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Abstract

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The suppressor method for screening forward mutants at the pyr-3N locus (Reissig 1963 J. Gen. Microbiol. 30: 317) was used to test whether or not acridine yellow is mutagenic for Neurospora. Microconidia, pre-germinated for 3 hours in liquid medium and washed, were treated with acridine yellow (5 mg/100 ml, and less than 10^9 conidia/mg acridine) in tartrate buffer, pH 5.6 at 25° C, and plated on the screening medium without washing. The treatment and all subsequent operations were performed in the dark or under a yellow light. The same procedure was followed with the control series, but omitting the acridine. Results of two experiments are shown below. Similar results were obtained with acridine orange under somewhat different conditions.

Experiment	Duration of treatment (min.)	Survival (%)	Mutants		
			Colony count	Per live treated ($\times 10^7$)	Per survivor ($\times 10^7$)
I	control	100	10	6.2	6.2
	85	2.0	5	0.6	30
	165	0.82	5	0.46	56
II	control	100	204	8.2	8.2
	210	0.52	24	0.13	26

From the results presented it appears that acridines have a slight mutagenic effect on Neurospora, although an interpretation based on differential killing is not excluded. If the mutagenic effect is real and is the consequence of base insertion and deletion like in phage (Crick, et al., 1961 Nature 192:1227), we might expect a sizable proportion of non-leaky or non-complementing mutants in the treated series. The following table shows that this is not the case.

Origin **	Number of mutants in each class*							
	Total tested	Prototrophs	Leaky		Complementation group			
			yes	no	alpha	beta	others	non-complementing
control	19	10	8	1	8	1	0	0
acridine yellow	29	19	8	2	8	2	0	0

* for description of each class and comparison with other mutagens, see Reissig (ibid.).
**Mutants from experiments I and II above.

The treated and untreated series differed, however, by the appearance in the former, but not in the latter, of morphologicals (gullivers) among the prototrophs. The significance of this observation and the nature of such mutants is currently under investigation in our laboratory.

In conclusion: no evidence was found for the induction of insertion/deletion mutants by acridines. In trying to reconcile this negative result with the occurrence of such a mechanism in phage, the following considerations should be borne in mind: - as Lerman (1963 Proc. Natl. Acad. Sci. U.S. 49:94) suggested, insertion/deletion mutants might only occur when recombinational events follow immediately after the acridine treatment; - the screening method used in this work detects forward mutation at the pyr-3 region provided it affects the N, but not the M function (Reissig, ibid.). It is conceivable that insertion/deletion mutants would be largely of the NM type. - - - Facultad de Ciencias Exactas, Peru 222, Buenos Aires.