Genet Sel Evol (1993) 25, 421-434

Original article

A genetic evaluation of male reproductive fitness at early and late age in *Drosophila melanogaster* treated with a mutagen

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(Received 26 February 1992; accepted 2 June 1993)

Summary – The effect of ethyl methane sulfonate-induced mutations in different germ cell stages on male reproductive fitness at early and late age, compared to an untreated control, was investigated in a laboratory population of *Drosophila melanogaster*. Indication of active DNA repair processes after mutagen treatment was obtained in the pre-meiotic germ cell stages. Genetic parameters for the male fitness trait, *ie* "number of progeny" were estimated in a succession of different broods at early and late ages. Heritability estimates for progeny size were found to vary between 0.13 and 0.97 in the different brood stages and over the 2 treatment groups. The estimates of genotype–environment interaction, as well as genetic correlations, suggest that the genetic determination of progeny size is different at an early age between EMS-treated and untreated individuals, but not at late ages.

reproductive fitness / *Drosophila melanogaster* / genetic parameter / ageing / mutagen

Résumé – Évaluation génétique des capacités de reproduction de mâles de différents âges exposés à un agent mutagène chez *Drosophila melanogaster*. *Les effets des mutations induites par l'éthyl méthanesulfonate (EMS) au cours des différentes phases de la gamétogenèse sur la capacité de reproduction de mâles, jeunes ou âgés, ont été étudiés sur une population de laboratoire de* Drosophila melanogaster. *Des processus actifs de réparation de l'ADN, après traitement par l'EMS, existent vraisemblablement au cours des phases préméiotiques. Les paramètres génétiques relatifs au nombre de descendants par mâle ont été estimés dans plusieurs séries de ponte correspondant à différents âges. Les estimations de l'héritabilité de ce caractère varient de 0,17 à 0,67 dans les différentes séries de ponte et dans les deux groupes de mâles traités et non traités. Les estimations des interactions génotype-milieu, ainsi que des corrélations génétiques suggèrent que le déterminisme génétique du nombre de descendants est différent chez les jeunes mâles*

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exposés à l'EMS par rapport aux mâles non traités. En revanche, aucune différence n'est détectée chez les mâles plus âgés.

capacité de reproduction / *Drosophila melanogaster* / paramètre génétique / vieillissement / mutagène

INTRODUCTION

Drosophila melanogaster is widely used to evaluate genetic damage resulting from exposure to chemical mutagens. Several standard techniques for feeding adult flies with mutagenic substances (Lewis and Bacher, 1968; Felix, 1971) can be used to induce a spectrum of relevant genetic damage in the different germ cell stages. To evaluate such genetic damage, assay test systems for the induction of recessive lethals in the X-chromosome, which represent one-fifth of the *Drosophila* genome, are the simplest and most commonly used. One characteristic feature of chemical mutagens is their specificity of action. In cases of very pronounced stage specificity, testing only one germ cell stage can lead to false negative results (Wurgler *et al*, 1984). In *Drosophila*, sensitivity differences between germ cell stages can be assessed by mating treated males to virgin females in a succession of different broods.

The assay systems often used in mutation research focus on standard genetic endpoints, *ie*, point mutations with major and discrete effects and chromosomal aberrations. However, when considering mutations affecting the polygenic systems of fitness characters, quantitative genetic analysis can contribute important information to the understanding of these mechanisms (see *eg* Ramel, 1983).

The genetic effect on male fertility after mutagen treatment in a succession of different broods depends on several factors such as: 1) the ability of the mutagen to reach the germ cells; 2) the kind of damage caused by the mutagen on the germ cells, which is dose-dependent for many mutagens; and 3) the extent to which this damage is eliminated through various repair mechanisms.

Ethyl methane sulfonate (EMS) is a known mutagen which reaches all germ cell stages. EMS produces alkylated purine adducts on the DNA in germ cells. These alkylated purine adducts are readily removed by excision repair systems. However, in late stages of post-meiotic cells, the repair ability is deficient (Sega, 1979; Sobel, 1972). In contrast to late stages of post-meiotic germ cells, pre-meiotic cells are believed to have DNA repair enzymes. Indications of efficient DNA repair systems have been found in spermatogonial germ cells of *Drosophila* (Smith *et al*, 1983; Vogel and Zijlstra, 1987) and mouse (Russel, 1986). In addition to DNA repair, segregational elimination of deleterious mutations during meiosis (germinal selection) seems to reduce the realization of EMS-induced genetic damage in pre-meiotic germ cells of *Drosophila* (Vogel and Zijlstra, 1987). An equal reduction in the number of both female and male offspring and a high sterility among individuals from pre-meiotic cells is an indication of germinal selection.

