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Effect of Host *lex, recA, recF,* and *uvrD* Genotypes on the Ultraviolet Light-Protecting and Related Properties of Plasmid R46 in *Escherichia coli*

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The ability of plasmid R46 to reduce the lethal but enhance the mutagenic effect of ultraviolet (UV) irradiation was tested in sets of *Escherichia coli* K-12 derivatives, wild type or with different mutations affecting DNA repair capacity, but otherwise isogenic. UV protection and enhancement of UV mutagenic effect were obtained in *uvrA6, uvrB5, uvrD3,* and *recF143* hosts, but not in a *recA56* strain. The plasmid gave some UV protection in two *lexA1* and two *lexA101* strains and in one *lexA102* host, but produced no such effect in another *lexA102* host. The plasmid restored UV mutagenic effect in a *lexB30* strain, the yield of induced mutants per survivor of irradiation (10 J/m^2) being about the same for the *lexB30*(R46) and *lex*$^+$(R46) strains; by contrast the plasmid, though it reduced the UV sensitivity of the *lexB30* strain, did not make it as UV-resistant as the *lex*$^+$ R$^-$ strain.

The presence of some plasmids (colicin factors or R factors) reduces the lethal effect of ultraviolet (UV) irradiation of *Salmonella typhimurium* or *Escherichia coli* but enhances the mutagenic effect of such irradiation (3, 8, 9, 16, 22, 24). R factor R46 (previously called R-Brighton) also increases the frequency of some classes of spontaneous host mutation (16). UV protection and enhancement of UV mutagenesis by R46 were observed in *uvrB* and *polA* mutant, as well as in wild-type, hosts, but not in a *recA* mutant of *S. typhimurium* (16). It was surmised that the plasmids produced their effects, or at least those of them involving UV irradiation, by enhancing the ability of their host to carry out some process of repair of UV-damaged DNA in which mutations were generated ("error-prone" repair) (25). UV irradiation of *E. coli* mutants of type *recA, lexA,* and *lexB* has no, or almost no, mutagenic effect, it is believed because the error-prone DNA repair process depends on normal function of the genes affected in such mutants (18, 25). If this is so, and if UV-protecting plasmids act by enhancing error-prone repair, it would be expected that they would be ineffective not only in *recA* but also in *lexA* and *lexB* mutant hosts. As noted above, R46 failed to confer UV protection or restore UV mutagenesis in *recA* mutants of *S. typhimurium,* and a similar dependence on host *recA* function has been observed for plasmid pKM101, which is a derivative of R46 (K. E. Mortelmans and B. A. D. Stocker, Mol. Gen. Genet., in press), and for the R46-related plasmid R205 (R-Utrecht), in *S. typhimurium* or *E. coli* (13, 14, 16, 24). The dependence or nondependence of UV-protecting plasmids on host *lexA* function is less clear. Walker (24) found that plasmid pKM101 housed in a *lexA3* subline of *E. coli* K-12 did not give UV protection, increase the frequency of spontaneous mutation, or restore UV mutagenic effect, but Monti-Bragadin and his colleagues (14) reported that the same plasmid gave UV protection and enhanced UV mutagenic effect even in a *lexA* mutant derivative of *E. coli* B. To further investigate the dependence of the UV-protecting, etc., properties of plasmid R46 on host genes affecting UV sensitivity, we have tested the behavior of this plasmid in *lexA, lexB, uvrD, recA,* and *recF* derivatives of *E. coli* K-12, as compared to its behavior in nearly isogenic *lex*$^+$, *uvrD*$^+$, and *rec*$^+$ control strains. In our experiments R46 exerted its typical effects in some *lexA* mutant strains but not in others, which perhaps provides an explanation for the apparent discrepancy noted above. In agreement with reports on plasmid pKM101 the UV-protecting and related effects of R46 were not prevented by *uvrD* or *recF* mutation. In a *lexB30* strain, R46 conferred UV protection and increased the frequency of spontaneous mutation; furthermore, the mutagenic effect of UV irradiation, absent or scarcely detectable in the *lexB30* parent strain, was brought

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to the same level as in the R46-bearing derivative of the *lex*$^+$ control strain; i.e., R46 "cured" the UV mutagenesis defect of *lexB30*.

MATERIALS AND METHODS

Bacterial strains and plasmids. The sets of nearly isogenic strains used, all derived from *E. coli* K-12, are listed in Table 1. One R46-bearing derivative of each strain listed was preserved and used in all the experiments described. Each such derivative was allotted a strain number; but in this paper we shall, for clarity, use not the strain number of the plasmid-bearing derivative but its "formula." For instance, we will use AB1157(R46), rather than the strain number, SL4593, to denote the R$^+$ derivative of AB1157 (and the symbol R$^-$ is sometimes appended to a strain number, e.g., AB1157 R$^-$, to emphasize the absence of the R factor).

