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Abstract

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Mutations to actidione resistance in Neurospora have been reported by Howe and Terry (1962 Can. J. Genet. Cytol. 4:447) and Hsu (1963 J. Gen. Microbiol. 32:341). Hsu determined that there were two loci for actidione resistance and that the seven mutants tested were dominant to wild type in heterocaryons. Thus, it seems probable that at least a high proportion of actidione resistant mutants are dominant. This heterocaryon dominance makes mutation to actidione resistance a potential tool in the study of mosaic mutations. Such a study is in progress in this laboratory.

Actidione resistant mutants were obtained by exposing water suspensions of microconidia from a pe, fl strain (Y8743m,L) (FGSC#867) to X-rays followed by plating the microconidia on minimal plating medium containing 2 µg/ml actidione. Actidione resistant colonies were isolated into minimal slants and incubated at 25°C for one week. Conidial suspensions of each mutant clone were prepared by adding water to the slants. A drop of this suspension was placed on a plate of minimal plating medium. This drop was serially spread over five plates without sterilizing the spreader between plates. After 4 or 5 days of incubation at 30°C, those plates having 5-200 colonies were overlayered with minimal plating medium to which 3 µg/ml of filter-sterilized actidione had been added. The plates were examined for mosaics after 5-7 additional days of incubation at 30°C. Actidione resistant colonies are recognized by their ability to continue growth after overlayering. Actidione sensitive colonies are distinguished by little or no additional growth after overlayering. The different actidione resistant isolates will produce either all actidione resistant colonies (scored as non-mosaics) or a mixture of resistant and sensitive colonies (scored as mosaics). The results of a series of X-ray induced actidione resistant mutation experiments are summarized in Table 1. In no case did the spontaneous mutants represent more than 10% of the mutants in the treated populations.

Table 1.

Dose	% Survival	No. mutants screened	No. mosaics
2000 r	97	117	4
4000 r	86	103	5
8000 r	77	116	4

Considering the duplex nature of DNA, the probable action(s) of ionizing radiations, and the low percentage kill in these experiments, the low frequency of mosaics is unexpected. These results, however, are consistent with back mutation studies using UV, X-ray and HNO_2 (Baylis, unpublished results) and with forward mutation studies using HNO_2 reported by Reissig (1963 J. Gen. Microbiol. 30:317) but inconsistent with the findings of Grigg and Sergeant (1961 Z. Vererbungslehre 92:380) and Royes (1962 NN#1:5). If defects in the screening procedure have not caused mosaics to be lost, then explanations of the low frequency of mosaics will have to consider the following: (1) the nature of the mutational event, (2) the manner in which genetic information is replicated, and (3) the action of repair mechanisms. Possible mechanisms for this failure to obtain segregation of mutation have been suggested in recent publications. A replication model in which all newly synthesized DNA strands are coded directly or indirectly from only one strand of the parent molecule has been proposed by Kubitschek (1964 Proc. Natl. Acad. Sci. U.S. 52:1374) and suggested by Witkin and Sciurella (1964 J. Mol. Biol. 8:610). The parental DNA strand complementary to the replication strand serves only as a template for messenger RNA. The concept that only one strand functions as a template for messenger RNA is supported by the findings of Guild and Robison (1963 Proc. Natl. Acad. Sci. U. S. 50:106). Suggestions for repair mechanisms consider the possibilities that (1) mistakes in repair may occur or (2) that the repair mechanism may act only to correct regions that do not have normal hydrogen bonding (Witkin and Sciurella ibid.) (Setlow 1964 J. Cell. Comp. Physiol. 64 Suppl. 1:51). The latter repair mechanism could either correct the lesion or extend the error to both DNA strands.

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