The objective of this study was to explore the effect of EMS-induced mutations in different germ cell stages on male reproductive fitness at early and late ages. Further, we investigated whether male reproductive fitness is a genetically different

trait after EMS treatment compared to normal reproductive fitness. Quantitative genetic parameters for number of offspring as well as more standard genetic endpoints (*eg*, sex proportion) were estimated in a succession of different broods at early and late age after an initial treatment with EMS and compared to a control not exposed to EMS.

MATERIAL AND METHODS

The population of *Drosophila melanogaster* used in this study was obtained from crosses between 4 laboratory wild-type strains of different origin, each contributing equally to a 4-way hybrid strain. This hybrid strain, consisting of > 400 individuals of each sex per generation, was allowed to attain linkage equilibrium through > 30 generations of random mating. A sample of 26 sires and 78 dams were taken at random and each sire was mated with 3 dams. The sons from these matings ($\approx$ 8 per dam) were collected within 12 h in order to obtain approximately the same stage of sexual maturity. All sons were kept in vials containing 2 cm standard medium (10 g agar, 60 g syrup, 50 g baker's yeast, 40 g powdered mashed potatoes, 0.75 g ascorbic acid and 2 ml propionic acid per l water). The flies were maintained in an incubator at 25°C and 55% relative humidity. Photoperiod was 16L:8D. All handling was performed at room temperature using carbon dioxide anaesthesia.

The sons in each full sib group were kept together until treatment, *ie* 3 d after eclosion. Half the number of sons from all full sib groups were individually exposed to EMS for 24 h using the method described by Lewis and Bacher (1968), but with a lower EMS concentration (5.0×10^{-3} M). The other half, the control group, was treated in the same manner, except that no EMS was added to the medium. Immediately after treatment each son was placed in a vial with 3 virgin "attached-X" ($\widehat{XX}$) females and kept in these vials for 2 consecutive egg-laying days. Each son was then transferred to a new vial with a new set of 3 virgin $\widehat{XX}$-females for another egg-laying period of 2 d, and the former set of $\widehat{XX}$-females were discarded. This procedure was repeated 5 times with egg-laying periods starting at 4, 6, 8, 10 and 12 d after eclosion and representing fertility at an early age.

Sex chromosomes of $\widehat{XX}$-females consist of 2 X-chromosomes and 1 Y-chromosome. The 2 X-chromosomes are attached to each other and segregate together during meiosis. Due to the genetic constitution of $\widehat{XX}$-female, male offspring of these females get their X-chromosomes from the sire (see fig 1). Thus, the proportion of male-to-female offspring from the cross between a wild-type male and an $\widehat{XX}$-female reflects the genetic load in the sire X-chromosome.

Each successive brood constitutes a sample of germ cells that received EMS at different stages of spermatogenesis. Thus, the first brood was produced from cells that were mature sperm at the time of treatment; the second brood from late spermatids; the third brood, early spermatids; the fourth brood, meiotic stages and the fifth brood from spermatogonia. During the egg-laying period in brood 5 starting at d 12, son groups were kept in vials for 3 consecutive d instead of 2. This was done in order to prolong the pre-meiotic period so that germ cells of all sons in brood 5 had reached the spermatogonial stage, since there is individual variation in the rate of spermatogenesis (Wurgler *et al*, 1984). Following the fifth