Media. The nutrient and defined media were as previously used (16). Revertants of amino acid auxotrophs were selected on defined medium supplemented with nutrient broth no. 2 (CM67; Oxoid Ltd.), 1% by volume, which we term enriched defined medium; Lac$^+$ revertants were selected on enriched defined medium with lactose as energy source, or, for semiquantitative experiments, on MacConkey base agar (Difco Laboratories) with lactose, filter-sterilized (5 g/liter).

Genetic methods. Plasmid R46 was transferred by growing donor and recipient together in broth, then selecting for recipient made ampicillin resistant (16). Purified transconjugants were tested for the four antibiotic resistances of R46 by the Kirby-Bauer method; nearly all those tested had acquired all four traits. Rates of spontaneous reversion of auxotrophs (to Leu$^+$, in the case of *leuB1* strains, to His$^+$ for *his-4* strains, or to His$^+$ Arg$^+$, by ochre suppressor mutation in *his-4 argE3* strains) were estimated essentially as described by Mortelmans and Stocker (16) by spreading 0.1-ml samples of washed cell suspensions from overnight broth cultures on plates of selective enriched defined medium, then incubating at 37°C until the number of revertant colonies no longer increased. Similarly inoculated plates of nonenriched selective medium were tested for the presence of preexisting revertants in the inoculum. Under these conditions for a nonleaky auxotroph, the final number of revertant colonies per plate is directly proportional to the probability of mutation per bacterium per generation (16). The frequency of reversion to Lac$^+$ was estimated similarly, and also semiquantitatively by counting red papillae on the thin film of lactose-negative growth on MacConkey lactose plates which had been similarly inoculated and incubated for 1 or 2 days. To estimate rates of UV-induced reversion, similarly inoculated plates were irradiated (see below), wrapped in aluminum foil to prevent photo-reactivation, and incubated. Revertant colonies were counted on day 2 or 3, and daily thereafter until they no longer increased in number. The mean number of revertant colonies per irradiated plate minus the mean number per nonirradiated plate gave the number of induced mutations per plate, from which the number of induced mutations per 10^8 survivors was calculated by taking into account the number of viable (nonirradiated) bacteria in the plate inoculum and the fraction surviving irradiation. UV dose/log survival curves were based on counts of col-

TABLE 1. *Strains used*

Strain no.[a]	Genotype[b]	Reference or source
Set 1		
DY178	*uvrB5 rha lac rpsL leuB1 thyA thyR*	6, 27
DY180	As DY178 but *lexA101*	27
DY179	As DY178 but *uvrD3*	27
DY155	As DY178 but *recA56 thyA*$^+$ *metE*	27
Set 2		
AB1157	*thr-1 leu-6 proA2 his-4 thi-1 argE3 lacY1 galK2 ara-14 xyl-5 mtl-1 tsx-33 strA31 sup-37*	15
JC9239	As AB1157 but *recF143*	19
JC10521	As AB1157 but *lexB30 thy*	A. J. Clark[c]
Set 3		
DY98	*lacZ-Y14 rpsL metE thyA thyR*	26
DY99	As DY98 but *lexA101*	26
SR205	As DY98 but *lexA1*	D. A. Youngs[c]
Set 4		
DM845	*uvrA6 lac thi-1 xyl gal tsx*	17
DM842	As DM845 but *lexA1*	17
Set 5		
DY239	*leuB1 bio rha lacZ-Y14 rpsL metE thyA thyR*	D. A. Youngs[c]
DY238	As DY239 but *lexA102*	D. A. Youngs[c]
Set 6		
PAM5811	*malB his-4 thr rpsL*	John Donch[c]
PAM5903	As PAM5811 but *lexA102*	John Donch[c]

[a] Strain numbers are as in cited publication, or as used by worker from whom obtained.
[b] All strains are *E. coli* K-12, known or presumed to be nonlysogenic for λ and F$^-$.
[c] Personal communication.

onies on sectors of nutrient agar plates inoculated with drops (0.01 ml each) of decimal dilutions of washed-cell suspensions (as used for selection of revertants), incubated without irradiation or after exposure to 256 nm of irradiation from a germicidal lamp (G8T5, General Electric Co.).

