, map units 3–21). Considering the relatively short regions examined by Green, however, it is possible that the strong mutator-induced polarity is unique to the tip of the X chromosome. Certainly, the absence of any reverse polarity (distal to proximal decrease in crossing over) in +/Df (3R)sbd^{105} heterozygotes (cf. Tables 6a and 6b) argues that both the decrease in crossing over (ca. 30% of control) and the absence of polarity are real. The slight decrease in crossing over in mutator heterozygotes, ca. 6–11%, lends further evidence for the semi-dominant effect of the mutator.

In Table 7, the tetrad analysis of the crossing over data (after Weinstein, 1936) is shown. As observed, the mutator-induced reduction in crossing over results in an increase of E_0's at the expense of both E_1's and E_2's, suggesting a generalized decrease in total crossing over along the entire X chromosome.

The observed crossing over reductions in and the frequency of specific tetrads recovered from the heterozygous-deficiency genotype (+/Df(3R)sbd^{105}) point to a further difference between the meiotic effect of the deficiency observed here and that reported by other investigators. Both Hinton (1966) and Lindsley *et al.* (1968)

Table 5a. Crossover and non-crossover F_1 ♂♂ recovered from crosses of ♀♀ *y²cv ct⁶*/+++; chromosome 3 genotype to ♂♂ *sn³*. Distal region

Interval[a]	Chromosome 3 genotype			
	+/+	+/*mu*	+/Df(3R)*sbd*^{105}	*mu*/Df(3R)*sbd*^{105}
0	3407	3967	4533	2823
1	656	676	715	359
2	369	354	374	190
1, 2	6	5	15	1
N	4438	5002	5637	3373
Σ XO ♂♂[b]	4	6	17	25
Freq. XO ♂♂[c] × 10^3	0.9	1.2	3.0	7.4

[a] Interval: 0 = non-crossovers; 1 = single crossovers in *y²-cv*; 2 = single crossovers in *cv-ct⁶*; and 1, 2 = double crossovers.
[b] XO ♂♂ = *sn³*/O ♂♂ exceptions from X non-disjunction in P_1 females. All *sn³*O ♂♂ were tested for expected sterility.
[c] Freq. XO ♂♂ = Σ *sn³*/O ♂♂/Σ ♂♂ scored.

Table 5b. Crossover and non-crossover F_1 ♂♂ recovered from crosses of ♀♀ *lz*^{50e}*v g odsy*/++++; chromosome 3 genotype × ♂♂ *sn³*. Proximal region

Interval	Chromosome 3 genotype			
	+/+	+/*mu*	+/Df(3R)*sbd*^{105}	*mu*/Df(3R)*sbd*^{105}
0	4158	3868	4786	2159
3	312	269	301	122
4	581	484	531	197
5	642	538	531	187
3, 4	3	3	12	2
4, 5	7	11	15	4
3, 5	16	20	13	3
3, 4, 5	1	1	1	0
N	5720	5194	6190	2674
Σ XO ♂♂[b]	3	6	16	21
Freq. XO ♂♂[c] × 10^3	0.5	1.2	2.6	7.9

[a] Intervals; 0 = non-crossovers; 3 = single crossovers in *lz*^{50e}-*v*; 4 = single crossovers in *v-g*; 5 = single crossovers in *g-odsy*; 3, 4 = double crossovers; 4, 5 = double crossovers; 3, 5 = double crossovers; and 3, 4, 5 = triple crossovers.
[b] XO ♂♂ = *sn³*/O ♂♂ exceptions from X non-disjunction in P_1 females. All *sn³*/O ♂♂ were tested for expected sterility.
[c] Freq. XO ♂♂ = Σ *sn³*/O ♂♂/Σ ♂♂ scored.

found a 50–67% reduction in X chromosome crossing over in +/Df(3R)*sbd*^{105} heterozygotes; and further, that the frequency of E_0's was increased at the expense of E_2's, but not E_1's. Summing the results of Tables 6a, 6b and 7 for the heterozygous deficiency demonstrates a net reduction in crossing over of roughly 15% over the entire X chromosome, the reduction occurring primarily at the expense of E_1's.

Table 6a. Crossover frequencies (%) with 95% confidence limits[b] in each chromosome 3 genotype for the intervals y^2-*cv*, *cv*-ct^6 and y^2-ct^6

Chromosome 3 genotype	Interval[a]			% of control (+/+) interval 1+2
	1	2	1+2	
+/+	14.92 (13.87–16.01)[b]	8.45 (7.64–9.31)	23.37 (22.12–24.65)	100
mu/+	13.62 (12.69–14.59)	7.18 (6.47–7.94)	20.79 (19.68–21.94)	89
+/Df(3R)sbd^{105}	12.95 (12.08–13.86)	6.90 (6.25–7.60)	19.85 (18.82–20.91)	85
mu/Df(3R)sbd^{105}	10.67 (9.65–11.76)	5.66 (4.88–6.50)	16.34 (15.10–17.63)	70

[a] Intervals: 1 = y^2-*cv*; 2 = *cv*-ct^6.
[b] Parentheses indicate 95% confidence limits computed from the standard error following the method and corrections of Stevens (1942).