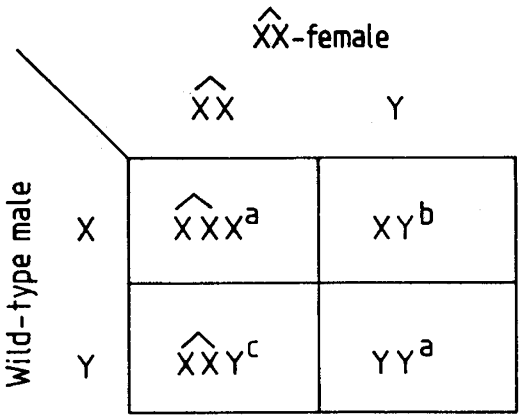

Fig 1. The distribution of sex chromosomes in parental gametes and the resulting constitution of sex chromosomes in embryos a: lethal combination of sex chromosomes, b: viable wild-type male embryo, c: viable $\widehat{XX}$-female embryo.

mating period, all sons were placed separately in new vials, twice a week, for 20 d. After the ageing period each son was mated with a new set of 3 virgin $\widehat{XX}$-females for 2 consecutive egg-laying d as described above. This procedure was repeated once more. Broods 6 and 7 represent fertility at a late age (35 and 37 d after eclosion). All $\widehat{XX}$-females used in the experiment were between 3 and 5 d of age. The total number of offspring from each son, the sex proportion (number of males divided by total number of progeny in each brood) and the proportion of sterile sons were calculated for each brood. In order to discriminate between males' and females' designation in different generations, a schematic representation of the experimental design is shown in figure 2.

Genetic parameters for number of progeny in the different broods within the 2 treatment groups were calculated by the method of multivariate-restricted maximum likelihood, using a random animal model with breeding value of sons as the only factor. A relationship matrix was used to take into account the covariance between relatives (Meyer, 1986). A restriction imposed was that only sons present in all broods within a treatment and having adult offspring in brood 1 were included in the analysis. Standard errors were calculated according to Meyer (1985, 1986).

In order to estimate genotype–environment interactions and genetic correlations for number of offspring between the 2 treatment groups, the appropriate variance components were also estimated (SAS Inc, 1982) using the model:

$$Y_{ijk} = \mu + B_i + f_j + (Bf)_{ij} + e_{ijk}$$

where:

Y_{ijk} = observed number of offspring;
μ = general mean;
B_i = fixed effect of the ith treatment ($i = 1, 2$);

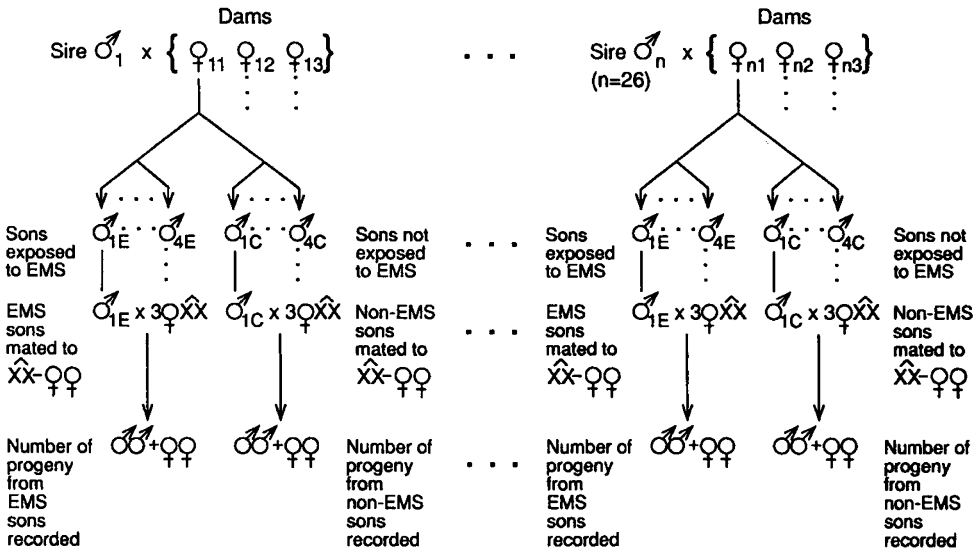

Fig 2. Schematic experimental procedure.

f_j = random effect of fullsib group ($j = 1 \ldots 78$), with mean 0 and variance σ_f^2;

$(Bf)_{ij}$ = interaction effect between fullsib group and treatment, with mean 0 and variance σ_{Bf}^2;

e_{ijk} = random residual effect associated with the ijkth record, with mean 0 and variance σ_e^2.

