JOURNAL OF BACTERIOLOGY, Aug. 1977, p. 572-582

Vol. 131, No. 2
Printed in U.S.A.

Characterization of *lexB* Mutations in *Escherichia coli* K-12

PHILIPPE MORAND, MANUEL BLANCO,* AND RAYMOND DEVORET

*Laboratoire d'Enzymologie, Centre National de la Recherche Scientifique, 91190 Gif-sur-Yvette, France, and Instituto de Investigaciones Citológicas, Valencia-10, Spain**

Received for publication 3 January 1977

Two mutations have been located at the *recA* locus and phenotypically characterized along with a third one, previously called *rec-34*. The three mutants behaved similarly to *lexA* mutants. They were sensitive to ultraviolet (UV) light and X rays, and λFec⁻ phages were able to plate on them. The three mutations were called *lexB* because they could be distinguished from *recA* mutations by the last property. *lexB* mutants were less sensitive to UV and X irradiations than were *recA* mutants and were, to various degrees, recombination proficient. UV light failed to induce prophage λ in all three *lexB* lysogens. In contrast, thymine starvation induced *lexB31* and *lexB34* lysogens. In *lexB34* mutants, but not in *lexB30* and *lexB31* mutants, UV reactivation occurred at a low level. In *Escherichia coli* K-12, the *recA* gene has basic functions in the repair of deoxyribonucleic acid lesions, deoxyribonucleic acid recombination, and prophage induction. The three *lexB* mutations alter unequally and independently the three functions. This suggests that the *recA* and *lexB* mutations affect the same gene.

In *Escherichia coli* K-12, *recA* mutants have a pleiotropic phenotype: they are deficient in genetic recombination, are sensitive to ultraviolet (UV) and X irradiations, and have a high rate of deoxyribonucleic acid (DNA) breakdown, spontaneously as well as after irradiation. Furthermore, λFec⁻ phages (46) are not able to plate on RecA⁻ bacteria, and, when lysogenic for prophage λ, *recA* mutants exhibit extremely low spontaneous phage production and are not inducible (see review in reference 10). The phenotype of the *lexA* mutants differs from that of the *recA* mutants in that the former are proficient in genetic recombination, have a wild-type rate of spontaneous DNA breakdown, permit λFec⁻ phage to form plaques, and, when lysogenic for prophage λ, can have a wild-type spontaneous phage production. *lexA* are similar to *recA* mutants in that they are sensitive to UV and X irradiations, have a high rate of DNA breakdown after irradiation, and, when lysogenic for prophage λ, are not inducible (8, 26, 36). The *recA* and *lexA* mutations are located at 57 and 90 min, respectively, on the chromosomal map of *E. coli* K-12 (3).

A mutant isolated as deficient in lysogenic induction by thymine starvation (16) and having the phenotype of LexA⁻ bacteria (17) has been found carrying a mutation located in the *recA* region and designated *lexB30* (5). We have isolated another mutant resistant to thymine starvation. The mutation responsible for this phenotype has been mapped in the *recA* region and called *lexB31*. Van de Putte et al. (44) found that mutation *rec-34* caused a partial loss of ability to recombine. The *rec-34* mutation was mapped in the *recA* region (25). It is here considered as a *lexB* mutation since, as for *lexB30* (5), we found that *lexB31* and *rec-34* mutations do not affect the efficiency of plating of a λFec⁻ phage. We propose to rename it *lexB34* for the sake of standardization.

Comparative characterization of the three mutations *lexB30*, *lexB31*, and *lexB34* should lead to a better understanding of the role of the gene products of the *recA* region in different cellular processes such as repair, recombination, and lysogenic induction.

MATERIALS AND METHODS

Bacterial and phage strains. The bacterial strains are listed in Table 1. Nomenclature in general conforms to that of Demerec et al. (15).

Phage 434*imm*λ (30) was used instead of phage λ with various Mal⁻ bacteria resistant to phage λ. A Fec⁻ derivative of 434*imm*λ was isolated by its ability to grow on strain C600(P2) (A. Goze and R. Devoret, unpublished data). Spontaneous production and induction of prophage λ was determined as described in Moreau et al. (35). Phage P1*vir* is a virulent derivative of P1*kc* (27).

Media and chemicals. LB contained 10 g of NaCl, 10 g of tryptone (Difco), and 5 g of yeast extract per liter of demineralized water and was adjusted to pH 7 with NaOH. LA contained 15 g of Biomar agar per liter of LB. LA 10 contained 10 g of Biomar agar, 0.5 g of glucose, and 0.3 g of $CaCl_2$ per liter of LB. LBT, LAT, and LA 10T were each supplemented with 40