RESULTS

Effect of plasmid R46 on growth of some *E. coli* K-12 lines on defined medium. The R46-bearing derivatives of *E. coli* strain AB1157 and its nearly isogenic relatives (set 2 in Table 1), and also those of several other sets of *E. coli* K-12 derivatives, grew much more slowly on defined medium than did their R^- parents, even when no antibiotic was present in the medium; but on nutrient agar there was no obvious difference in rate of growth of R^+ and R^- strains. Because of their slower growth on defined medium, colonies of revertant mutants, spontaneous or UV induced, of plasmid-bearing strains took longer to appear than did those of the related R^- strains. However, we think that our procedure of continuing incubation until the number of revertant colonies per plate no longer increased insured that this slow growth of R46-bearing strains did not invalidate our estimates of their rates of spontaneous mutation. (The slow growth on defined medium caused by R46 in these *E. coli* strains appears to result from the action of a gene or genes of R46 which can be lost by deletion mutation [Waleh et al., unpublished data].)

Behavior of R46 in *lexA101, uvrD3,* and *recA56* forms of a *uvrB5* strain. We tested the effect of R46 in several of a set of nearly isogenic K-12 derivatives described by Youngs and Smith (27), all *uvrB5* and therefore unable to effect excision repair of pyrimidine dimers (Table 1, set 1). The mutation *leuB1* in these strains, probably a missense mutation (5, 6), was nonleaky and subject to only very low frequency of spontaneous reversion, therefore suitable for experiments on UV mutagenesis. The *lexA101* and *uvrD3* strains were, as expected, moderately UV sensitive (Fig. 1). R46 gave some UV protection in the control strain, although protection was, as expected, less than is observed in *S. typhimurium* strain LT2 (16); the plasmid gave rather more protection in the *lexA101* strain than in the control strain, and rather less in the *uvrD3* strain, but it did not reduce the extreme UV sensitivity of the *recA56* strain. The presence of R46 increased the frequency of induced Leu^+ revertants per 10^8 survivors of exposure to UV (0.5 J/m^2), from 74 to 264 in the control strain, but only from 15 to 26 in the *uvrD3* strain (Table 2). In the *lexA101*(R46) strain, the same irradiation had a significant mutagenic effect, 17

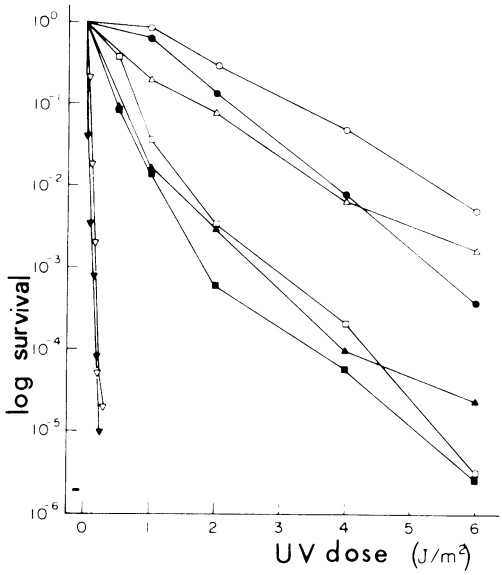

FIG. 1. *Log survival/UV fluence curves for R^- (filled symbols) and R46-bearing (open symbols) forms of strains of first isogenic set (all uvrB5). Symbols: ●, ○, DY178 (lex^+ rec^+ $uvrD^+$); ▲, △, DY180 (lexA101); ■, □, DY179 (uvrD3); ▼, ▽, DY155 (recA56).*

induced mutations per 10^8 survivors, compared with less than one (i.e., no significant mutagenic effect) in the *lexA101* R^- strain. No UV mutagenic effect was detected in the *recA56* R^- or the *recA56*(R46) strain. Because of low spontaneous reversion frequencies, these experiments did not test whether R46 has a mutator effect in these strains.

Behavior of R46 in *recF143* and *lexB30* forms of a *uvr*$^+$ host strain. The effect of plasmid R46 was similarly tested in a uvr^+ K-12 derivative, AB1157, and in its nearly isogenic related strains (Table 1, set 2). The *recF143* and *lexB30* strains were, as expected, moderately UV sensitive, and their survival curves lacked the conspicuous shoulder seen in the control strain (Fig. 2). The plasmid gave some protection in all three hosts, but somewhat less in the *recF143* strain than in the other two; the presence or absence of a shoulder in the curves was not altered by acquisition of the plasmid.

