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Guoqiang Wu Lanzhou University, China

Jiejun Xi Lanzhou University, China

Aike Bao Lanzhou University, China

Suomin Wang Lanzhou University, China

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The XXI International Grassland Congress / VIII International Rangeland Congress took place in Hohhot, China from June 29 through July 5, 2008.

Proceedings edited by Organizing Committee of 2008 IGC/IRC Conference

Published by Guangdong People's Publishing House

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Molecular cloning and characterization of a vacuolar Na^+/H^+ antiporter gene from the succulent xerophyte $Z_{ygophyllum xanthoxylum}$

Guo-Qiang Wu, Jie-Jun Xi, Ai-Ke Bao, Suo-Min Wang

 $Key\ Laboratory\ of\ Grassland\ A\ gro-ecosystem\ ,\ Ministry\ of\ A\ griculture\ ,$

School of Pastoral A griculture Science and Technology, Lanzhou University, Lanzhou 730000, People's Republic of China. * Corresponding author E-mail: smwang@lzu.edu.cn

Key words : Xerophyte ,vacuolar Na^+ / H^+ antiporter ,Zx NHX

Introduction Salinity is an environment factor that adversely affects plant growth and development. High salinity causes osmotic and oxidative stresses, and the damage in ion homeostasis. To cope with salt stress, plants have developed a variety of adaptation mechanisms. One of them is to compartmentalize Na⁺ into the vacuole through the operation of a vacuolar Na⁺/H⁺ antiporter (NHX) (Apse *et al.*, 1999). So far, NHX genes have been isolated from the glycophytes, such as AtNHXI in *A rabidopsis thaliana*, OsNHXI in rice and GhNHXI in cotton, and the halophytes, such as SsNHX in *Suaeda salsa* and SeNHXI in *Salicornia europaea*. However, NHX genes from the xerophytes have been not reported. In this study, a homologous gene (ZxNHX) of vacuolar Na⁺/H⁺ antiporter was cloned and characterized from a typically succulent xerophyte, $Z\gammagophyllum xanthoxylum$.

Materials and methods The core fragment of ZxNHX was cloned by RT-PCR. The full-length cDNA ends of ZxNHX were isolated by RACE system. Sequence analysis was run on the software DNAstar. Homology comparison was analyzed using DNAMAN software.

Results The full-length cDNA of vacuolar Na⁺ / H⁺ antiporter gene from Z. xanthoxylum was obtained by assembling the core cDNA fragment (Figure 1 a) , 5 -RACE fragment (Figure 1 b) and 3 -RACE fragment (Figure 1 c) . This cDNA consists of 2127 bp with a 5 -untranslated region of 213 bp , an open reading frame of 1558 bp that encodes a protein of 532 amino acids with a calculated molecular weight of 58 .8 KDa , and a 3 -untranslated region of 315 bp containing a ploy (A) tail . It was named as ZxNHX and deposited in the GenBank database (accession no . EU103624) . The deduced amino acid sequence of ZxNHX shares a high homology with those of the vacuolar Na⁺ / H⁺ antiporters of higher plants , ranging from 80 .7% homology with that of *Populus euphratica PeNHX*² to 72 .7% with that of *Oryzasativa OsNHX*1 , and contains the consensus amiloride-binding domain . Semi-quantitative RT-PCR analysis showed that the expression of *ZxNHX* in *Z*. xanthoxylum was induced and regulated by salt stress .

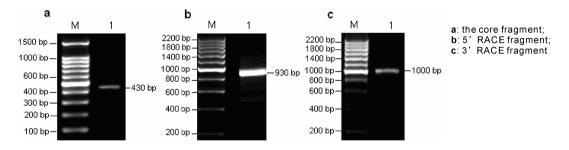


Figure 2 A garose gel electrophoresis of the core fragment and RACE fragments of ZxNHX.

Conclusions The results indicated that the gene ZxNHX isolated from $Z \cdot x$ anthox y lum is a vacuolar type Na⁺ / H⁺ antiporters. This gene may play important roles in the drought tolerance, its mechanism is under further investigation.

Reference

Apse MP, Aharon, GS, Snedden WA, Blumwald E. 1999. Salt tolerance conferred by overexpression of a vacuolar Na⁺ / H⁺ antiporter in A rabidopsis .Science . 285 : 1256-1258.