TABLE 1. *Bacterial strains*

Strain	Sex	Genotype	Origin and references
AB1157	F⁻	*thi-1 thr-1 leu-6 proA2 his-4 argE3 lacY1 galK2 ara-14 xyl-5 mtl-1 str-31 tsx-33 sup-37*	2
AT713	F⁻	*thi-1 lysA22 argA21 cysC43 malA1 xyl-7 mtl-2 lam str-104 rel-1*	Coli Genetics Stock Center, Yale, New Haven, Conn.
AT2427	Hfr PO1	*thi-1 cysC43 rel-1*	Coli Genetics Stock Center
C600	F⁻	*thi-1 thr-1 leu-6 lacY1 tonA21 supE44*	2
JC5547	F⁻	Same markers as AB1157, *recA13 recB21 recC22*	2
JM692	F⁻	Same markers as AB1157, *cysC43 ilvK633*	Castellazzi et al., in press.
KMBL49	F⁻	*thi-1 thr-1 leu-6 pyrF47 thyA6 dra-1 lacY1 tonA21 supE44*	43
KMBL336	F15 PO45	Same markers as KMB49, *str*/F- *thy*⁺	Medical Biological Laboratory, Rijswijk, The Netherlands
MH5	Hfr PO45	*thi-1? rel-1? nalA*	23
P4X6B	Hfr PO3	*metB1 rel-1*	S. Brenner via E. Signer; P4X6 cured of λ (2)
PC0150	F⁻	*thi-A his trp tyrA purC lacY1 gal-6 xyl-7 mtl-2 malA1 lam tonA2 tsx phx str*	42
3000	Hfr PO1	*thi-1 rel-1*	2
GY515	F⁻	*uvrA16 str-515 tol1468 supD*	4
GY550	F42 PO65	*thi-1 thr-1 leu-6 thyA6 dra-1 lacY1 dnaB87 tonA21 str supE44*/F- *lac*⁺	Strain no. 7 of Bonhoeffer (6) having received F42 from 200PS F⁻ *lac*⁺ (2)
GY3242	F⁻	*bio-87 endA101? metS120? his-108 ilvA106? cysC43 pheA97 str-8*	*thy*⁺ *cys*⁻ recombinant of GY3450 (Morand and Devoret, in press) mated with AT2427
GY3254	F⁻	*cysC43 lac str* (λ)	*thy*⁺ *cys*⁻ recombinant of MM 384 (34), mated with AT 2427, lysogenized with λ
GY3257	F⁻	Same markers as GY3242, (λ)	GY3242 lysogenized with λ
GY3491	F143 PO45	*thi-1 thr-1 leu-6 pyrF47 his-108 thyA6 argG103 ilvA106 cysB112 pheA97 lacY1 recA36 uvrB97 tonA21 tsx cod dra-1 phx supE44*/F- *thy*⁺ *rec*⁺ *phe*⁺	Morand and Devoret (in press)
GY4015	F⁻	Same markers as C600, *ampA601*	C600 resistant to 20 mg of ampicillin per liter (35)
GY5207	F⁻	Same markers as AB1157, *ilvK633, recA11*	*cys*⁺ *recA11* transductant of JM692 from donor GY3451
GY5208	F⁻	Same markers as AB1157, *ilvK633, lexB31*	*cys*⁺ *lexB31* transductant of JM692 from donor GY5206
GY1015	F⁻	Same markers as KMBL49, *lam mal* (λ)	KMBL49 lysogenized with λ and rendered λ^r
GY1163	F⁻	Same markers as KMBL49, *lexB30*	GY6030 cured of λ (16)
GY1164	F⁻	Same markers as KMBL49, *lexB30* (λ)	GY1163 lysogenized with λ
GY1165	F⁻	Same markers as KMBL49, *lexB30 tsx*	GY1163 rendered T6^r
GY1166	F⁻	Same markers as KMBL49, *lexB30 tsx str*	GY1165 selected for spontaneous streptomycin resistance
GY1167	F⁻	Same markers as KMBL49, *lexB30 lam mal* (λ)	GY1164 rendered λ^r
GY2549	F101 PO1	Same markers as KMBL49, *recA128*/F- *thr*⁺ *leu*⁺	8
GY3201	F42 PO65	Same markers as KMBL49, F- *lac*⁺	GY1163 having received F42 from GY550
GY3246	F⁻	*thi-1 thr-1 leu-6 pyrF47 dra-1 lacY1 tonA21 supE44 lexB30 tsx str recB21*	*thy*⁺ *recB21* transductant of GY1166 from donor AB2470
GY3247	F⁻	*thi-1 thr-1 leu-6 pyrF47 dra-1 lacY1 tonA21 supE44*	*thy*⁺ transductant of KMBL49 from donor AB2470
GY3248	F⁻	*thi-1 thr-1 leu-6 pyrF47 dra-1 lacY1 tonA21 supE44 lexB30 tsx str*	*thy*⁺ transductant of GY1166 from donor AB2470
GY3262	F⁻	*thi-1 thr-1 leu-6 pyrF47 dra-1 lacY1 tonA21 supE44 str*	*str*⁻ transductant of GY3247 from donor GY1166
GY3263	F⁻	Same markers as KMBL49, *str*	*str*⁻ transductant of KMBL49 from donor GY1166
GY3266	F⁻	*thi-1 thr-1 leu-6 pyrF47 dra-1 lacY1 tonA21 supE44 str recB21*	*thy*⁺ *recB21* transductant of GY3263 from donor AB2470
GY3422	F⁻	Same markers as AT713, *thyA61*	AT713 made *thy*⁻ by selection with trimethoprim
GY3424	F⁻	*thi-1 lysA22 thyA61 argA21 cysC43 malA1 xyl-7 mtl-2 lam spc-339 rel-1*	*spc*⁻ *str*⁺ transductant of GY3422 from donor GY2339

TABLE 1—*Continued*

Strain	Sex	Genotype	Origin and references
GY3428	F⁻	Same markers as GY3424, *lexB31*	This paper
GY3431	F⁻	Same markers as GY3424, *lexB31 pheA97*	This paper
GY3442	F⁻	Same markers as GY3424, *pheA97*	*lex*⁺ transductant of GY3431 from donor PC0150
GY3444	F⁻	Same markers as GY3424, *tyrA*	*phe*⁺ *tyrA lex*⁺ transductant of GY3431 from donor PC0150
GY3445	F⁻	Same markers as GY3424, *recA1*	*phe*⁺ *recA1* transductant of GY3442 from donor KL16-99
GY3446	F⁻	Same markers as GY3424, *recA13*	*phe*⁺ *recA13* transductant of GY3442 from donor AB2463
GY3447	F⁻	Same markers as GY3424, *recA36*	*phe*⁺ *recA36* transductant of GY3442 from donor GY3440
GY3448	F⁻	Same markers as GY3424, *lexB30*	*phe*⁺ *lexB30* transductant of GY3442 from donor GY1163
GY3449	F⁻	Same markers as GY3424, *lexB34*	*phe*⁺ *lexB34* transductant of GY3431 from donor KMBL239
GY5202	F⁻	Same markers as GY3424, *recA11*	*tyr*⁺ *recA11* transductant of GY3444 from GY3451
GY5209	F⁻	Same markers as GY3424, (434*imm*λ)	GY3424 lysogenized with 434*imm*λ
GY5210	F⁻	Same markers as GY3424, *lexB31* (434*imm*λ)	GY3428 lysogenized with 434*imm*λ
GY5218	F⁻	Same markers as GY3424, *recA128*	*phe*⁺ *recA128* transductant of GY3442 from donor GY6128
GY5219	F⁻	Same markers as GY3424, *lexB34* (434*imm*λ)	GY3449 lysogenized with 434*imm*λ
GY5222	F⁻	Same markers as GY3424, *lexB30* (434*imm*λ)	GY3448 lysogenized with 434*imm*λ

mg of thymine per liter. These media were used as rich liquid and solid media.

GT contained 5 g of NaCl, 5 g of tryptone (Difco), 8 g of peptone, and 12 g of Biomar agar per liter of demineralized water and was adjusted to pH 7 with NaOH. This medium was used for plating phage on indicator bacteria.

Soft agar contained 7.5 g of agar (Difco) per liter of demineralized water.

EMBO contained 5 g of NaCl, 12.5 g of EMB broth base (Difco), 1 g of yeast extract, and 13.5 g of Biomar agar per liter of demineralized water. EMB-mal, -lac, or -gal contained 10 g of maltose, lactose, or galactose, respectively, per liter of EMBO.

YM9 contained 11 g of $Na_2HPO_4 \cdot 7H_2O$, 3 g of KH_2PO_4, 1 g of NH_4Cl, and 5 g of NaCl per liter of double-distilled water. This medium served as a buffer or was used as a culture medium when supplemented with sugar and growth factors: 2 g of glucose, 40 mg of thymine, 40 mg of uracil, 9 mg of adenine, 1 mg of biotin, 1 mg of thiamin, 60 mg of arginine, 20 mg of cysteine, 20 mg of histidine, 30 mg of isoleucine, 60 mg of leucine, 30 mg of lysine, 30 mg of methionine, 30 mg of phenylalanine, 30 mg of proline, 75 mg of threonine, 10 mg of tryptophan, 20 mg of tyrosine, 40 mg of valine, and 10 g of Casamino Acids (vitamin-free; Difco) per liter. YM9-supplemented solid medium was the same as YM9-supplemented liquid medium except that it contained 4 g of glucose and 15 g of agar (Difco).