Genetic correlations between the 2 treatment groups for number of offspring were calculated according to Yamada (1962).

RESULTS

Number of progeny

The mean values for female and male progeny in the different broods are presented in table I. Total number of offspring after EMS treatment was significantly lower than the control for broods 1–5 ($p < 0.001$), and brood 6 $p < 0.01$), but not brood 7. However, the effect of treatment was consistently larger in males than in females. Within the control group, the total number of progeny in brood 1 was significantly lower ($p < 0.001$) than the other stages at an early age (3–14 d). In the EMS-treated group there was a considerable variation in this trait.

Sex proportion

The difference in sex proportion (table II) between the control and the EMS treated group is high in broods 1 to 3 (13.13–14.38), whereas only small differences remain in

Table I. Means for the traits female progeny size and male progeny size.

Egg laying period (brood)	*Age (d)*	*Control*			*EMS treatment*			*Difference* [a]	
			Females	*Males*		*Females*	*Males*	*Females*	*Males*
		N	$\overline{X}_{Cf}$	$\overline{X}_{Cm}$	N	$\overline{X}_{Ef}$	$\overline{X}_{Em}$	$\overline{X}_{Cf} - \overline{X}_{Ef}$	$\overline{X}_{Cm} - \overline{X}_{Em}$
1	3–4	356	21.11	29.73	348	19.04	15.80	2.07	13.93
2	5–6	351	27.50	38.55	335	21.20	17.19	6.30	21.36
3	7–8	338	25.64	37.19	330	23.60	19.17	2.04	18.02
4	9–10	331	25.63	37.72	316	1.80	2.38	23.83	35.34
5	11–13	321	22.54	36.75	306	9.16	11.72	13.38	25.03
6	35–36	236	8.61	12.08	222	6.10	8.32	2.51	3.76
7	37–38	219	2.06	3.38	202	1.60	2.09	0.46	1.29

N: number of sons tested; $\overline{X}_{Cf}$: mean for the trait female progeny size in the control group; $\overline{X}_{Cm}$: mean for the trait male progeny size in the control group; $\overline{X}_{Ef}$: mean for the trait female progeny size in the EMS-treated group; $\overline{X}_{Em}$: mean for the trait male progeny size in the EMS-treated group; [a] the difference between total number of progeny $[(\overline{X}_{Cf} + \overline{X}_{Cm}) - (\overline{X}_{Ef} + \overline{X}_{Em})]$ for the control and the EMS-treated group in different broods was significant except for brood 7.

broods 4 to 7 (0.68–5.82). Within the control group the variation in sex proportion was small between different broods. This finding is consistent with an earlier study by Björklund *et al* (1988) where sex ratio was calculated at 3 different age periods and no significant differences were obtained. In the EMS treated group, a lower sex proportion was obtained in broods 1 to 3 (44.78–45.35%) than in broods 4 to 7 (56.16–57.70%). The sex proportion in broods 1 to 3 after EMS treatment were on average 23% lower than the same broods in the control group, which is parallel to an investigation by Vogel and Natarajan (1979). At the same concentration of EMS used as in this investigation, frequency of recessive lethal mutations was found by them to be $\approx$ 20%.

Sterility

Within the control group the proportion of sterile sons increased linearly from 1.7% in the first brood to 84.9% in the seventh brood (table II). In the EMS-treated group the proportion of sterile sons increased from its minimum value, 2.1%, in the second brood to 89.1% in the seventh brood. Brood 4 deviated from this pattern with a considerably higher proportion of sterile sons (65.4%).

Quantitative genetic parameters

Heritabilities as well as genetic and phenotypic correlations for total number of offspring in the different broods of the control group are presented in tables III and IV for the EMS-treated group. Genotype–environment interactions and genetic correlations estimated between the 2 treatment groups and within brood stage are presented in table V. Due to the small number of progeny obtained (brood 7 in the control and the EMS group; brood 4 in the EMS-treated group), brood 7 was

Table II. Means for the traits sterility and sex proportion.[a].