The *his* and *arg* mutations of AB1157 are both ochre (4, 10). His^+ Arg^+ mutants, i.e., ochre suppressor mutants, were therefore selected for measurements of mutation frequency (Table 2). The UV dose used (10 J/m^2) evoked 553 revertants per 10^8 survivors from the R^- control strain, AB1157, and 1,364/10^8 from its R^+ derivative. In the *recF143* R^- strain, the yield of induced revertants was about one-fifth that from the control R^- strain; the presence of the plasmid

TABLE 2. *Effect of R46 on frequency of UV-induced and spontaneous reversion mutation in sets of strains isogenic except for rec, lex, or uvrD character*

Strain			Nonirradiated, revertant colonies/plate[b]		UV irradiated						
No.	Relevant genotype[a]	Revertant class selected	R⁻	R⁺	Dose (J/m²)	% Survival		Revertant colonies/plate[b]		Induction mutations/10⁸ survivors[c]	
						R⁻	R⁺	R⁻	R⁺	R⁻	R⁺
Set 1[d]											
DY178	*uvrB5*	Leu⁺	1	2	0.5	75	90	81	238	74	264
DY180	*uvrB5 lexA101*	Leu⁺	0	0	0.5	33	43	0.25	17	(0.8)	17
DY179	*uvrB5 uvrD3*	Leu⁺	0.2	1	0.5	9	37	15	82	15	26
Set 2[e]											
AB1157	*uvr*⁺	His⁺ Arg⁺	2.6	4.2	10	100	100	437	1,500	535	1,364
JC9239	*uvr*⁺ *recF143*	His⁺ Arg⁺	6.6	25	10	20	28	17	263	97	588
JC10521[f]	*uvr*⁺ *lexB30*	His⁺ Arg⁺	0.3	11	10	8	14	1.7	339	(3.8)	1,312
Set 3[g]											
DY98	*uvr*⁺	Lac⁺	1	6	5	95	100	43	170	49	167
DY99	*uvr*⁺ *lexA101*	Lac⁺	2.7	6	5	15	54	5.7	20	(27)	45
SR205	*uvr*⁺ *lexA1*	Lac⁺	4.7	3.7	5	8	16	2	4	—	—
Set 5[h]											
DY239	*uvr*⁺	Leu⁺	0.3	1.6	5	70	80	15	37	177	488
DY238	*uvr*⁺ *lexA102*	Leu⁺	0	0.3	5	5.1	7.5	0	1.3	—	—
DY239	*uvr*⁺	Lac⁺	24	22	5	100	100	92	371	486	4,190
DY238	*uvr*⁺ *lexA102*	Lac⁺	5	6	5	8	8	3.5	4	—	—
Set 6[i]											
PAM5811	*uvr*⁺	His⁺	0.7	6	10	90	100	125	~1,000	1,200	~10,000
PAM5903	*uvr*⁺ *lexA102*	His⁺	0.6	0.3	2	6	9	0	1	—	—

[a] Genotype in respect of genes affecting UV sensitivity; loci not shown indicate wild type.

[b] Values indicate mean number of revertant colonies on plates (usually 3) of selective medium.

[c] —, No UV-induced mutations detected. Numbers in parentheses indicate estimates based on very small numbers of colonies on irradiated plates, or on small differences between mean numbers on irradiated and nonirradiated plates, therefore of low precision.

[d] Isogenic *uvrB5*, either *uvrD*⁺ *rec*⁺ *lex*⁺ or *lexA101* or *uvrD3*. Data not shown for DY155 (*recA56* strain in set 1) because of very low survival of irradiated bacteria.

[e] Isogenic *uvr*⁺, either *rec*⁺ *lex*⁺ or *recF143* or *lexB30*.

[f] See text for results of additional experiments on the *lexB30* R⁻ and *lexB30* (R46) strains.

[g] Isogenic *uvr*⁺ *lex*⁺ or *lexA101* or *lexA1*.

[h] Isogenic *uvr*⁺, either *lex*⁺ or *lexA102*.

[i] Isogenic *uvr*⁺, either *lex*⁺ or *lexA102*.

caused about a fivefold increase in the mutagenic effect of irradiation. The UV fluence used in this experiment (10 J/m²) allowed only 8% survival of the *lexB30* R⁻ strain; mutagenic effect of this exposure was hardly detectable (calculated number of induced revertants per 10^8 survivors was 3.8, i.e., <1% of the number evoked from the control, *lex*⁺ R⁻ strain, AB1157). The survival of the similarly exposed *lexB30*(R46) strain was 14%, and the yield of induced mutants per 10^8 survivors was 1,312, i.e., about the same as that for the R⁺ form of the *lex*⁺ control strain. The ability of R46 to restore a normal level of mutagenic response in the *lexB30* strain was confirmed in an additional experiment, in which selection was made for Arg⁺, His⁺, and Arg⁺ His⁺ revertants of the *lexB30* R⁻ and *lexB30*(R46) strains, unirradiated or after exposure to UV (10 J/m²). The survival of the R⁻ strain was 2.9%, and no mutagenic effect of irradiation was detected. For the *lexB30*(R46) strain, survival was 15%, and the yields of induced revertants per 10^8 survivors were 2.9×10^4 for Arg⁺ His⁺, 3.2×10^4 for Arg⁺ and 2.3×10^4 for His⁺ selection (which indicates that nearly all UV-induced His⁺ or Arg⁺ revertants of this strain arise by ochre suppressor mutation). UV dose/log survival curves for the two strains, determined in this experiment, were virtually identical to those shown in Fig. 2. This indicates that restoration of UV mutability by R46 did not result from reversion to *lex*⁺. The presence of the plasmid caused moderate increases in frequency of spontaneous ochre suppressor mutation in both the control strain and its *recF143* and *lexB30* relatives. (In the second experiment on the R⁻ and R46-bearing forms of the *lexB30* strain, selection was made for spontaneous reversion to His⁺ or Arg⁺, as well as for Arg⁺ His⁺, i.e., ochre suppressor, mutants. For each strain about half the mutants selected as His⁺, or as Arg⁺, retained their other requirement, and so

