UV-PROTECTING PLASMIDS INCREASE POST-UV DNA SYNTHESIS IN *ESCHERICHIA COLI*

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Models to explain mutagenic DNA repair invoke the transient appearance of an error-prone DNA polymerase activity, which inserts random nucleotides opposite non-coding lesions in the DNA template. Many plasmids carry *muc* (mutagenesis caused by UV and chemicals) genes (Pinney 1980; Upton and Pinney 1981b), and we now report the effect of Muc+ plasmids on DNA synthesis in *Escherichia coli* after ultra-violet (UV) irradiation.

Cultures were grown overnight in fully supplemented Davis and Mingioli's medium (DM) (Davis and Mingioli 1950) at 37°. 0.5ml was then added to 9.5ml of fresh prewarmed DM and incubated in an orbital incubator at 37° for 1 h . The 10ml exponential cultures were then stirred and UV irradiated in 9cm glass Petri dishes. 0.25ml samples of the irradiated suspensions and of control, unirradiated cultures were added to 0.25ml volumes of fully supplemented DM containing 400µg ml^{-1} deoxyadenosine and 5µCi methyl-^{3}H thymidine (50 Ci $mmol^{-1}$). Assay mixtures were incubated at 37° for up to 6 h and 50µl samples taken at regular intervals. These were treated as described by Upton and Pinney (1981a) and DNA synthesis was determined as the incorporation of radioactivity into acid-insoluble material.

Resumpton of DNA synthesis in the DNA repair-proficient *E.coli* strain AB1157 was delayed for 40 and 60 min after UV doses of 90 and 135 Jm^{-2}, respectively. The UV-protecting plasmid R46 did not affect the time at which DNA synthesis was resumed in AB1157, although the amount of radioactivity incorporated was slightly but consistently higher than that incorporated by AB1157 lacking the plasmid after either dose of UV irradiation. R46 increases post-UV survival in strain AB1157 tenfold, whereas its protection factor in the UV-sensitive *E.coli* strain TK501 *uvrB umuC* is almost 10^4-fold (Upton, 1982). Experiments were therefore repeated using strain TK501 to test whether the large phenotypic effect on its survival produced by R46 could also be detected in its DNA synthesis after UV irradiation. A UV dose of 20 Jm^{-2} was found to delay DNA synthesis in strain TK501 for 75 min. This delay was reduced to less than 50 min by the presence of R46. The plasmid also significantly increased the rate of DNA synthesis. After 6 h post-UV incubation approximately five times as much label was incorporated by strain TK501 (R46) compared with the same strain lacking the plasmid (Table 1). Similar results were obtained with two other UV-protecting plasmids, R124 and R446b, whereas plasmids R1, RP4 and R6K, which do not affect post-UV survival (Pinney 1980), had no effect on post-UV DNA replication (Table 1).

TABLE 1. Effect of plasmids on post-UV DNA synthesis in *E.coli* strain TK501.

Plasmid	none	R1	RP4	R6K	R46	R124	R446b
Radioactivity (cpm) in culture sample at 6 hr	750	650	765	450	3,770	2,725	4,810

It has been suggested (Trgovcevic et al 1980) that DNA polymerisation after UV irradiation results from DNA repair, rather than from DNA replication. It is thus possible that Muc+ plasmids enhance post-UV survival by increasing DNA repair synthesis.

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