Egg-laying period (brood)	*Age (d)*	*Control*			*EMS treatment*			*Difference*	
		N	*Sterility (%)* $\overline{X}_{CS}$	*Sex proportion (%)* $\overline{X}_{CP}$	N	*Sterility (%)* $\overline{X}_{ES}$	*Sex proportion (%)* $\overline{X}_{EP}$	*Sterility (%)* $\overline{X}_{CS}$-$\overline{X}_{ES}$	*Sex proportion (%)* $\overline{X}_{CP}$-$\overline{X}_{EP}$
1	3–4	356	1.68	58.48	348	4.02	45.35	− 2.34	13.13
2	5–6	351	1.99	58.36	335	2.09	44.78	− 0.10	13.58
3	7–8	338	2.66	59.19	330	4.24	44.81	− 1.58	14.38
4	9–10	331	6.95	59.55	316	64.54	56.80	− 57.59	2.75
5	11–13	321	10.28	61.98	306	29.08	56:16	− 18.80	5.82
6	35–36	236	63.98	58.38	222	70.72	57.70	− 6.74	0.68
7	37–38	219	84.93	62.13	202	89.11	56.64	− 4.18	5.49

[a] Sex proportion calculated as (number of males divided by total number of progeny in each brood)×100; N: number of sons tested; $\overline{X}_{CS}$: mean for the trait sterility in the control group; $\overline{X}_{CP}$: mean for the trait sex proportion in the control group; $\overline{X}_{ES}$: mean for the trait sterility in the EMS treated group; $\overline{X}_{EP}$: mean for the trait sex proportion in the EMS treated group.

excluded from all analysis of quantitative genetic parameters, whereas brood 4 was only excluded from the analysis of genotype–environment interactions, genetic correlations estimated between the 2 treatment groups and genetic parameters estimated within the EMS-treated group.

Heritabilities

The heritability for number of progeny in the first brood was very high both for the control (0.79) and for the EMS-treated group (0.97). In broods 2 to 6 heritabilities varied from 0.17 to 0.66 in the control group and from 0.13 to 0.67 in the EMS-treated group, but were not significant in broods 3 and 6 for either the control or the EMS-treated groups.

Genetic correlations within treatment

A clear pattern of genetic correlations was not discernible within early age broods, nor between early and late age broods, and most genetic correlations were not significant in the control group. Most of the phenotypic correlations were significant and positive. In the EMS-treated group, there were significant positive genetic correlations between broods close to each other in time, except for the correlation between brood 1 and brood 2, which was negative. Most phenotypic correlations in the EMS-treated group, although lower than the phenotypic correlations in the control group, were positive and significant.

Genotype–environment interaction and genetic correlations between treatments

Genotype–environment interactions in broods 1 and 2 were significant at the $P < 0.0001$ level and in brood 5 at the $P < 0.01$ level. There were no significant genotype–environment interactions in broods 3 and 6. The method used to calculate genetic correlations within brood stage for number of offspring between the 2 treatment groups gave a correlation of -1.00 in brood 1 and 1.02 in brood 6.

DISCUSSION

Several experiments have investigated the genetic covariation of fitness traits in populations not exposed to mutagens. According to Falconer (1981), at a negligible rate of mutations, additive genetic variance in major components of fitness can be maintained only in the presence of a negative genetic correlation. However, both negative (Rose and Charlesworth, 1981; Luckinbill *et al*, 1984; Tucic *et al*, 1988) and positive genetic correlations (Giesel 1986; Engström *et al*, 1989) have been found among fitness traits measured at different life history stages. Classical quantitative genetic theory generally assumes that inbreeding causes spurious patterns of positive genetic correlations (Rose, 1984). However, genetic correlations do not measure the inherent action of genes, but rather provide statistical descriptions of populations within particular contexts of environments and genotypic frequencies. Therefore, it may not be meaningful to discuss genetic correlations for life history fitness components found in different investigations (Clark, 1987).

Table III. Heritabilities (diagonal), genetic correlations (lower triangle) and phenotypic correlations (upper triangle) with their SEs for the trait "number of progeny" at different brood stages in the control group. The analysis is based on 232 sons tested.

