EXTRACTION, PURIFICATION AND CHARACTERIZATION OF PROTEASE OF Pseudomonas fluorescens ISH AND IT'S ROLE IN DETERIORATION OF IRAQI SOFT CHEESE 2- EXTRACTION, PURIFICATION AND CHARACTERIZATION OF PROTEASE OF P. fluorescens ISHAND EFFECT OF IT ON MILK PROTEINS

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ABSTRACT

The enzyme was purified by 30-80% saturation of ammonium sulphate precipitation, ion exchange chromatography on DEAE-Sepharose column and by gel filtration on Sephacryl S-200, with 18.5 fold and 32.8% recovery.

The results of the enzyme characterization showed that it's estimated molecular weight was 47.2 kD. The optimum pH of activity was 8.0 and it'soptimum pH of stability was 7.5. The optimum temperature for the activity was 40° C, and the enzyme was retained its original activity after incubation for 10 minutes at temperatures ranging from 30-40 $^{\circ}$ C.

The protease was metalloprotease because it was affected by the chelating agent EDTA.

The protease showed high affinity toward β -casein, compared with its activity towards α s-casein and k-casein.

Keywords: Pseudomonas fluorescens, Psychrotrophic bacteria, proteases, soft cheese.