# EFFECTS OF OXYGEN ON THE INDUCTION OF VISIBLE MUTATIONS (AT THE *YELLOW*, *WHITE*, *MINIATURE* AND *FORKED* LOCI) IN *DROSOPHILA* MATURE SPERM*

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It is a well known fact that fewer chromosome aberrations are induced following X-irradiation of organisms under nitrogen condition as compared to those treated under oxygen.

Recently, Miyamoto (1978) has investigated the effects of oxygen on the induction of different kinds of *dumpy* (*dp*) mutations (*o*, *v*, *ol*, *lv*, *ov* and *olv* types; see Carlson and Oster 1962) in *Drosophila* mature sperm. He has revealed that the oxygen effects on the mutations which result often from chromosome breakage events (those of *ov* and *olv* types) are great as compared to those on the mutations which are rarely or partly associated with structural changes (those of *o*, *v*, *ol* and *lv* types).

In the course of the above study, the oxygen effects on the induction of mutations at the *yellow*, *white*, *miniature* and *forked* loci located on the X-chromosome were also elucidated. In the present report, the data on these mutations are considered in relation to the nature of their mutation.

## MATERIALS AND METHODS

The experimental procedures are entirely the same as those used by Miyamoto (1978). Briefly, 7-day-old males of Canton-S, a wild type strain of *D. melanogaster* were X-ray irradiated with an exposure of 3000 R in $O_2$ or in $N_2$, using the dose rate of about 105 R/min. (200 kV, 25 mA, filter 1.0 mm Al plus 1.5 mm Cu). The flies were pretreated with each gas for 15 min. The gasses used were oxygen (99.5% purity) and nitrogen (99.999% purity), and both were made to flow at the rate of 1.0 ℓ/min. during irradiation. Immediately after treatment, the males were mated individually with

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* This work was done at Zoological Laboratory, Faculty of Science, Hiroshima University, Hiroshima 730.

Abbreviation: OER, oxygen enhancement ratio.

four virgin females with genetic constitution, *y w m f*; *dp* for a 24 h period. This mating procedure permitted the detection of *y*, *w*, *m* and *f* mutations among the female progeny, although the stocks were employed primarily for the detection of *dp* mutations (Miyamoto 1978). The $F_1$ flies were examined for complete and mosaic mutations at the four sex-linked loci, *y*, *w*, *m* and *f*. Since the yield of mosaic mutations detected at these loci was very low, the data on these mutations were excluded from the analysis in the present report.

The statistical significance of the difference between the data obtained from these two kinds of irradiation condition were tested by using Kastenbaum and Bowman's tables (Kastenbaum and Bowman 1970).

## RESULTS AND DISCUSSION

The results of five replicate experiments on the induction of complete mutations (at the *y*, *w*, *m* and *f* loci) following an X-ray exposure of 3000 R in mature sperm under two different irradiation conditions, oxygen and nitrogen, are summarized in Table 1. Since the number of *y*, *w*, *m* and *f* mutants detected at each experiment are not so large, the mutation frequencies

Table 1. Frequencies of complete mutations (at the *y*, *w*, *m* and *f* loci) induced by 3000 R of X-irradiation under $O_2$ or $N_2$ condition in *Drosophila* mature sperm. The mutation frequencies given present the sum of those observed for the four loci

| Expt. No. | Mutation frequency (%) | | P($O_2$ versus $N_2$ 2-sided test) |
|---|---|---|---|
| | Under $O_2$ condition | Under $N_2$ condition | |
| 1 | 0.3688 (6/1627) | 0.1323 (4/3023) | |
| 2 | 0.6889* (18/2613) | 0.1740 (13/7473) | |
| 3 | 0.6747** (9/1334) | 0.1760 (8/4545) | |
| 4 | 0.6472* (10/1545) | 0.0993 (4/4028) | |
| 5 | 0.4479* (12/2679) | 0.0957 (5/5227) | |
| Total | 0.5613 (55/9798) | 0.1339 (34/24296) | <0.001 |

*, ** Significant at 5 and 1% level from the control, respectively.

in the table are given as the sum of those observed for these four mutants. As can be seen from the table, the total frequency of 0.5613% under oxygen is significantly higher than that of 0.1399% observed after the same exposure under nitrogen; the OER is thus 4.01. Throughout the 5 replicates, the frequencies of these mutations are consistently higher in oxygen series than in nitrogen. Moreover, in 4 out of the 5 replicates (Expt. Nos. 2, 3, 4 and 5) the yield of such mutations under oxygen condition is significantly higher than that under nitrogen. These findings indicate that the yield of *y*, *w*, *m* and *f* mutations induced by 3000 R of X-rays is affected to a considerable extent by the difference in the irradiation condition such as oxygen and nitrogen.