Chemicals were used at the following final concentrations, per liter: 10 mg of trimethoprim, 0.2 g of streptomycin, 0.1 g of spectinomycin, 10 mg of ampicillin, and 10^7 U of penicillin. Trimethoprim, spectinomycin, and ampicillin were donated by Wellcome Laboratories, The Upjohn Co., and Bristol Laboratories, respectively.

Unless otherwise stated, all cultures and plates were incubated at 37°C.

Isolation of thymine auxotrophs. Thymine-requiring mutants were isolated by a procedure derived from that of Stacey and Simson (41). Bacteria were grown in minimal medium with thymine to stationary phase; then the culture was diluted 10-fold, and portions were plated on plates containing minimal medium plus thymine and trimethoprim. After 2 days, many colonies were picked and purified.

Thymine starvation. The cultures were incubated in YM9 supplemented with glucose, Casamino Acids, and thymine until cells had reached a concentration of about 3×10^8 cells/ml; then the cultures were centrifuged, washed with YM9, left for 10 min at room temperature, and centrifuged again. The pellet was suspended in YM9 supplemented with glucose and Casamino Acids (method I) or in YM9 and then diluted 100-fold in supplemented YM9 (method II). Method I was used to isolate GY3428 after 210 min of starvation.

Genetic crosses. (i) Matings. Exponential cultures at about 3×10^8 cells/ml were mixed in the ratio of 1 Hfr or F′ donor to 10 F⁻ recipients and incubated for 30 min without shaking. Mating was interrupted by diluting a sample into YM9 with or without streptomycin and by vortexing for 2 min. The bacteria were then plated on appropriate selective media.

(ii) Transductions. Lysates of P1*vir* phage were prepared by the confluent-plate lysis technique. Transductions were done by a procedure derived from that of Arber (1). After dilution in 0.01 M $MgSO_4$, the lysate was exposed to UV light at a dose of 130 J/m² and mixed (about 1:1) with the recipient culture at 2×10^8 bacteria/ml. The mixture was incubated 30 min and then diluted 5- to 10-fold in prewarmed LBT and incubated for 30 min with shaking. It was then centrifuged and washed, and the pellet was suspended and plated on selective medium.

Recombination index. The recombinational abil-

ity of an F^- strain has been calculated with the following formula, which defines a recombination index (R) for the transmission of given markers: $R = (R_1/R_0)/(S_1/S_0)$. R_1 and R_0 are the numbers of recombinants obtained in crosses of an Hfr donor with the mutant F^- recipient tested and with the F^- parental strain respectively; S_1 and S_0 are the corresponding numbers of F′ transconjugants obtained in crosses of an F′ donor having the same point of origin of transfer as the Hfr donor.

This formula is derived from that of Castellazzi et al. (8).

Irradiation techniques. UV irradiation was performed with a 15-W General Electric germicidal UV lamp on bacteria diluted in 0.01 M $MgSO_4$. Doses were measured with a Latarjet dosimeter. Two techniques were used: bacteria in the exponential phase of growth in LBT medium were harvested by centrifugation and suspended in 0.01 M $MgSO_4$ or diluted at 10^{-2} or lower concentrations in 0.01 M $MgSO_4$. Survival was measured on LAT plates.

X irradiation was performed with a Machlett tube OEG60 (19) on bacteria in the exponential phase of growth in LBT medium. Before cells were plated on LA plates, bacteria either were or were not diluted in YM9 buffer. Under our irradiation conditions, the adsorbed dose was about 100 rads/s as measured according to Cottin and Lefort (12).

Colonies replica plated on LAT medium were submitted to UV light and X rays to check their radioresistance.

Reactivation of UV-irradiated phage λ. Bacteria in the exponential phage of growth in LBT medium were centrifuged and suspended in 0.01 M $MgSO_4$. One-half of the culture was irradiated, and the control and irradiated samples were both diluted with an equal volume of double-strength LBT. After 30 min of aeration at 37°C, the cells were infected with UV-irradiated phage λ at a multiplicity of infection of 1 phage for 10 or 100 bacteria or with nonirradiated phage at a multiplicity of infection of 0.01 or 0.001. After 20 min, the infected bacteria were mixed with a culture of GY515 indicator bacteria and plated on GT plates.

RESULTS

Mutations *lexB30* (16), *lexB31* (this article), and *lexB34* (44), are characterized here in comparison with *recA* mutations such as *recA1* (11), *recA11* (20), *recA13* (26), *recA36* (44), and *recA128* (16). Plaque formation of λFec^- phages on *lexB*$^-$ bacteria was our criterion for differentiating *lexB* from *recA* mutants: the efficiency of plating of 434*imm*λFec^- on *lexB* mutants was 70 to 100% of that found with the parental strain, whereas it was about 10^{-5} on the *recA* mutants.

***lexB* mutations are located in the *recA* region.** To map the *lexB30* mutation, we performed two crosses between GY1166 (F^- *leu-6 thyA6 lexB30 str*) and Hfr strains P4X6B and MH5, selecting for Thy^+ Str^r or Leu^+ Str^r recombinants. These crosses gave an approximate location of *lexB30* near *thyA* (Table 2). A

TABLE 2. *Map location of lexB30 with respect to thyA, cysC, and pheA*[a]

Donor	Recipient	Selected marker	Total no. of tested recombinants	Unselected marker	No. of recombinants
P4X6 (Hfr PO3 *leu*$^+$ *lex*$^+$ *thy*$^+$)	GY1166 (*leu lexB30 thy*)	*leu*$^+$	24	*lexB*$^-$	24
				lexB$^+$	0
		thy$^+$	48	*lexB*$^-$	24
				lexB$^+$	24
MH5 (Hfr PO45 *thy*$^+$ *lex*$^+$)	GY1166 (*thy lexB30*)	*thy*$^+$	80	*lexB*$^-$	58
				lexB$^+$	22
GY3201 (*phe*$^+$ *lexB30 cys*$^+$/F42)	GY3257 (*phe lex*$^+$ *cys*)	*phe*$^+$	364	*cys*$^-$ *lex*$^+$	21
				cys$^-$ *lex*$^-$	44
				cys$^+$ *lex*$^+$	17
				cys$^+$ *lex*$^-$	282
		cys$^+$	354	*phe*$^-$ *lex*$^+$	50
				phe$^-$ *lex*$^-$	114
				phe$^+$ *lex*$^+$	20
				phe$^+$ *lex*$^-$	170
P1/GY1166 (*lexB30 cys*$^+$)[b]	GY3254 (*lex*$^+$ *cys*)	*cys*$^+$	596	*lexB*$^+$	572 (4.0%)
				lexB$^-$	24
P1/GY116 (*phe*$^+$ *lexB30*)	GY3257 (*phe lex*$^+$)	*phe*$^+$	162	*lexB*$^+$	159
				lexB$^-$	3 (1.8%)

[a] The *lexB30* phenotype was checked by its sensitivity to UV light and subsequently by the absence of induction of prophage λ.

[b] P1/GY1166 is a transducing stock of P1 phage grown on strain GY1166.

cross between GY3257 (F⁻ *cysC43 pheA97*) and GY3201 (F' *lexB30*), transferring the chromosome at low frequency from PO65, indicated that *lexB30* probably lies between *cysC* and *pheA* as deduced from the distribution of the unselected markers. Finally, cotransduction of *lexB30* with *cys*⁺ or *phe*⁺ assigned *lexB30* to a location similar to that of *recA* mutations.