Table 6b. Crossover frequencies (%) with 95% confidence limits[b] in each chromosome 3 genotype for the intervals lz^{50e}-*v*, *v*-*g*, *g*-*odsy* and lz^{50e}-*odsy*

Chromosome 3 genotype	Interval[a]				% of control (+/+) for 3+4+5
	3	4	5	3+4+5	
+/+	5.82 (5.23–6.46)[b]	10.35 (9.58–11.16)	11.63 (10.81–12.48)	27.80 (26.24–28.97)	100
+/*mu*	5.64 (5.03–6.30)	9.61 (8.82–10.44)	10.97 (10.14–11.85)	26.22 (25.03–27.44)	94
+/Df(3R)sbd^{105}	5.28 (4.75–5.86)	9.03 (8.34–9.76)	9.05 (8.33–9.80)	23.36 (22.31–24.44)	84
mu/Df(3R)sbd^{105}	4.75 (3.98–5.62)	7.59 (6.60–8.66)	7.26 (6.30–8.30)	19.60 (18.10–21.16)	70

[a] Intervals: 3 = lz^{50e}-*v*; 4 = *v*-*g*; 5 = *g*-*odsy*.
[b] Parentheses indicate 95% confidence limits computed from the standard error following the method and corrections of Stevens (1942).

Discussion

The present studies demonstrate two new parameters of the third chromosome mutator gene, *mu*, in *D. melanogaster*. The mutator significantly alters both meiotic chromosome disjunction and crossing over in females, but not in males. Thus, in meiotic phenotype the mutator resembles several recently isolated "meiotic" mutants in *D. melanogaster* (Sandler *et al.*, 1968; Sandler and Lindsley, 1974).

The function of the mutator, however, although limited to females is not restricted to meiosis. As shown earlier (Green, 1970; Green and Lefevre, 1972; Gold and Green, 1973) the mutator is also active during mitotic cell divisions in

Table 7. Tetrad analysis[a] of crossing over between the intervals y^2-*cv*-ct^6 and lz^{50e}-*v*-*g*-*odsy* in the tested chromosome 3 genotypes

Chromosome 3 genotype	E_0	E_1	E_2	E_3
1. y^2-*cv*-ct^6				
+/+	0.538	0.457	0.005	
+/*mu*	0.588	0.408	0.004	
+/Df(3R)sbd^{105}	0.614	0.376	0.010	
mu/Df(3R)sbd^{105}	0.675	0.324	0.001	
2. lz^{50e}-*v*-*g*-*odsy*				
+/+	0.463	0.520	0.016	0.001
+/*mu*	0.503	0.472	0.024	0.001
+/Df(3R)sbd^{105}	0.559	0.416	0.024	0.001
mu/Df(3R)sbd^{105}	0.662	0.365	0.013	0.000

[a] Method after Weinstein (1935).
E_0 = No exchange tetrad; E_1 = Single exchange tetrad; E_2 = Double exchange tetrad; and E_3 = Triple exchange tetrad.

both premeiotic germinal and somatic tissues. More recently, Martensen and Green (unpublished data) have obtained evidence that the mutator alters mitotic recombination as well.

As discussed by Green and Lefevre (1972), speculation about specific mutator cellular function on purely genetic grounds is only conjecture. However, given the mutator phenotypes described to date (including the results here), it is not difficult to imagine the mutator function as part of the cellular mechanism(s) involved in DNA repair and/or DNA replications. This was suggested by Green (1970) and Green and Lefevre (1972) from analogy with both microbial mutators (Speyer, 1965; Drake *et al.*, 1969) and mutants of *E. coli* defective in DNA polymerase (Coukell and Yanofsky, 1969). This suggested mutator function is not directly contradicted by the results presented here, provided at least one assumption is met. That is, it must be assumed that the same mechanisms of DNA repair and/or replication which operate at mitotic cell divisions, function similarly during meiosis. Since it is known that crossing over in higher organisms requires chromosome breakage and reunion (Creighton and McClintock, 1931; C. Stern, 1931), some replication and/or repair must occur during meiosis. Data directed to this point are afforded by the elegant studies of H. Stern and co-workers (see Stern and Hotta, 1974) on *Lilium* microsporocytes. Here, it was found that several enzymes presumably involved in DNA excision-repair accumulate during the short period of non-conservative DNA synthesis in meiotic prophase. By analogy, a similar situation likely exists for *D. melanogaster*, *i.e.*, enzymes promoting DNA repair and/or replications during mitotic divisions function similarly at meiosis.

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