

## SHORT PAPERS

### Conditional induction of $\lambda$ prophage in *exrA* mutants of *Escherichia coli*

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#### SUMMARY

Strains of *Escherichia coli* which contain the UV sensitivity gene *exrA*, repress UV induction of  $\lambda$  prophage in a novel manner. While *exrA*<sup>+</sup> strains can be induced by UV irradiation and express this induction in a 2 h period, *exrA* mutants delay the expression of induction for almost 4 h, and the induction maximum is approximately 10% of the wild-type value. The kinetics of the delayed induction and superinfection experiments indicate that the lifting of immunity and induction of  $\lambda$  prophage occur simultaneously.

*exr* mutants of *Escherichia coli* are sensitive to both ultraviolet (UV) and x-irradiation (Rörsch *et al.* 1966; Mattern, Zwenk & Rörsch, 1966). In this paper we are concerned with the *malB* linked *exr* gene referred to as *exrA* (Donch & Greenberg, 1968). It was reported (Donch, Greenberg & Green, 1970) that *exrA* mutants of *Escherichia coli* repressed ultraviolet (UV) induction of  $\lambda$  prophage. In this respect *exrA* resembles *recA* (Brooks & Clark, 1967). *exrA* and *recA* resemble each other further in that both suppress UV induced filamentation in *lon* strains (Donch, Green & Greenberg, 1968; Green, Greenberg & Donch, 1969) and both suppress UV induction of mutations (Witkin, 1967; Witkin, 1969). We now report on further investigations on the repression of UV induction of  $\lambda$  by *exrA*.

It was possible that  $\lambda$  was not inducible by UV in *exrA* strains because of the presence of excessive amounts of  $\lambda$  repressor. However,  $\lambda$ vir (obtained from D. Kaiser) plated with equal efficiency on *exrA* ( $\lambda$ ) and *exrA*<sup>+</sup> ( $\lambda$ ) strains. This would not be expected, if an elevated immunity system were present (Matsubara & Kaiser, 1968).

The possibility of a sterically altered repressor, which would produce a difference in the kinetics of induction of an *exrA* ( $\lambda$ ) and *exrA*<sup>+</sup> ( $\lambda$ ), was considered. This was tested by using the temperature sensitive  $\lambda$  mutant,  $\lambda$ hc 1857 (Sussman & Jacob, 1962), which was found to be heat inducible but not UV inducible in an *exrA* strain (Donch & Greenberg, 1968). No difference was observed between the kinetics of heat induction of an *exrA* ( $\lambda$ hc 1857) and *exrA*<sup>+</sup> ( $\lambda$ hc 1857). If the *exrA* gene produced an alteration in the repressor protein, one would expect this to be reflected in altered kinetics of heat induction.

The conclusions from the foregoing experiments were that repressor was present in normal amounts and was apparently structurally unaltered. It had been noted earlier (Brooks & Clark, 1967; Donch *et al.* 1970) that while both *recA* and *exrA* repressed  $\lambda$  induction, *exrA* repression was incomplete, since the spontaneous induction of an *exr* ( $\lambda$ ) strain was 10% of the wild-type value as compared to < 10<sup>-2</sup>% observed with *recA* ( $\lambda$ ) mutants. It was suspected that the repression of UV induction in *exrA* ( $\lambda$ ) strains involved an abnormality in the kinetics of induction. Unusual kinetics of induction has been reported by Iwo (1968) for Bsl (80).

In earlier experiments (Donch *et al.* 1970) streptomycin was applied in an overlay to kill the bacteria 2 h after exposure to UV (70 ergs/mm<sup>2</sup>). In experiments reported here streptomycin was added at intervals of 1 h up to 4 h after UV irradiation. During this time any induction, which might occur late because of an unusual host property, should be expressed. Fig. 1 shows the results of such an experiment. It can be seen that under these conditions evidence of induction occurred only after 2.5 h and reached a maximum of approximately 10% in 4 h. Also shown in wild-type *exrA*<sup>+</sup> ( $\lambda$ ) in which induction occurred in 30 min after UV and approached 100% by 2 h.

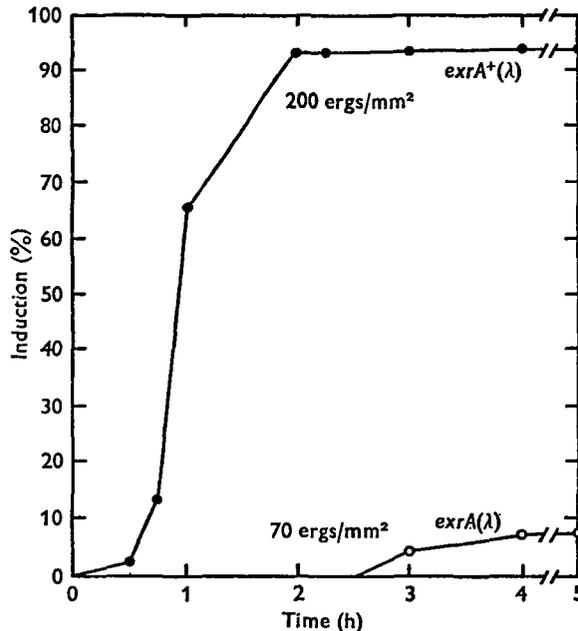


Fig. 1. Effect of time on the expression of induction of  $\lambda$  prophage following UV irradiation of *exrA* ( $\lambda$ ) and *exrA*<sup>+</sup> ( $\lambda$ ) strains

The results of these experiments indicated that in an *exrA* mutant the lifting of immunity requires a considerably longer period of time as compared to the time required for an *exrA*<sup>+</sup> strain. In order to test this hypothesis an *exrA* ( $\lambda$ ) mutant was superinfected with  $\lambda$ hc 1857 at 37 °C then irradiated with UV (70 ergs/mm<sup>2</sup>) and incubated at 42 °C. The second soft agar layer containing streptomycin was added at 1 h intervals up to 5 h. Under these conditions mottled plaques were observed at a frequency of approximately 6%, due to the lysing of induced *exrA* ( $\lambda$ ) which produce wild type (turbid plaques) and superinfecting phages,  $\lambda$ hc 1857 (clear plaques). No mottled plaques were observed earlier than 3 h. This implies that immunity to superinfection was lifted simultaneously with the occurrence of induction of prophage.

It appears that lifting of immunity (inactivation of repressor) following UV irradiation requires an extended period of time in an *exrA* ( $\lambda$ ) strain as compared to the wild-type strain *exrA*<sup>+</sup> ( $\lambda$ ). This delay is not a consequence merely of UV sensitivity, since *uvr* strains show no such repression of prophage induction (Donch *et al.* 1970). Delayed induction is peculiar to *exrA* mutants, and, therefore, may be related to a deficiency in post-replicative repair. *recA* ( $\lambda$ ) strains, also defective in post-replicative repair (Howard-Flanders, Rupp, Wilkins & Cole, 1968), are not inducible by UV even after long delays

(Brooks & Clark, 1967). It is possible that *recA*<sup>+</sup> and *exrA*<sup>+</sup> are both involved at different points in a common repair system, since both are needed for complete expression of UV induced mutations and prophage induction.

*exrA* represses only UV induction of prophage. Recent experiments in this laboratory have shown that *exrA* ( $\lambda$ ) are induced as efficiently as *exrA*<sup>+</sup> ( $\lambda$ ) in the presence of nalidixic acid, *N*-methyl-*N'*-nitro-*N*-nitrosoguanidine and mitomycin C.

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