The *lexB31* mutant strain GY3428 was isolated in order to introduce a mutation in the *recA* locus in a strain carrying multiple genetic markers near the *recA* locus. Since *recA* mutants are resistant to thymineless death (16, 28) and relatively resistant to treatment with penicillin as shown by their survival being one order of magnitude higher than that of wild-type bacteria (data not shown), GY3424 (*rec*⁺) was submitted to two cycles of growth and thymine starvation and growth and treatment with penicillin, and to one more cycle of growth and thymine starvation. After this treatment, the surviving bacteria were plated on synthetic medium. Colonies were picked and tested by replica plating for their sensitivity of UV light and X rays. Out of the first 40 *thyA*⁻ surviving clones tested, one was radiosensitive. The mutation carried was mapped by introducing into the mutant strain GY3428 (*lysA22 thyA61 argA21 cysC43*) the F143 sex factor bearing the genes from *lysA* to *pheA*, including *recA*, and the F15 sex factor carrying only the genes from *lysA* to *argA* and not the *recA* locus (Fig. 1). F143 but not F15 complemented the mutation (data not shown). Since 434*imm*λ Fec⁻ was able to plate on GY3428, the mutation was then called *lexB31*. More precise location of *lexB31* in the *recA* region was obtained by transducing *cys*⁺ with phage P1 into GY3428 (*cysC43 lexB31*); the frequency of cotransduction *cys*⁺-*lex*⁺ was 15%, and there was no cotransduction between *lex*⁺ and *arg*⁺ (data not shown).

During the course of the mapping of *lexB34*, the frequencies of cotransduction of this mutation with *cysC*⁺ and *pheA*⁺ were found to be low: <0.2 and 0.6%, respectively (25). Similarly, using strain GY3442 (*cysC43 pheA97*) as recipient, no cotransduction of *lexB34* with *cysC*⁺ and *pheA*⁺ was obtained for 100 Cys⁺ and 360 Phe⁺ transductants tested. We have no explanation for this anomalous behavior. Nevertheless, with strain GY3431 (*cysC43 lexB31 pheA97*) as recipient, we were able to cotransduce *lexB34* with *cysC*⁺ and *pheA*⁺ with frequencies of 17 and 6%, respectively. In this experiment, *lexB34* transductants, in contrast to *lexB31*, were able to form replica patches after irradiation with a UV dose of 20 J/m². No Lex⁺ transductant was found in this transduction. This indicates that *lexB31* and *lexB34* are very close to each other.

Introduction of markers in the *recA* region by abortive transduction: construction of isogenic strains. To compare *lexB* with *recA* mutants, it was necessary to construct isogenic strains with various markers around the *recA* locus. In the first step, we developed a method to recover abortive P1 transductants and introduced *pheA97* into GY3428. In the second step, in newly marked strains, we introduced *recA* and *lexB* mutations by cotransduction with phage P1.

We made use of the fact that when the *rec*⁺ gene is transduced into *recA*⁻ recipients, the *rec*⁺ gene is expressed in both abortive and stable transductants (24). If *lexB31* were recessive, upon P1 transduction from a *lex*⁺ *pheA97* donor into a *lexB31 phe*⁺ recipient, it would then be possible to select transductants, some of which would be transiently resistant to UV light after infection with the transducing

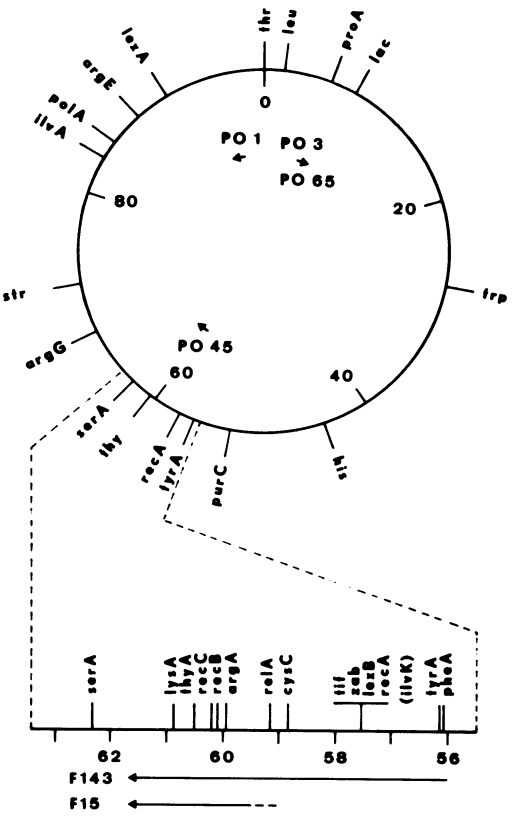

FIG. 1. *Expanded map of the E. coli K-12 chromosome region covered by F15 and F143 (3, 33). PO are points of origin of transfer for Hfr or F' strains relevant to this study (2).*

phage. Among the transductants that would have received the *lex*$^+$ *pheA97* chromosomal segment, after recombination and segregation, some would have inherited only the *pheA97* allele from the donor. Using GY3428 as recipient and GY3242 as donor, we selected transductants resistant to a UV dose of 10 J/m^2 2 h after infection. Of 1,134 resistant colonies picked, 85 were Lex$^+$ and one was Lex$^-$ Phe$^-$. Thus we obtained strain GY3431 (*cysC43 lexB31 pheA97*). The low proportion of Lex$^+$ colonies obtained was expected because of the relatively high residual survival of *lexB31* bacteria at this dose.

Strain GY3442 (*cysC43 lex*$^+$ *pheA97*) was also constructed by selection of bacteria resistant to 10 J/m^2, using GY3431 as recipient and PCO150 (*lex*$^+$) as donor. The frequency of UV-resistant bacteria among the P1-infected bacteria surviving irradiation was about 7.5%; the number of UV-resistant bacteria expected by reversion was at most 20 times lower than this value.

Isogenic strains were constructed by transducing *phe*$^+$ into GY3442 along with one of following markers: *lexB30*, *recA1*, *recA13*, *recA36*, or *recA128*. An isogenic *lexB34* strain was derived from GY3431 (*lexB31 pheA97*), since we did not obtain any *phe*$^+$ *lexB34* cotransductant in GY3442. The isogenic *recA11* strain was obtained from GY3444 (*rec*$^+$ *tyrA*) using *tyr*$^+$ as a selective marker, the donor strain carrying *pheA97*.

lexB mutants are less sensitive to UV and X irradiations than are recA mutants. *lexB31* and *recA1* mutants were more radiosensitive to X rays and to low UV doses than were *lexB34* and *lexB30* mutants, which were themselves equally radiosensitive (Fig. 2). However, the UV survival curves of *lexB31* and *lexB34* mutants tailed off so that, at doses higher than 3 J/m^2, their survival was higher than that of *recA1* and *lexB30* mutants. Survival curves of other *recA*$^-$ isogenic strains were similar to that of *recA1* (these curves are deleted from the figure for the sake of clarity). To summarize, *lexB* mutants were less sensitive to UV light and, except for *lexB31*, to X rays than were *recA* mutants.

