Molec. gen. Genet. 122, 287—290 (1973)

Short Communication

Sensitivity to X-Radiation of Strains of *Escherichia coli* K-12 which Lack DNA Polymerase II

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Received, February 7, 1973

Summary. The *polB1* and *polA1 polB1* strains of *E. coli* K-12, wihch are deficient in DNA polymerase II and in DNA polymerases I and II, respectively, were found to have essentially the same sensitivity to anoxic or aerobic X-irradiation as their related wild-type and *polA1* strains, respectively. Thus, DNA polymerase II appears to play no major role in the repair of X-ray damage.

Introduction

Mutant strains of *E. coli* K-12 which lack each of the three known DNA polymerases have been isolated. The *polA1* mutation results in the loss of DNA polymerase I activity (de Lucia and Cairns, 1969) and sensitizes the cell to ultraviolet (Gross and Gross, 1969) and X-radiation (Paterson *et al.*, 1971; Town *et al.*, 1971). The *dnaE* mutation results in the temperature-sensitive loss of DNA polymerase III activity (Gefter *et al.*, 1971) and also increases sensitivity to X-radiation (D. A. Youngs and K. C. Smith, in preparation). The increased X-ray sensitivity of *polA* and *dnaE* strains appears to be due at least partially to the deficiency of these strains in the Type II (buffer) repair of X-ray-induced single-strand breaks in DNA (Town *et al.*, 1971; Youngs and Smith, in preparation).

A *polB* strain lacking DNA polymerase II has been isolated and was found to be no more sensitive than the *polB*$^+$ strain to ultraviolet radiation (Campbell *et al.*, 1972). Genetic recombination was not altered by the presence of a *polB1* mutation (Campbell *et al.*, 1972).

Hirota *et al.* (1972) have confirmed these results with an independently isolated *polB* mutant. In addition they have found that the *polB* mutation does not alter sensitivity to treatment with mitomycin C or methylmethane sulfonate.

The present study examines the X-ray sensitivity of strains containing the *polB1* mutation.

Materials and Methods

1. Bacterial Strains

The following closely related F$^-$ strains of *E. coli* K-12 were used: (JG139) *thy lac rha str* and (JG138) *thy lac rha str polA1*, obtained from Dr. J. D. Gross; (HMS83) *thy lac rha str lys polA1 polB1* and (HMS85) *thy lac str lys polB1*, described by Campbell *et al.* (1972).

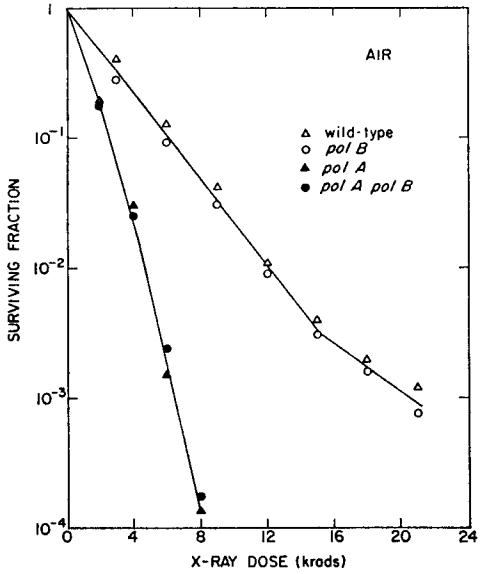

Fig. 1. Survival after aerobic X-irradiation. Cells were grown in MM medium to exponential phase, irradiated in DTM buffer while bubbling with air, and plated on MM agar plates. The data represent the average of two experiments. Symbols are: (△) JG 139 wild-type; (○) HMS83 *polB1*; (▲) JG138 *polA1*; (●) HMS85 *polA1 polB1*

2. X-Ray Survival

The methods used for examining X-ray survival have been described (Youngs and Smith, 1973). Cells were grown in a minimal medium (MM; Ganesan and Smith, 1968) to exponential growth phase, filtered on 0.45 μm Millipore filters, and resuspended in DTM (MM without glucose, thiamine, or amino acids). Samples were bubbled with air for 3 min or N_2 (> 99.996 purity) for 5 min prior to, and during, X-irradiation. The cells were diluted in buffer (11.7 g Na_2HPO_4 and 7.1 g KH_2PO_4 per liter, pH 7.0) and plated on MM agar (MM medium solidified with 0.9% Oxoid agar agar No. 3) supplemented with 10 μg/ml thymine and 0.5 μg/ml thiamine hydrochloride. The MM medium and agar for strains HMS83 and HMS85 also contained 50 μg/ml L-lysine.

Results and Discussion

The survival curves for the wild-type, *polA1*, *polB1*, and *polA1 polB1* strains for aerobic X-irradiation are shown in Fig. 1; those for anoxic X-irradiation are presented in Fig. 2. The data points for the *polB1* strains were always slightly lower than those for the parent wild-type strains, so the possibility exists that the *polB1* mutation has a small effect on survival in the $polA^+$ background. However no such effect was observed when the *polA1* and *polA1 polB1* strains were compared. Thus, these results indicate that the *polB1* mutation has little or no effect on X-ray survival, either in wild-type or DNA polymerase I deficient strains. A similar conclusion has been reached concerning the UV sensitivity of these strains (Campbell *et al.*, 1972).

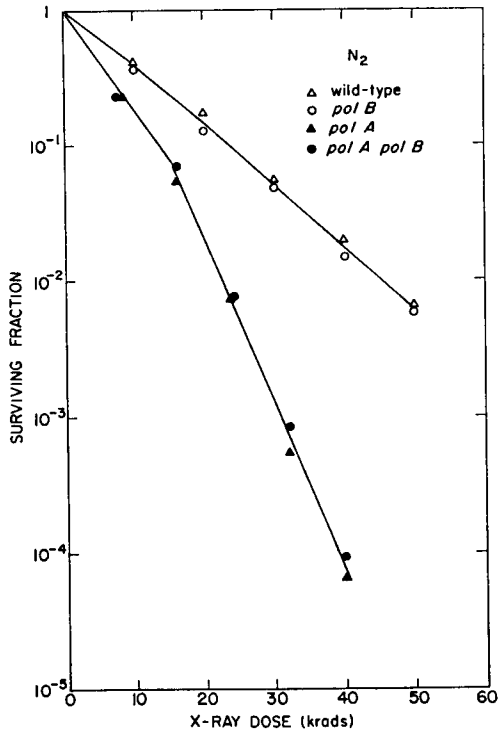

Fig. 2. Survival after anoxic X-irradiation. Procedure and symbols are as in Fig. 1 except that the cells were bubbled with N_2 instead of air

Acknowledgements. The *polB* strains were obtained from Dr. C. C. Richardson via Dr. H. S. Kaplan and we gratefully acknowledge this contribution.

The work was supported by Public Health Service Research Grant CA-02896 and Research Program Project Grant CA-10372 from the National Cancer Institute.

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Communicated by E. Witkin

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