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PROPERTIES OF CLONED DOMINANT Gam18 ALLELE

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Mutant alleles Gam^r capable to enhance radiation-resistance of *E. coli* wild-type cells were cloned from the Gam^{r}444 mutant in a plasmid mini-Mu vector [1]. After introduction of these pGam^r plasmids into *recA13* or *rpoH15* mutants, high level of radiation-resistance was not obtained. Therefore expression of the cloned dominant gam^r mutationsl depends on *SOS* and *HSP* regulons.

Null mutation in chromosomal gene corresponding to *gam^{r}18* allele was constructed. Random insertions of mini-Tn10-Km^r transposon [2] into pGam18 were obtained by hop-transposition from λ1105 vector and a plasmid pGam^{s}18::Km^r (Cm^r), which had lost ability to transfer Gam^r phenotype and carries a *gam^{s}18*::Km^r insertion in the cloned Gam^r gene, was selected. Very slowly growing Km^{r}Cm^s transformants were selected after transformation of *recBC sbcB* recipient with pGam^{s}18::Km^r (Cm^r) plasmid DNA cut with EcoRI endonuclease. They are extremely sensitive to γ-rays and presumably are carrying chromosomal insertion *gam^{s}18*::Km^r. The behavior of this mutation in *recBC sbcB* background is typical for mutations in the *RecF* pathway.

Plasmid pGam18 restores radioresistance of *recBC sbcB gam^{s}18*::Km^r mutant and partly compensates defect in *uvrD* gene (Table 1), i.e. *gam^{r}18* mutation may be concerned with a helicase involved in repair and recombinant process. Transduction of *Kan* marker by Kohara phages [3] revealed homology of plasmid-borne allele with some helicases which had 3'→5' polarity (Table 2).

Thus, the dominant mutation *gam^{r}18* is related to RecF pathway of repair and recombination and mutant protein can be a reparative helicase.

Table 1

Effect of pGam18 plasmid on
radiosensitivity of *E. coli* strains

E. coli strains	Genotype /pGam18	LD_{10} Gy −	LD_{10} Gy +	DMF
AB1157	*wild type*	350	667	1.90
JC7623	*recB21 C22 sbcB15*	470	680	1.45
JC8679	*recB21 C22 sbcA23*	400	333	0.83
JC7902	*recB21 C22 sbcB15 recF144*	275	200	0.73
JC7636	*recB21 C22 sbcB15 recJ148*	215	60	0.28
HA1156	*uvrD210 ilv::Tn10*	187	271	1.45
KS114	*uvrD502*	170	247	1.45
JC9239	*recF143*	170	140	0.82
RDK1541	*rec01504::Tn5*	165	165	1.00
SP256	*recN262*	140	155	1.11
KD1996	*recQ61 polA12(ts)*	130	160	1.23
TY108	*recR::Tn5*	113	120	1.06
FB154	*recB21 C22 ruv52*	105	110	1.05
JC8171	*recB21 C22 sbcB15 uvrD152*	50	255	5.10

Table 2

λ-mediated replacement of chromosomal
alleles by plasmid-borne ones

Kohara phage	Relevant traits	Polarity of helicase	Kam insertion	MC1061 (lysogen) Transduction frequency $\cdot 10^7$	MC1061 (lysogen) Fraction Cm^s (%)
λ222	*helD*	3' to 5'	*gam18*	15	7
λ459	*recBCD*	No	"	10	0.6
λ538	*priA*	3' to 5'	"	170	23
λ552	*uvrD*	3' to 5'	"	1300	76
λ555	*rho*	3' to 5'	"	9	33
λ556	*rep rho*	3' to 5'	"	200	25

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