

CHAPTER V

SUMMARY

The results of the present investigation can be summarized as follows :

1. The $^1\text{H-NMR}$ is a suitable method for the determination of glycine betaine in *A. halophytica*.

2. The amount of glycine betaine in *A. halophytica* grown in non salt-stressed condition (0.5 M NaCl) was 9.7 nmol/ 10^6 cells. The glycine betaine accumulation was increased 8-fold in salt-stressed condition (2 M NaCl).

3. The *A. halophytica* BADH was partially purified by ammonium sulfate fractionation and DEAE-cellulose column chromatography. The BADH purification was 18-fold with a specific activity of 290.8 $\mu\text{mol min}^{-1} \text{mg}^{-1}$ and a recovery of 8.6%. K_m values for betaine aldehyde and NAD^+ were 91 and 71.4 μM , respectively. NADP^+ was a less preferred coenzyme ($K_m = 100 \mu\text{M}$). The V_{max} of BADH was 175.4 $\mu\text{mol min}^{-1} \text{mg}^{-1}$.

4. The optimal condition for *A. halophytica* BADH was pH 7.5 at 25°C. The enzyme was inhibited by substrate analogs such as ethanolamine and acetaldehyde. Aldehyde analog appeared to be a more potent inhibitor than N-methylated analogs.

5. The enzyme activity was initially stimulated by increasing concentration of NaCl and KCl from 0 to 0.1 M above which the BADH activity was decreased. CaCl₂ and MgCl₂ appeared to have a particularly strong inhibition on BADH.

6. *A. halophytica* BADH activity was strongly inhibited by sulfhydryl-reactive compound such as PCMS but incubating the enzyme with reducing agent such as DTT before or after PCMS treatment was able to relieve the inhibition by PCMS.

7. The molecular weight of *A. halophytica* BADH was 120,000 dalton. The holoenzyme was most likely a tetramer of 30,000 dalton subunits.

8. The high external salinity resulted in the increased specific activity of *A. halophytica* BADH.