UV light fails to induce prophage λ normally in lexB mutants. The spontaneous phage production in a *lexB30* (λ) lysogen was extremely low, with almost no free λ phage production after UV irradiation (Table 3). In this lysogen, prophage λ was not inducible by UV light.

The spontaneous phage production during exponential growth of GY5210 (*lexB31*)

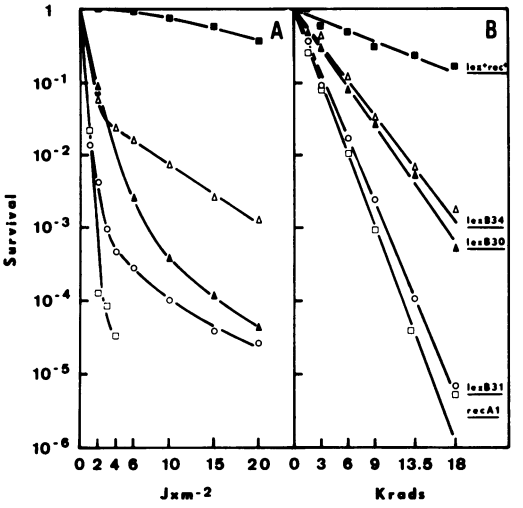

FIG. 2. *Bacterial survival after UV light (A) and X-ray (B) irradiation of GY3424 (lexB*$^+$ *recA*$^+$*) (■), GY3428 (lexB31) (○), GY3445 (recA1) (□), GY3448 (lexB30) (▲), and GY3449 (lexB34) (△).*

TABLE 3. *Phage released by UV-irradiated lexB30 (λ) bacteria*

Strain	UV dose (J/m^2)	PFU/ml[a]	Clear plaques (%)
GY1167 (*lexB30*)	0	2.5 × 10^3	72
	10	1.5 × 10^4	29
	30	5.5 × 10^3	2
	60	1.5 × 10^3	2
GY1015 (*lex*$^+$)	0	8.0 × 10^6	<1
	10	6.0 × 10^9	<1
	30	6.0 × 10^{10}	<1
	60	8.3 × 10^9	<1

[a] PFU, Plaque-forming units. Bacteria at a concentration of 2 × 10^8 cells/ml were centrifuged and suspended in YM9 buffer and then UV irradiated. The suspensions were diluted fivefold in LB and incubated with aeration at 37°C for 3.5 h. The cultures were then chloroformed, and the free phage was assayed on C600 indicator bacteria.

(434*imm*λ) was found to be one order of magnitude higher than that of the parental strain (Fig. 3). Prophage UV induction was determined by the number of infective centers plating on ampicillin-resistant indicator bacteria (Fig. 4); with this technique, delayed spontaneous production of phage was eliminated by the ampicillin in the plates (35). Two kinds of plaques were visible: large plaques, whose number decreased slightly with increasing doses, and small plaques, whose number increased rapidly at doses ranging from 0 to 2 J/m^2 and then decreased as did the large plaques.

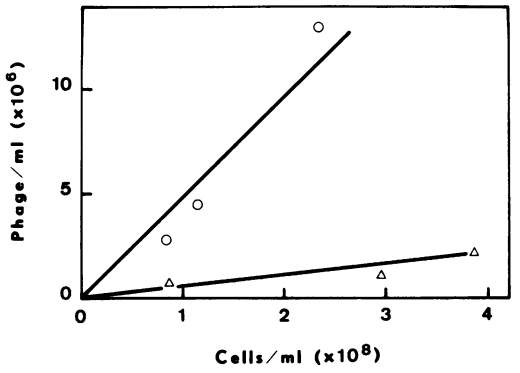

FIG. 3. *Spontaneous production of 434immλ phage during the exponential growth of GY5209 (lexB⁺) (△) and GY5210 (lexB31) (○). Culture samples were chloroformed at different times, and 0.1 ml of the supernatant was plated on C600 indicator bacteria.*

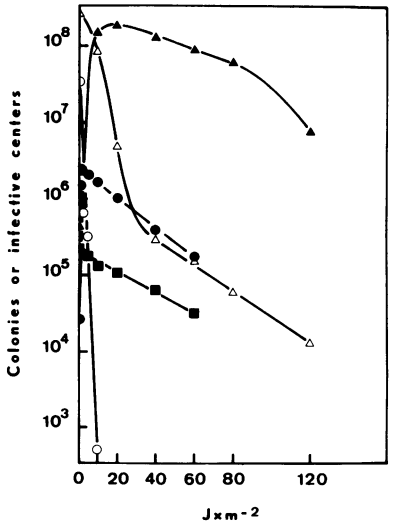

FIG. 4. *Colonies (open symbols) and infective centers (closed symbols) after UV irradiation of 434immλ lysogenic bacteria GY5209 (lexB⁺) (△) and GY5210 (lexB31) (○). In the case of lexB31, small-size infective centers are indicated by (●); the large plaques are indicated by (■). The indicator strain was GY4015, and the plates contained ampicillin.*

This led us to suppose that prophage induction was delayed, the formation of small plaques being due to late liberation of phage λ.

Van de Putte et al. (44) showed that the spontaneous phage production of *lexB34* lysogens equals that of wild-type bacteria; that the number of infective centers after UV irradiation increases first and then decreases as a function of the dose; and that the release of free phage from the infective centers does not occur before 3 h of incubation. Since the number of free phage after UV irradiation is low, Hoekstra et al. (25) concluded that prophage λ is not inducible in *lexB34* mutants.

Thymine starvation induces prophage λ in *lexB31* and *lexB34* mutants. The *lexB31* mutant, although very sensitive to UV light, is resistant to thymineless death. Therefore, if induction occurred in the *lexB31* mutant but was not detected after UV irradiation, one expects that thymine deprivation would allow better expression of lysogenic induction.

The survival of lysogens and nonlysogens, as well as the number of infective centers produced, is plotted in Fig. 5 as a function of the thymine deprivation period. We have not distinguished large and small plaques, since the proportion of small plaques was low. As expected, *lexB31* and *lexB34* lysogens were induced, but maximal induction was obtained about 60 min later than in wild-type lysogens. The efficiency of thymineless induction was 80% for the parental strain, 60% for the *lexB31* strain and 20% for the *lexB34* strain. Bacterial survival after 180 min of thymine starvation was, respectively, 5×10^{-3}, 1.5×10^{-2}, and $3 \times$

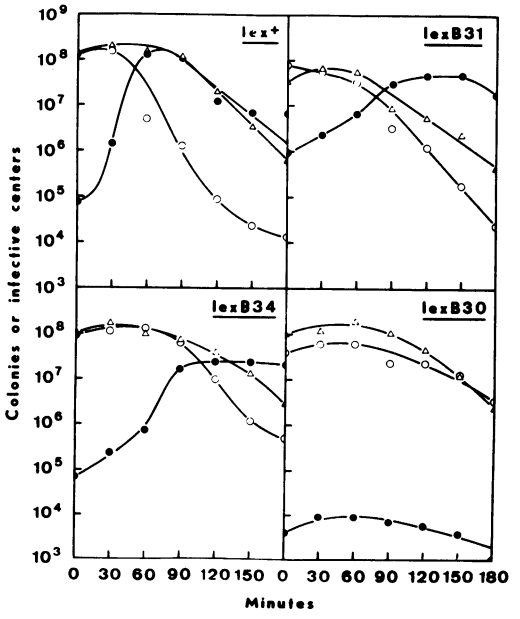

FIG. 5. *Thymineless death and induction of nonlysogenic (△) and 434immλ lysogenic (○) lexB⁻ and lexB⁺ bacteria. Colonies (open symbols) and infective centers (closed symbols) of parental strains GY3424 and GY5209; lexB31 GY3428 and GY5210; lexB34 GY3449 and GY5219; and lexB30 GY3448 and GY5222. Thymine starvation was performed according to method II, and infective centers were plated with GY4015 indicator bacteria on plates containing ampicillin.*

10^{-2} for the nonlysogenic strains; this indicates that the last two strains are relatively resistant to thymineless death.