Brood	*Brood*					
	1	*2*	*3*	*4*	*5*	*6*
1	0.79 ± 0.10	0.06 ± 0.06	0.15 ± 0.06	− 0.07 ± 0.07	− 0.01 ± 0.05	0.01 ± 0.05
2	− 0.38 ± 0.21	0.43 ± 0.12	0.25 ± 0.06	0.14 ± 0.06	0.15 ± 0.06	0.02 ± 0.06
3	− 0.20 ± 0.24	− 0.17 ± 0.31	0.26 ± 0.16	0.14 ± 0.07	0.15 ± 0.07	0.05 ± 0.07
4	− 0.18 ± 0.25	0.26 ± 0.20	− 0.62 ± 0.29	0.66 ± 0.16	0.51 ± 0.04	0.18 ± 0.06
5	0.01 ± 0.21	0.03 ± 0.27	− 0.64 ± 0.40	0.44 ± 0.20	0.33 ± 0.15	0.30 ± 0.06
6	0.24 ± 0.25	0.11 ± 0.34	− 0.94 ± 0.16	0.41 ± 0.30	0.79 ± 0.33	0.17 ± 0.12

Table IV. Heritabilities (diagonal), genetic correlations (lower triangle) and phenotypic correlations (upper triangle) with their SEs for the trait "number of progeny" at different brood stages in the EMS-treated group. The analysis is based on 218 sons tested.

Brood	*Brood*				
	1	*2*	*3*	*5*	*6*
1	0.97 ± 0.01	− 0.01 ± 0.03	0.03 ± 0.01	− 0.01 ± 0.02	− 0.04 ± 0.01
2	− 0.35 ± 0.16	0.24 ± 0.13	0.18 ± 0.07	0.16 ± 0.07	0.07 ± 0.07
3	0.08 ± 0.03	0.87 ± 0.15	0.13 ± 0.11	0.12 ± 0.07	0.05 ± 0.07
5	0.00 ± 0.03	0.54 ± 0.28	0.75 ± 0.36	0.67 ± 0.17	0.26 ± 0.07
6	− 0.13 ± 0.07	0.07 ± 0.47	0.29 ± 0.59	0.62 ± 0.31	0.18 ± 0.14

Table V. Genotype–environment interaction and genetic correlations for number of progeny between the 2 groups of treatment and within brood stage.

		Genotype–environment interaction		
Brood	N	*Genetic correlation*	F-*value*	*Prob* < F
1	348	− 1.00	4.03	0.0001
2	335	− 0.85	1.80	0.0001
3	330	0.67	1.10	0.2574
5	306	− 0.26	1.51	0.0030
6	222	1.02	0.83	0.8540

N: number of sons tested.

This study deals with the effect of EMS-induced genetic damages in germ cells on male reproductive fitness at early and late age. The EMS concentration used in this investigation is not expected to produce genetic changes other than point mutations, unless sperm is being stored (Vogel and Natarajan, 1979). Due to the genetic constitution of the $\widehat{XX}$-females, male offspring from these females get their X-chromosome from their father. Thus, X-linked deleterious mutations affecting viability will reduce the number of male progeny and result in female-biased sex proportion.

At an early age, broods 1 to 3 after EMS treatment are similar in that they have lower sex proportion and rates of sterility approximately twice that of the untreated flies. The reduced number of male offspring accounts for most of the reduction in total number of progeny, indicating that the induced mutations are mainly recessive. Almost normal sex proportion in broods 4 and 5 after EMS treatment suggests that efficient DNA repair processes exist in the meiotic and spermatogonial germ cell stages. The reduction in both female and male offspring and the higher sterility in broods 4–5 indicate that germinal selection is an important mechanism in eliminating deleterious mutations. Vogel and Zijlstra (1987) reported similar results.

At later ages, differences in number of offspring and sterility between the EMS-treated group and the control group are smaller, and no significant difference in

number of offspring was obtained for brood 7. However, the sex proportion is slightly lower in the EMS-treated group. These results suggest that most, but not all, of the deleterious mutations induced early in life are eliminated.

Heritability estimates found in the literature for fecundity of female *Drosophila* vary between 0.02 and 0.40 (Rose and Charlesworth, 1981; Tucic *et al*, 1988; Engström *et al*, 1989). The total number of progeny in brood 1 had a higher estimated heritability than was expected. Furthermore, there were significantly fewer offspring in brood 1 of the control group than in the other early age brood stages. This and the negative correlation between brood 1 and brood 2 suggest that number of progeny in brood 1 is a trait that includes both fertility and sexual maturity. High heritability estimates have been found for sexual maturity in other species, such as for example laying hens (Liljedahl *et al*, 1984).

