Is MS 222 Mutagenic: A Sex-Linked Lethal Test

Karen A. Fahrner
University of California San Diego
Department of Biology
La Jolla, CA. 92093

Some of you may have wondered about the anesthetic, MS 222 (3-aminobenzoic acid ethyl ester methanesulfonate, Tricaine), in regard to the safety of its use. This compound is superficially similar to ethyl methanesulfonate (EMS), a well-known mutagen, having an ethyl ester of m-aminobenzoic acid moiety in place of the ethyl ester of EMS. In consideration of the blithe use of the drug in our lab, we decided to investigate its mutagenicity, by measuring the rate of X-linked lethals arising in Drosophila melanogaster upon ingestion of MS 222.

Three concentrations of MS 222 were tested according to the procedure of Lewis and Bacher (1968): 1mM, 10mM, and 100mM. Three to five day-old wild-type adult males were placed in bottles containing tissues soaked with 1% sucrose and the solution to be tested. They were allowed to feed for 24 hours, and then were mated with virgin females of the X-chromosome balancer strain, FM7, (FM7 y^{31d} w^{a} v B; heterozygous FM7/+ females are distinguished by the effects of the dominant Bar eye factor). Offspring from paired matings of F_1 females (FM7/treated chromosome) by F_1 males (FM7/Y) were scored for the presence or absence of wild-type males, the latter indicating an X-linked lethal mutation. These were compared to the X-linked mutation rate in flies fed only 1% sucrose, and that induced by 25mM EMS.

The results are given in Table 1. MS 222 did not cause a mutation rate greater than that of the control in this experiment. Lethal and semi-lethal mutations occurred in less than 1% of the chromosomes scored. In contrast, such mutations were induced in 23% of EMS-treated chromosomes.

Thus, MS 222 does not appear to be a mutagen. It should, however, not be used with impunity. The drug can be toxic to fish and amphibia. The mode of action of MS 222 has not been well characterized, although Maeno (1966) has concluded that it inhibits neurotransmitter release at the neuromuscular junction, and suggested other means of action at other sites were possible. MS 222 is probably less toxic to mammals, since it is metabolized 40-fold more rapidly by mouse liver than frog liver (Wayson et al. 1976). However, the products of metabolism have yet to be completely identified, and those that are known (such as m-aminobenzoic acid, methane sulfonate, and acetylated derivatives) have not been tested for toxicity or mutagenicity.

References

Lewis and Bacher (1968). Drosophila Inform. Service. 43: 193.

Maeno, T. (1966). Effects of m-amino benzoic acid ethylester on neuromuscular transmission in the frog. J. Pharmacol. Exp. Therapeutics. 151: 449-455.

Wayson, K.A., Downes, H., Lynn, R.K., and Gerber, N. (1976). Studies on the comparative pharmacology and selective toxicity of tricaine methanesulfornate: metabolism as a basis of the selective toxicity in poikilotherms. J. Pharmacol. Exp. Therapeutics. 198: 695-708.

Table 1. Effects of MS 222 on the Induction of Mutations in Drosophila.

Substance Tested	#Chromosomes Tested	#X-linked Lethals	(%)	#Semilethals	(%)
Control*	666	1	(0.15)	1	(0.15)
25mM EMS	421	94	(22.00)	6	(1.00)
1mM MS 222	662	2	(0.30)	0	(0)
10mM MS 222	628	0	(0)	0	(0)
100mM MS 222	615	2	(0.33)	0	(0)

* Control was 1% sucrose; experimental substances were made up in 1% sucrose.

**Semilethals were scored for the presence of one wild-type male in 12 or more males.