The nonlysogenic *lexB30* cells were as resistant to thymine deprivation as the *lexB34* bacteria, and, as expected, the *lexB30* lysogen was not inducible since it was selected as such (16).

lexB mutants are able to perform some genetic recombination. The ability of *lexB* mutants to produce Thr$^+$ Leu$^+$ Str^r recombinants was tested, for *lexB30*, in crosses with Hfr donors 3000 and P4X6B and, for *lexB31*, in crosses with Hfr donor 3000. An F$^-$ *recA11* was used as a negative control (Table 4). When *lexB31* or *recA11* recipients were used, we checked the ability of the recipients to give rise to F- *thr*$^+$ *leu*$^+$ transconjugants.

We determined the recombination index of various *lexB* and *recA* mutants. Strains *lexB30*, *lexB31*, and *recA11* had recombination indexes of about 0.30, 0.015, and 0.0002, respectively, compared with about 0.01 for *lexB34* (25) and 0.0002 for *recA13* (9). *lexB* mutants retain a proficiency in genetic recombination that the *recA* mutants do not.

Host cell reactivation and UV reactivation of phage λ. Host cell reactivation of UV-irradiated phage λ depends on host repair enzymes. In *lexB* mutants, except those carrying *lexB30*, host cell reactivation has been found to be altered as much as in RecA$^-$ bacteria (4). Host cell reactivation was slightly higher with *lexB30* than with *lexB31* (Table 5). The decrease in host cell reactivation in LexB$^-$ bacteria was due not entirely to DNA degradation by the *recBC* nuclease, but also to a function proper to *recA* as suggested by the deficiency of host cell reactivation in *recA recBC* bacteria.

UV reactivation (see review in reference 18) of UV-irradiated phage λ depends on an inducible error-prone host repair system (13, 21). Survival of UV-damaged phage λ was higher in UV-irradiated *lexB31* and *lexB34* than in the *lexB30* mutant. There was a slight UV reactivation of phage λ in *lexB34* bacteria (Table 5). UV-irradiated *lexB30* bacteria were unable to reactivate UV-irradiated phage λ.

DISCUSSION

In *E. coli* K-12, three mutations, located between *cysC* and *pheA* in the *recA* region, have been characterized (16, 44; this paper). We have designated them *lexB30*, *lexB31*, and *lexB34* because of their similarity with *lexA* mutations.

The *lexA* and *lexB* mutations are similar to *recA* in causing: (i) a decrease in repair capacity; (ii) a decrease in inducibility of prophage λ after UV irradiation; and (iii) high DNA degradation after UV irradiation (5, 25). *lexB* mutants differ from *recA* in that: (i) the loss of repair capacity is not as high; (ii) they still have a certain proficiency of recombination (almost wild type for *lexB30*); (iii) for both *lexB31* and *lexB34*, they display a delayed induction of prophage λ; (iv) their DNA is not spontaneously degraded (B. W. Glickman, personal communication); and (v) a λFec$^-$ phage is able to plate on them. The last property provides a clear-cut distinction between *lexB* and *recA* mutations.

Some other mutations, called *zab*, have a phenotype and genetic location very similar to that of *lexB* (8). They have been selected as suppressors of *tif-1*, a mutation conferring thermosensitivity for induction and filamentation to the cell (7, 31). *zab* mutations were located

TABLE 4. *Ability of lexB30 and recA11 recipients to produce recombinants*[a]

Recipient	*lex, rec* markers	Mating time (min)	No. of sexductants or recombinants per ml				Recombination index
			Thr$^+$ Leu$^+$ Str^r in crosses with:			Pyr$^+$ Str^r in crosses with 3000	
			GY2549	3000	P4X6		
GY3262	*lex*$^+$ *rec*$^+$	20		8.7×10^5	1.0×10^5		1*
		60		4.0×10^6	9.0×10^5	2.7×10^4	1*
GY3248	*lexB30 rec*$^+$	20		3.2×10^5			0.37*
					3.3×10^5		3.3*
		60		8.0×10^5			0.20*
					1.8×10^6		2*
						2.0×10^3	0.07*
GY3266	*lex*$^+$ *recB21*	60			4.0×10^2		0.004*
GY3246	*lexB30 recB21*	60			1.0×10^2		0.001*
JM692	*lex*$^+$ *rec*$^+$	30	1.7×10^4	3×10^6			1
GY5207	*lex*$^+$ *recA11*	30	1.25×10^4	4.5×10^2			0.0002
GY5208	*lexB31 rec*$^+$	30	1.5×10^4	4×10^4			0.015

[a] The donor strains are 3000 (Hfr PO1), P4X6B (Hfr PO3), and GY2549 (F101 PO1). The values of the recombination index in which the capacity of the recipients to give rise to sexductants was not taken into consideration are indicated by an asterisk.

TABLE 5. *Host cell reactivation (HCR) and UV reactivation (UV-R) of phage 434immλ in lexB strains*

Host strain	UV dose (J/m²) to:			No. of phage/ml		Repair efficiency (%)[a]				Frequency of clear mutations ($\times 10^4$) (expt II)
	Phage					Expt I		Expt II		
	Expt I	Expt II	Bacteria	Expt I	Expt II	HCR	UV-R	HCR	UV-R	
GY3424 (*lex*⁺)	0	0	0	1.8×10^8	1.3×10^{10}					2
	90	150	0	4.1×10^6	6.5×10^7					1
	0	0	50	1.4×10^8	1.3×10^{10}					2
	90	150	50	1.55×10^7	3.4×10^8		42		30	36
GY3428 (*lexB31*)	0	0	0	1.6×10^8	1.2×10^{10}					2
	90	150	0	1.1×10^6	1.6×10^7	−32		−25		1
	0	0	10	1.4×10^8	1.1×10^{10}					1
	90	150	10	5.0×10^5	1.3×10^7		−13		−2	2
GY3448 (*lexB30*)	0	0	0	1.4×10^8	1.4×10^{10}					0.5
	90	150	0	1.7×10^6	4.0×10^7	−16		−11		0.5
	0	0	10	1.4×10^8	1.2×10^{10}					0.5
	90	150	10	8.0×10^5	1.4×10^7		−17		−15	<1
GY3449 (*lexB34*)	0	0	0	1.7×10^8	1.3×10^{10}					<2
	90	150	0	1.1×10^6	1.5×10^7	−33		−28		1
	0	0	10	1.4×10^8	1.0×10^{10}					1
	90	150	10	1.3×10^6	1.7×10^7		7		6	6
AB1157 (*rec*⁺)		0	0		1.5×10^{10}					2
		150	0		3.5×10^7					2
		0	50		1.5×10^{10}					1
		150	50		3.0×10^8				35	30
JC5547 (*recA13 recB21 recC22*)		0	0		1.2×10^{10}					1
		150	0		1.3×10^7			−13		1
		0	10		1.2×10^{10}					0.5
		150	10		6.3×10^6				−12	<2