In this context, it is perhaps worthy to compare the OER value estimated for the *y*, *w*, *m* and *f* mutations with that for the other kinds of genetic effects reported thus far in the literature. This is summarized in Table 2.

Table 2. The oxygen enhancement ratios (OER) for the induction of various kinds of genetic effects in *Drosophila* mature sperm

| Effects | Treatment | OER | References |
|---|---|---|---|
| *dp* mutations (complete) | 2000 R | 5.3 | Oster (1963) |
| *dp* mutations (mosaic) | 2000 R | 1.2 | Oster (1963) |
| *dp* mutations (complete) | 3000 R | 2.6 | Miyamoto (1978) |
| *dp* mutations (mosaic) | 3000 R | 1.1 | Miyamoto (1978) |
| Sex-linked recessive lethal mutations | 2000 R | 2.0 | Oster (1963) |
| Sex-linked recessive lethal mutations (in ring X: *R (1) 2*) | 3000 R | 1.6-2.0 | Sobels (1965) |
| Sex-linked recessive lethal mutations (in ring X: *R (1) 2*) | 2000 R | 2.2 | Leigh (1968) |
| Translocations | 2000 R | 4.5 | Oster (1963) |
| Translocations | 2000 R | 3.6 | Leigh (1968) |
| Translocations | 3000 R | 4.5 | Leigh (1968) |
| Hyperploid males | 3000 R | 4.2 | Miyamoto (1978) |
| Partial loss of Y | 2000 R | 2.0 | Oster (1963) |
| Loss of Y (or X) | 2000 R | 2.5 | Oster (1963) |
| XO males (in ring X: *R (1) 2*) | 2000 R | 1.0 | Leigh (1968) |
| XO males (in ring X: *R (1) 2*) | 3000 R | 1.1 | Leigh (1968) |

It is evident from the table that the OER value estimated for the *dp* mosaics, the majority of which represent gene mutations not associated with detectable structural changes (Carlson and Southin 1962; Fujikawa *et al.* 1975; Inagaki *et al.* 1977; Miyamoto 1978), is nearly 1.0, and that for the translocations and the hyperploid males, both of which result from chromosome breakage and rejoining (Lefevre 1967; Auerbach 1975), fairly high OER values are estimated. The OER value estimated for the *y*, *w*, *m* and *f* mutations in the present study is nearly the same as that for the translocations. Such a correspondence in the OER values estimated between these visible mutations and the translocations possibly suggests that potential lesions leading to these two kinds of genetic effects are not different from one another.

In the meantime, several investigations on the nature of radiation-induced visible mutations, using cytological technique (salivary gland chromosome), reported that about half of them are associated with detectable chromosome aberrations, such as translocations, inversions and deficiencies (Painter and Patterson 1935; Ward and Alexander 1957; Alexander 1960; Roberts 1974). However, the proportion of visible mutations associated with structural changes seems to be greater than 50%, in view of Roberts' indications (Roberts 1974). They are: 1) the cytological analysis often cannot be made for the mutants associated with gross rearrangements because of their drastically reduced fertility; and 2) rearrangements, particularly small deficiencies, may be misclassified as cytological normal, because their size is too small to analyze within the resolving power of the light microscope. Therefore, it may be said that a large portion of the *y*, *w*, *m* and *f* mutations induced by 3000 R of X-rays is associated with structural changes of some form, thus a relatively high OER value is estimated for these mutations.

## SUMMARY

The effects of oxygen on the induction of visible mutations (at the *y*, *w*, *m* and *f* loci) following an X-ray exposure of 3000 R to mature sperm were investigated.

The results indicate that the yield of *y*, *w*, *m* and *f* mutations under oxygen condition is significantly higher than that obtained after the same exposure under nitrogen, thus a relatively high OER value of 4.01 is estimated.

This finding indicates that the magnitude of oxygen effects on the induction of the above mutations is fairly great, suggesting that the majority of them may be associated with structural changes of some form at this exposure level.

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