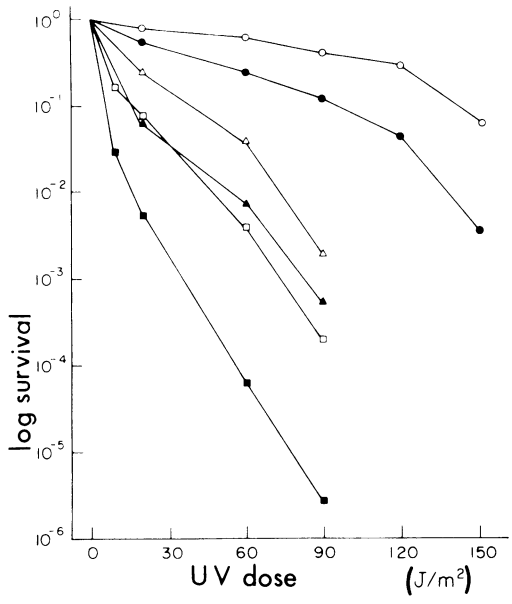

FIG. 2. *Log survival/UV fluence curves for R⁻ (filled symbols) and R46-bearing (open symbols) forms of strains of second isogenic set (all uvr⁺). Symbols: ●, ○, AB1157 (rec⁺ lex⁺); ▲, △, JC9239 (recF143); ■, □, JC10521 (lexB30).*

presumably arose by mutation at *his*, or *arg*; the yields [data not shown] indicated that carriage of R46 by the *lexB30* host raised the spontaneous mutation frequency for all three sorts of reversion, i.e., ochre suppressor, to His^+ and to Arg^+.)

Behavior of R46 in strains with different *lexA* mutations or with the same *lexA* mutation in different backgrounds. R46 was introduced into further sets of nearly isogenic strains, each set made up of one or more *lexA*-mutant strains and a lex^+ control (Table 1, sets 3–6). Set 3, all uvr^+, comprised a strain which carried *lexA101* (an allele already tested in a *uvrB* background [Table 2, set 1]), and its *lexA1* and lex^+ relatives. Both *lexA*-mutant strains were UV sensitive, with survival curves which were concave upwards, instead of with the conspicuous shoulder, seen in the lex^+ control (Fig. 3). R46 reduced UV killing in all three strains without altering the general shape of the survival curves. Exposure to UV (5 J/m²) evoked 49 Lac^+ revertants per 10^8 survivors from the R^- form of the lex^+ strain, and 167 from its R46-bearing form (Table 2). The same exposure had no detectable mutagenic effect on the *lexA1* strain, either R^- or R46 bearing. However, it evoked 45 Lac^+ revertants per 10^8 survivors from the *lexA101*(R46) strain, though it had no, or only a weak, mutagenic effect on the *lexA101* R^- strain. The presence of the plasmid in the lex^+ host increased the frequency of spontaneous reversion to Lac^+, but had no, or at most a slight, mutator effect in the two *lexA* strains.

In a further set (Table 1, set 4), lex^+ and *lexA1*, both *uvrA6*, R46 gave protection, rather less in the *lexA1* strain (and only at fluences allowing <1% survival) than in the lex^+. Each of a further two pairs (Table 1, sets 5 and 6) comprised a *lexA102* strain and a nearly isogenic lex^+ control strain, all uvr^+. In the *lexA102* strain of set 5, DY238, the plasmid conferred UV protection at fluences allowing survivals <2% but in the *lexA102* member of the sixth set, PAM5903, the plasmid gave no protection, though it did protect the related lex^+ strain, PAM5811. Reversion to Leu^+ and Lac^+ was tested in the strains of the fifth set, DY238 and DY239, which are *leuB1* and *lacZ-Y14*. In the lex^+ member, DY239, R46 enhanced the mutagenic effect of irradiation, in respect of both characters, but no, or almost no, UV-induced reversion of either character was detected in the *lexA102* strain, DY238, in either its R^- or R46-bearing form (Table 2). The plasmid also caused a significant increase in frequency of spontaneous reversion to Leu^+ in the lex^+ member but not in the *lexA102* member of set five. In the