[a] The evaluation of repair efficiency (E) is given by the formula:

$$E = \frac{(\log S_1 - \log S_0) - (\log S_1' - \log S_0')}{\log S_1 - \log S_0}$$

where S_0 and S_1 are, respectively, the survivals of the nonirradiated and irradiated phage in the control, and S_0' and S_1' are corresponding values in the assay. For HCR determination, the control is performed with nonirradiated wild-type bacteria and the assay is performed with nonirradiated mutant bacteria; for UV-R determination, the control is performed with nonirradiated bacteria and the assay is performed with irradiated bacteria. This formula, derived from Jagger (29), is used because λ survival curves have an exponential shape (4).

close to *tif-1* (8), and it has not yet been possible to study *zab* out of a *tif* genetic background. The *lexB* mutations are not subject to the same limitation.

The characteristics and mapping of the *lexB*, *zab*, *tif*, and *recA* mutations suggest that they may be in the same gene (M. Castellazzi, P. Morand, J. George, and G. Buttin, Mol. Gen. Genet., in press; P. Morand, A. Goze and R. Devoret, Mol. Gen. Genet., in press; K. McEntee, personal communication).

The properties of the *lexB* mutants may help to clarify the different cellular processes dependent on the *recA* gene. These processes may result from two types of functions: (i) constitutive functions—recombination, inhibition of exonuclease V activity (as exemplified by the spontaneous DNA degradation of *recA* mutants and by the inability of λFec⁻ phage to plate on *recA* mutants), and recombinational repair (39, 40); and (ii) induced functions—leading to prophage development, error-prone repair (14), filament formation (45), synthesis of protein X (22), and enhancement of DNA degradation (37) (see reviews in references 10, 38).

The *lexB30* mutant appears to be able to perform all the constitutive functions: it is almost as recombination proficient as the wild-type strain and allows λFec⁻ to plate. It is unable to perform induced functions as seen by lack of UV reactivation, of control of DNA degradation (5), and of prophage induction. Conversely, Lloyd and Low (32) have described a mutation, *recA255*, that appears to affect only the constitutive and not the inducible functions: the *recA255* mutant is recombination de-

ficient, does not allow λFec⁻ to grow, has high spontaneous DNA degradation but no DNA degradation increased by UV irradiation, and is inducible for prophage λ.

At least two models are possible to explain the pleiotropy of *recA* mutants. In model 1, *recA* is a gene that regulates the expression of other genes (some of them located next to *recA*) that are involved in the numerous processes that appear modified in a RecA mutant. Different mutations in the *recA* region lead to different regulatory activities and phenotypes. An example of this type of model is given by Gudas and Pardee (22). In model 2, the *recA* protein is a protein that interacts (alone or complexed with other proteins) with the DNA. Different mutations located at the *recA* locus alter the *recA* protein, and the various phenotypic characters observed are a consequence of this alteration.

According to the first model, it is hard to explain how three properties, radioresistance, inducibility, and recombination proficiency, may be affected differently and independently in every *lexB* mutant.

The second model accounts for the variable properties of all the *lexB* mutants on the assumption that *lexB* mutations are in the *recA* locus.

T. Kato and Y. Shinoura have isolated mutants of *E. coli* in which mutations are not induced with UV light (in preparation). Three of them have been also characterized as *lexB* mutants (Glickman, personal communication). The varied phenotypes of six *lexB* mutants provide strong support for the second model.

ACKNOWLEDGMENTS

This work benefited from the technical assistance of Danièle Foiret and Mélanie Pierre. We are thankful to Albert Goze, Steve Sedgwick, and Brooks Low for helpful discussions. We are grateful to Barbara Bachmann and Jacqueline George for providing strains.

This report was supported by grant 147-75-1-BIO F from EURATOM.