The heritability estimates obtained in the other brood stages are, on average, higher than estimates of female fecundity reported in the literature. Heritability for the male reproductive component was about twice the size of the heritability of female fecundity in an investigation reported by Tucic *et al* (1988). Brittnacher (1981) found that genetic load due to virility was greater than that due to the female reproductive component. This suggests that the male reproductive component may contain more genetic variance than the female component. Heritability at late ages (brood 6) is not significant, probably due to increased environmental variation as well as non-additive genetic variation. This has been observed in several studies where the animals' reactions to stressful environments and different ages has been investigated (Flock, 1977; Liljedahl *et al*, 1984; Rose and Charlesworth, 1981; Burla and Taylor, 1982).

As only a few of the genetic correlations estimated were significant, caution is necessary when interpreting the covariance structure among fitness components. However, many fewer negative genetic correlations were obtained in the EMS-treated group than in the control group. Whether the difference in genetic correlations between the control and the EMS-treated group is due to sampling variance or caused by EMS treatment cannot be clearly distinguished in the present investigation, but this difference is believed to be an effect of EMS, because of the smaller sampling variances in EMS-treated group.

The genetic correlations estimated in this experiment were based on reproduction data from *Drosophila* males. However, most other studies which found significant covariance structures among life history fitness components have used *Drosophila* females (Rose and Charlesworth, 1981; Luckinbill *et al*, 1984; Giesel, 1986; Engström *et al*, 1989). Tucic *et al* (1988) estimated genetic correlations of life history fitness components on both sexes and concluded that the genetic covariance structure between fitness components differs between sexes.

At an early age, genotype–environment interactions as well as genetic correlations between the 2 treatment groups and within broods vary depending on the different genetic events in spermatogenesis. In the post-meiotic period, (broods 1–3) highly significant genotype–environment interactions were obtained for number of progeny in broods 1 and 2, which is indicated by high negative genetic correlations. No significant genotype–environment interaction was obtained in brood 3 and the genetic correlation was moderate to high and positive. This finding is unexpected, because the sex proportion, sterility rate and total number of progeny obtained in

the 3 broods were all very similar. A possible explanation for the high negative correlations found in broods 1 and 2 could be variation in consumption and uptake of EMS, which may be negatively coupled to fertility in the sense of sexual activity. More sexually active males may require greater amounts of energy and therefore consume more EMS, which in turn could lead to a smaller number of progeny as a result of more serious DNA damage in the germ cells. In the spermatogonial stage (brood 5) a significant genotype–environment interaction, as well as a low negative genetic correlation was obtained. This, together with an almost restored sex proportion and reduced number of progeny suggests that DNA repair and germinal selection are interfering with "normal" fertility.

At a late age (brood 6), no significant genotype–environment interactions were observed and the genetic correlation between the 2 treatment groups was high and positive. This finding suggests that the same set of genes are determining male fertility measured as "number of progeny" in the 2 treatment groups, which is confirmed by the fact that sex proportion and sterility are restored to an almost normal level and the difference in number of progeny is diminishing.

We conclude that DNA repair processes are actively present in the meiotic and earlier stages after an initial EMS treatment, because sex proportion was restored to an almost normal level in these germ cell stages. The heritability for number of progeny in brood 1 is very high for both the EMS treated group and the control. This finding suggests that number of progeny includes both fertility and sexual maturity. In the other brood stages, heritabilities were found to vary between 0.17 and 0.67. Finally, we suggest that the trait "number of progeny" at an early age is, to a great extent, genetically expressed in the EMS-treated individuals differently from the untreated control. This conclusion is based on the partly different pattern of genetic correlations within the 2 treatment groups, the genotype–environment interaction, and the genetic correlations between the 2 treatment groups found. At lates ages, the trait "number of progeny" is essentially expressed in the same way because the genetic correlation between the 2 treatment groups is positive and high and there is no significant genotype–environment interaction.

ACKNOWLEDGMENTS

The authors are grateful to S Johansson and A Schultz for excellent technical assistance and to H Jorjani, C Glynn and M Blackmore for helpful comments.

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