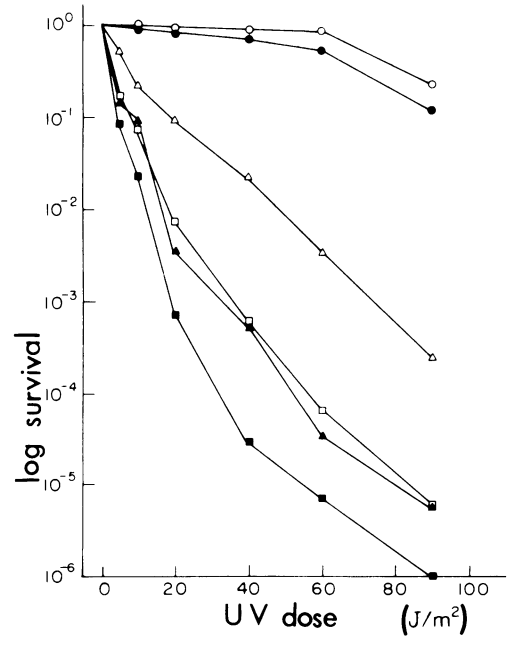

FIG. 3. *Log survival/UV fluence curves for R⁻ (filled symbols) and R46-bearing (open symbols) forms of strains of third set (all uvr⁺). Symbols: ●, ○, DY98 (lex⁺); ▲, △, DY99 (lexA101); ■, □, SR205 (lexA1).*

sixth set, which carry the ochre *his-4* allele, R46 increased the frequency of UV-induced His^+ revertants of the lex^+ strain about eightfold, and also increased its frequency of spontaneous reversion; no mutagenic effect of irradiation was detected in the *lexA102* member of the pair, either in its R^- or R46-bearing form, and the plasmid did not show the mutator effect seen in the lex^+ control strain.

DISCUSSION

Table 3 summarizes our observations on the effect of R46 on the lethal and the mutagenic actions of UV irradiation of *lexA, lexB, recA, recF,* and *uvrD* mutant strains, compared with its effects in nearly isogenic control strains, wild type for the character in question. To obtain a numerical estimate of UV-protecting ability, we measured, at three equally spaced points on each dose/survival curve, the log units killing of R^- parent strain and of its R46-bearing derivative, and took the average of the three values of (log units killing of R^+/log units killing of R^-). If there is no protection, this parameter has value one, whereas if there is protection it has a value less than one; the greater the protection, the lower its value. If both strains show exponential killing, the parameter is equal to slope for R^+/slope for R^-. In the six "control" strains (four uvr^+, one *uvrB5,* and one *uvrA6*) the plasmid gave UV protection (killing R^+/killing R^-, 0.39 to 0.69) and, in the five strains tested, enhanced UV mutagenesis (yield of induced revertants per 10^8 survivors for R^+/yield per survivor for R^-, ca. 2.5 to ca. 8) (Table 3). The plasmid also increased spontaneous mutation frequency in at least some of the "control" strains (Table 2). Thus, the presence of R46 in our control strains, either of wild-type UV sensitivity or of deficiency only in excision repair, had the three results—UV protection, enhancement of UV mutagenesis, and mutator effect—previously reported for this plasmid, its derivative pKM101, the related R-Utrecht (R205), and some other plasmids in the corresponding classes in *S. typhimurium* and/or *E. coli* (16, 22–24; K. E. Mortelmans, Ph.D. thesis, Stanford University, Stanford, Calif., 1975).

The failure of R46 to give UV protection or to restore the mutagenic effect of UV irradiation in the *recA56* host (strain DY155 in set 1, Fig. 1; Table 3) agrees with earlier observations on this and other UV-protecting plasmids tested in *recA*-type mutants of *S. typhimurium, E. coli,* or *Pseudomonas pyocyanea* (11, 13, 14, 16, 22–24). But note that Siccardi (20) found that two R factors reduced UV killing, in some circumstances, of two *rec* mutants of *E. coli* K-12 now designated as *recA*. In the *recF143* host, R46 conferred some UV protection, increased the weak mutagenic effect of UV irradiation about sixfold, and also increased spontaneous mutation frequency. Correspondingly, Walker (24) found that plasmid pKM101 exerted its UV-protecting ability and enhanced methyl methane sulfonate (MMS) mutagenesis when tested in another *recF143* host. It seems that these plasmids exert their effects without dependence on host *recF*

TABLE 3. *Summary of effect of R46 on UV sensitivity and UV mutagenesis in wild-type, uvr, lex, and rec hosts*

