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Characterization of *RAD4* gene in *Schizosaccharomyces pombe*

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The *RAD4* gene of *Saccharomyces cerevisiae* is one of the five genes (*RAD1, RAD2, RAD3, RAD4, and RAD10*) that are absolutely required for the incision step in nucleotide excision repair. Mutation in any of these five genes renders yeast cells abnormally sensitive to UV light and other DNA-damaging agents. In our previous studies we have demonstrated that the cloned *RAD4* gene is composed of 2,190 nucleotides encoding a putative protein of 730 amino acids, and transcribed into 2.3 kb mRNA. We have further observed that the *RAD4* gene is neither essential gene for viability of the haploids under normal growth condition nor UV- inducible. The *RAD4* gene is essential for the nucleotide excision repair in *Saccharomyces cerevisiae*. It has been known that the deduced amino acid sequence of Rad4 protein contains three DNA-dependent ATPase/ helicase motifs. To determine the biochemical activities and functional role of *RAD4* gene, Rad4 protein was expressed and purified. Immunoblot analysis showed a specific band of 21 kDa, which was very well matched with the size of open reading frame of *RAD4* gene. The purified Rad4 protein had no detectable helicase activity, however, the protein could interact with double stranded oligonucleotides, as judged by mobility shift assay. This result suggests that the Rad4 protein is a DNA binding protein.