GENETIC CHARACTERIZATION OF NOVEL MUTANTS OF RecA GENE IN ESCHERICHIA COLI K-12

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Genetical properties of 7 novel and one known (recA200^{ts}) mutants localized in 3 different regions of the RecA protein structure proposed by Story and Steitz (1992) have been analyzed. Six mutants were localized in the region "αH-loop-β9" of the structure. Their amino acid substitutions are as follows: 283Leu→Pro (recA200), 283Leu→Glu, 284Ile→Asp, 278Gly→Pro, 278Gly→Thr+275, Val→Phe, and 277Leu→Asp. The analysis of recombination repair and SOS properties of the mutants permits to separate a group of thermosensitive mutants with the following phenotypes: REC^{ts}-UV^{ts}-SOS^{ts}- (L283P), REC^{ts}-UV$^{ts\pm}$SOS^{ts}- G278P and REC^{ts}-UV^{+}SOS$^{ts\pm}$ (G278T+V275F). The biochemical properties of the latter most intriguing mutant protein are under study now. According to preliminary results it contains the deficiency in the site for binding the RecA protein to single-stranded DNA.

The amino acid substitution 223Leu→Met in the β6 region provides the mutant phenotype $REC^{ts\pm}UV^{ts\pm}SOS^{ts-}$ with clearly expressed deficiency in the SOS response. Summarizing with the properties of other known mutants in this region we can suppose that the protein region is responsible for the RecA corepressor activity.

The substitution 183Lys→Met in the αF helix produces a strong $REC^{-}UV^{-}SOS^{-}$ phenotype. According to our preliminary data with purified mutant protein and data of other authors the region contains a possible site for the double strand DNA binding.