Isogenic set: genetic background; mutation selected	Bacterial strain		UV killing, (R^+/R^-)[a]	UV-induced mutations/10^8 survivors[b]		
	Strain	Mutation		R^-	R^+	R^+/R^-[c]
Set 1: *uvrB5*; Leu^+	DY178	—	0.59	74	246	3.6
	DY180	*lexA101*	0.47	—	17	+ (>10)
	DY179	*uvrD3*	0.78	15	26	1.7
Set 2: uvr^+; ochre suppressor	AB1157	—	0.43	535	1,364	2.5
	JC9239	*recF143*	0.67	97	588	6.1
	JC10521	*lexB30*	0.45	—	1,312	++ (>600)
Set 3: uvr^+; Lac^+	DY98	—	0.48	49	167	3.4
	DY99	*lexA101*	0.49	(27)	45	(1.6)
	SR205	*lexA1*	0.68	—	—	
Set 4: *uvrA6*; (none tested)	DM845	—	0.39	NA	NA	
	DM842	*lexA1*	0.68	NA	NA	
Set 5: uvr^+; Leu^+	DY239	—	0.69	171	488	2.9
	DY238	*lexA102*	0.79	—	—	
Set 6: uvr^+; His^+	PAM5811	—	0.68	125	ca. 1,000	ca. 8
	PAM5903	*lexA102*	1.0	—	—	

[a] Average value of log units killing R^+ strain/log units killing R^- strain for three equally spaced points on UV dose/log survival curve.

[b] —, No UV-induced mutations detected; NA, not applicable because UV-induced reversion was not tested.

[c] +, ++, No exact value could be calculated, because mutagenic effect of UV in R^- strain was undetectable or too weak for reliable estimation. Numbers in parentheses indicate inferred minimum values.

function, which is thought to be involved in a minor pathway for recombination and for repair of UV-damaged DNA (27).

In the *uvrD3 uvrB5* host, R46 gave some UV protection and caused a 1.6-fold increase in the small yield of UV-induced mutations (Fig. 2; Tables 2 and 3). Venturini and Monti-Bragadin (23) similarly found that pKM101 and another protecting plasmid enhanced both UV and MMS mutagenesis in a *uvrD3* derivative of *E. coli* strain B. At least a part of the UV-protecting and mutagenesis-enhancing effects of several plasmids thus does not require host *uvrD* function, supposedly needed for one branch of the post-replication repair process (21, 27).

We tested R46 in six *lexA*-mutant strains, two strains for each of three alleles (Tables 2 and 3). In five of the six strains the plasmid reduced UV killing (Fig. 1, 3, 4, 5), but in strain PAM5903 (one of the two *lexA102* strains tested) no protection was observed (Fig. 6). A (weak) UV mutagenic effect was seen in only one of the five *lexA*-mutant R^- strains tested, i.e., strain DY99, *lexA101*; in this strain R46 caused an increase (about 1.6-fold) in UV mutagenic effect (Tables 2 and 3). The introduction of R46 into the other four *lexA*-mutant strains tested resulted in the appearance of a weak mutagenic response to UV irradiation in strain DY190, one of the two

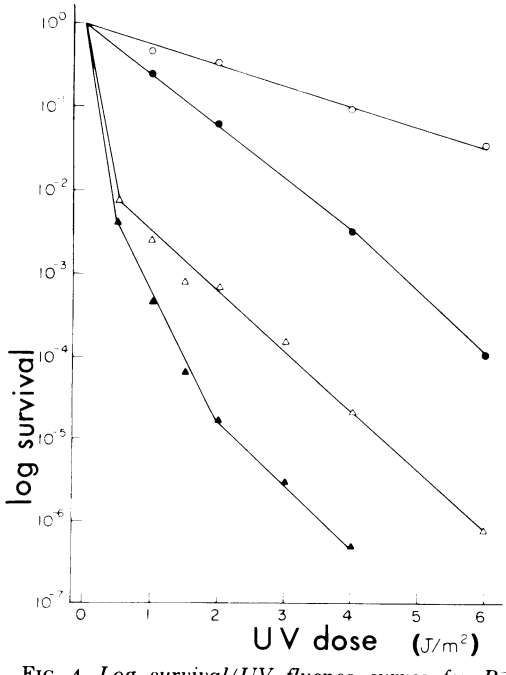

FIG. 4. *Log survival/UV fluence curves for R^- (filled symbols) and R46-bearing (open symbols) forms of strains of fourth set (both uvrA6). Symbols: ●, ○, DM845 (lex⁺); ▲, △, DM842 (lexA1).*