LITERATURE CITED

1. **Arber, W.** 1960. Transduction of chromosomal genes and episomes in *Escherichia coli*. Virology 11:273–288.
2. **Bachmann, B. J.** 1972. Pedigrees of some mutant strains of *Escherichia coli* K-12. Bacteriol. Rev. 36:525–557.
3. **Bachmann, B. J., K. B. Low, and A. L. Taylor.** 1976. Recalibrated linkage map of *Escherichia coli* K-12. Bacteriol. Rev. 40:116–167.
4. **Blanco, M., and R. Devoret.** 1973. Repair mechanism involved in prophage reactivation and UV-reactivation of UV-irradiated phage λ. Mutat. Res. 17:293–305.
5. **Blanco, M., A. Levine, and R. Devoret.** 1975. *lexB:* a new gene governing radiation sensitivity and lysogenic induction in *Escherichia coli* K12, p. 377–380. *In* P. Hanawalt and R. B. Setlow (ed.), Molecular mechanisms for repair of DNA, part A. Plenum Press, New York.
6. **Bonhoeffer, F.** 1966. DNA transfer and DNA synthesis during bacterial conjugation. Z. Vererbungsl. 98:141–149.
7. **Castellazzi, M., M. George, and J. Buttin.** 1972. Prophage induction and cell division in *E. coli*. I. Further characterization of a thermosensitive mutation *tif-1* whose expression mimics the effect of UV irradiation. Mol. Gen. Genet. 119:139–152.
8. **Castellazzi, M., J. George, and G. Buttin.** 1972. Prophage induction and cell division in *E. coli*. II. Linked (*recA, zab*) and unlinked (*lex*) suppressors of *tif-1* mediated induction and filamentation. Mol. Gen. Genet. 119:153–174.
9. **Clark, A. J.** 1967. The beginning of a genetic analysis of recombination proniciency. J. Cell. Physiol. 70(Suppl. 1):165–180.
10. **Clark, A. J.** 1973. Recombination-deficient mutants of *E. coli* and other bacteria. Annu. Rev. Genet. 7:67–86.
11. **Clark, A. J., and A. D. Margulies.** 1965. Isolation and characterization of recombination-deficient mutants of *Escherichia coli* K12. Proc. Natl. Acad. Sci. U.S.A. 53:451–459.
12. **Cottin, M., and M. Lefort.** 1956. Etalonnage absolu du dosimètre au sulfate ferreux. Rayons X mous de 10 et 8 KeV. J. Chim. Phys. 53:267–273.
13. **Defais, M., P. Caillet-Fauquet, M. S. Fox, and M. Radman.** 1976. Induction kinetics of mutagenic DNA repair activity in *E. coli* following ultraviolet irradiation. Mol. Gen. Genet. 148:125–130.
14. **Defais, M., P. Fauquet, M. Radman, and M. Errera.** 1971. Ultraviolet reactivation and ultraviolet mutagenesis of λ in different genetic systems. Virology 43:495–503.
15. **Demerec, M., E. A. Adelberg, A. J. Clark, and P. E. Hartman.** 1966. A proposal for a uniform nomenclature in bacterial genetics. Genetics 54:61–76.
16. **Devoret, R., and M. Blanco.** 1970. Mutants of *Escherichia coli* K12 (λ) non-inducible by thymine deprivation. I. Method of isolation and classes of mutants obtained. Mol. Gen. Genet. 107:272–280.
17. **Devoret, R., M. Blanco, and A. Bailone.** 1972. Mutations in *E. coli* K12 which render prophage λ noninducible, p. 321–330. *In* N. P. Dubinin and D. M. Goldfarb (ed.), Molecular mechanism of genetic processes. Nauka Publishers, Moscow.
18. **Devoret, R., M. Blanco, J. George, and M. Radman.** 1975. Recovery of phage λ from ultraviolet damage, p. 155–171. *In* P. Hanawalt and R. B. Setlow (ed.), Molecular mechanisms for repair of DNA, part A. Plenum Press, New York.
19. **Devoret, R., and J. George.** 1967. Induction indirecte du prophage λ par le rayonnement ultraviolet. Mutat. Res. 4:713–734.
20. **Fuerst, C. R., and L. Siminovitch.** 1965. Characterization of an unusual defective lysogenic strain of *Escherichia coli* K12 (λ). Virology 27:449–451.
21. **George, J., R. Devoret, and M. Radman.** 1974. Indirect ultraviolet-reactivation of phage λ. Proc. Natl. Acad. Sci. U.S.A. 71:144–147.
22. **Gudas, L. J., and A. B. Pardee.** 1975. Model for regulation of *Escherichia coli* DNA repair functions. Proc. Natl. Acad. Sci. U.S.A. 72:2330–2334.
23. **Hane, W., and T. H. Wood.** 1969. *Escherichia coli* K-12 mutants to nalidixic acid: genetic mapping and dominance studies. J. Bacteriol. 99:238–241.
24. **Hertman, I., and S. E. Luria.** 1967. Transduction studies on the role of a *rec*⁺ gene in the ultraviolet induction of prophage lambda. J. Mol. Biol. 23:117–133.
25. **Hoekstra, W. P. M., P. K. Storm, and E. M. Zuidweg.** 1974. Recombination in *Escherichia coli*. VI. Charac-

terization of a recombination-deficient mutation with unusual properties. Mutat. Res. 23:319-326.
26. Howard-Flanders, P., and R. P. Boyce. 1966. DNA repair and genetic recombination: studies on mutants of *Escherichia coli* defective in these processes. Radiat. Res. Suppl. 6:156-184.
27. Ikeda, H., and J. Tomizawa. 1965. Transducing fragments in generalized transduction by phage P1. I. Molecular origin of the fragments. J. Mol. Biol. 14:85-109.
28. Inouye, M. 1971. Pleiotropic effect of the *recA* gene of *Escherichia coli:* uncoupling of cell division from deoxyribonucleic acid replication. J. Bacteriol. 106:539-542.
29. Jagger, J. 1960. Photoreactivation, p. 352-377. *In* A. Hollaender (ed.), Radiation protection and recovery. Pergamon Press, New York.
30. Kaiser, A. D., and F. Jacob. 1957. Recombination between related temperate bacteriophage and the genetic control of immunity and prophage localization. Virology 4:509-521.
31. Kirby, E. P., F. Jacob, and D. A. Goldthwait. 1967. Prophage induction and filament formation in a mutant strain of *Escherichia coli*. Proc. Natl. Acad. Sci. U.S.A. 58:1903-1910.
32. Lloyd, R. C., and B. Low. 1976. Some genetic consequences of changes in the level of *recA* gene function in *Escherichia coli* K12. Genetics 84:675-695.
33. Low, K. B. 1972. *Escherichia coli* K-12 F-prime factors, old and new. Bacteriol. Rev. 36:587-607.
34. Monk, M., and J. Kinross. 1972. Conditional lethality of *recA* and *recB* derivatives of a strain of *Escherichia coli* K-12 with a temperature-sensitive deoxyribonucleic acid polymerase I. J. Bacteriol. 109:971-978.
35. Moreau, P., A. Bailone, and R. Devoret. 1976. Prophage λ induction in *Escherichia coli* K12 *envA:* a highly sensitive test for potential carcinogens. Proc. Natl. Acad. Sci. U.S.A. 73:3700-3704.
36. Mount, D. W., K. B. Low, and S. J. Edmiston. 1972. Dominant mutations (*lex*) in *Escherichia coli* K-12 which affect radiation sensitivity and frequency of ultraviolet light-induced mutations. J. Bacteriol. 112:886-893.
37. Pollard, E. C., and E. P. Randall. 1973. Studies of the inducible inhibitor of radiation-induced DNA degradation of *Escherichia coli*. Radiat. Res. 55:265-279.
38. Radman, M. 1974. Phenomenology of an inducible mutagenic DNA repair pathway in *E. coli*. SOS repair hypothesis, p. 128-142. *In* M. Miller (ed.), Molecular and environmental aspects of mutagenesis. Charles C Thomas Publisher, Springfield, Ill.
39. Radman, M., L. Cordone, D. Krsmanovic-Simic, and M. Errera. 1970. Complementary action of recombination and excision in the repair of ultraviolet irradiation damage to DNA. J. Mol. Biol. 49:203-212.
40. Rupp, C. W., C. E. Wilde, D. L. Reno, and P. Howard-Flanders. 1971. Exchanges between DNA strands in ultraviolet-irradiated *Escherichia coli*. J. Mol. Biol. 61:25-44.
41. Stacey, K. A., and E. Simson. 1965. Improved method for the isolation of thymine-requiring mutants of *Escherichia coli*. J. Bacteriol. 90:554-555.
42. Stouthamer, A. H., P. G. de Haan, and J. J. Nijkamp. 1965. Mapping of purine markers in *Escherichia coli* K12. Genet. Res. 6:442-453.
43. Van de Putte, P., J. Van Dillewijn, and A. Rörsch. 1964. The selection of mutants of *Escherichia coli* with impaired cell division at elevated temperature. Mutat. Res. 1:121-128.
44. Van de Putte, P., H. Zwenk, and A. Rörsch. 1966. Properties of four mutants of *Escherichia coli* defective in genetic recombination. Mutat. Res. 3:381-392.
45. Witkin, E. M. 1967. The radiation sensitivity of *Escherichia coli* B: a hypothesis relating filament formation and prophage induction. Proc. Natl. Acad. Sci. U.S.A. 57:1275-1279.
46. Zissler, J., E. Signer, and F. Schaeffer. 1971. The role of recombination in growth of bacteriophage lambda. I. The gamma gene, p. 455-475. *In* A. D. Hershey (ed.), The bacteriophage lambda. Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y.