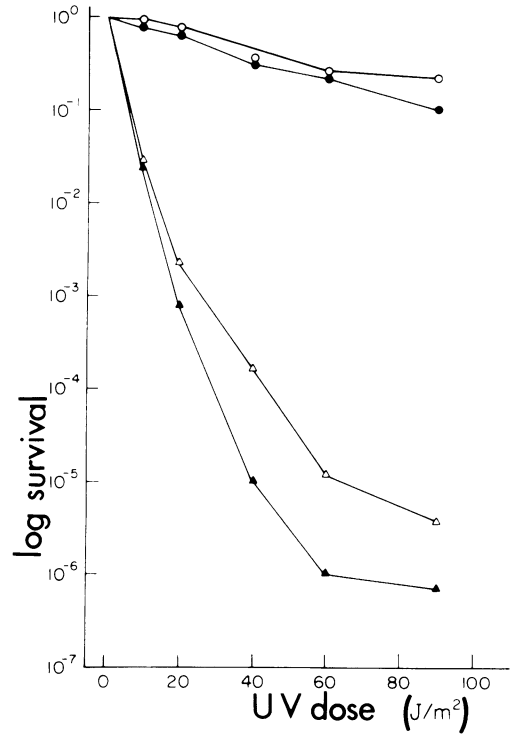

FIG. 5. *Log survival/UV fluence curves for R^- (filled symbols) and R46-bearing (open symbols) forms of strains of fifth set (both uvr⁺). Symbols: ●, ○, DY239 (lex⁺); ▲, △, DY238 (lexA102).*

lexA101 strains tested, but the plasmid had no such effect in the other three *lexA*-mutant strains. It appears from our results that the ability of R46 to confer UV protection and to restore, or enhance, mutagenic effect of UV in *lexA*-mutant hosts varies, both according to allele tested (compare DY99, *lexA101*, with the nearly isogenic SR205, *lexA1*, Tables 2 and 3) and according to genetic background (compare the two *lexA102* strains, DY238 and PAM5903 [Fig. 5 and 6; Tables 2 and 3]). It is therefore not surprising that, as noted in the introduction, earlier reports (1, 14, 24) differed as to the dependence of UV-protecting and mutagenesis-enhancing effect of plasmids on *lexA* function.

Mutants termed *lexB* resemble *recA* mutants in most phenotypic properties but are recombination-proficient and are thought to result from mutation of gene *recA*, which specifies protein X (2, 12). R46 conferred UV protection about as much in the *lexB30* host as in the *lex*⁺ control (Fig. 2; Table 3). UV irradiation had no, or at most a very weak, mutagenic effect on the R^- form of the *lexB30* strain, but in two experiments evoked as many revertants (ochre suppressor mutations) per 10^8 survivors from the

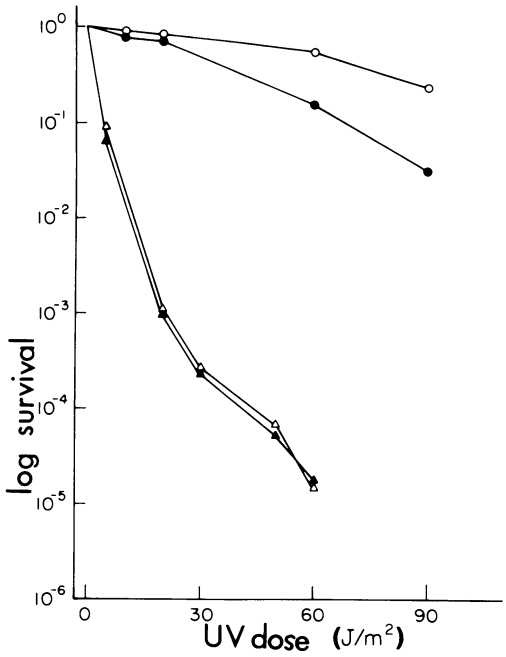

FIG. 6. *Log survival/UV fluence curves for R⁻ (filled symbols) and R46-bearing (open symbols) forms of strains of sixth set (both uvr⁺). Symbols: ●, ○, PAM5811 (lex⁺); ▲, △, PAM5903 (lexA102).*

lexB30(R46) strain as from the *lex*⁺(R46) control strain (Tables 2 and 3); the plasmid also increased the frequency of spontaneous mutation (ochre suppressor, also to His⁺ and to Arg⁺) in the *lexB30* host as well as in the *lex*⁺ control strain. Why R46 should restore mutagenic effect (though not wild-type UV resistance) in a host carrying *lexB30* (inferred from genetic evidence to be a missense *recA* allele [K. McEntee, personal communication]) but not in a host carrying *recA56* remains to be discovered. The possibility that R46 causes an increase in the amount of the (presumably abnormal) protein X synthesized in the irradiated *lexB30* cells has not yet been tested.

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