Isolation and Bioinformatics Analysis of Zma1158 Protein

Yibadaiti·Kadier[1, a], Zu Yiyi[1], Pan Jinbao[2], Sun Qingpeng[3], Lu Min[4, b *]

[1-5]Plant Science and Technology College, Beijing University of Agriculture, Beijing, 1002206, P.R. China

[a]940518026@qq.com, [b]Lumin.sdau@Gmail.com

[1]Both authors contributed equally to this work

*Corresponding author

Keywords: NAC transcription factor, bioinformatics analysis, maize

Abstract. Zma1158 protein, consisted of 386 amino acids, was analyzed by bioinformatics tools. The result showed that Zma1158 was a typical NAC transcription factor in maize, and it may involve into abiotic stress response for the stress related *c*is-elements that detected in the promoter region of Zma1158.

Introduction

NAC proteins are plant-specific transcription factor superfamily that has significant roles in the progress of plants development and stress resistance [1]. NAC proteins share a typical NAC domain (~150 amino acids) in the N-terminal region that contains five conserved motifs based on sequence similarity. In contrast, the C terminus of NAC proteins was quite divergent and was considered as the transcription activation region. Phylogenetic analysis indicated that NAC proteins could be divided into two large groups including eighteen subgroups [2-5]. The NAC members in subgroups OsNAC3, ATAF, NAP were reported to response to various abiotic stresses such as drought and salinity. To data, a series of stress-related NAC proteins were identified and well characterized form Arabidopsis, rice, soybean, wheat and barley. However, little information of stress-responsive NAC genes was obtained in maize. In this study, a NAC gene was isolated from maize. The amino acid sequence analysis, prediction of the *c*is-elements in the promoter region, prediction of Physical and chemical parameters, gene structure analysis and prediction of trans-membrane motifs were performed by bioinformatics tools. This will provide evidence for further functional characterization of the candidate stress responsive NAC genes.

Results

Cloning of the full-length cDNA of *Zma1158* and the amino acid sequence analysis

Specific primers were designed to amplify the full-length cDNA of *Zma1158*. Sequencing results indicated that the full-length of *Zma1158* was 1162 bp that encodes 386 amino acid residues. Multiple sequence alignment showed Zma1158 protein contained one typical NAM domain in the N-terminus that can be divided into five subdomains based on protein sequence similarity（data not shown）. In contrast, the sequences of C-terminal region were quite divergent. Fig.1 shows that the N- and C-terminal regions were considered as the DNA binding domain and the trans-activation domain, respectively.

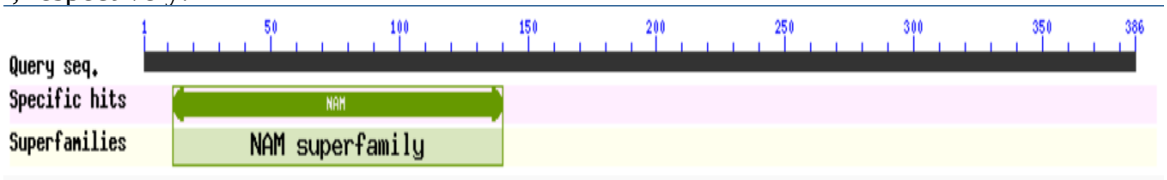

Fig. 1. The amino acid sequences analysis of maize Zma1158.

Prediction of Physical and chemical parameters of *Zma1158*

The theoretical pI and molecular weight of Zma1158 was 6.88 and 41008.44 respectively using Compute pI/Mw tool (http://us.expasy.org/tools/pi_tool.html).

Gene structure analysis of Zma1158

Gene structure analysis indicated that *Zma1158* have three exons and two introns. The Fig.2 was performed with (http://gsds.cbi.pku.edu.cn/) online software.

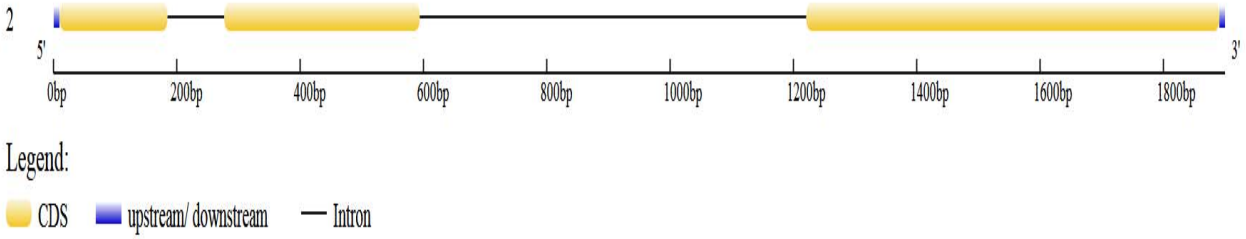

Fig. 2. Gene structure analysis of Zma1158.

Prediction of trans-membrane motifs in maize *Zma1158*

The online software TMHMM version 2.0 (http://www.cbs.dtu.dk/services/TMHMM) was used to identify the potential trans-membrane (TM) motifs in Zma1158 protein. Fig.3 shows that Zma1158 protein was considered as a maize NAC member that have non trans-membrane region.

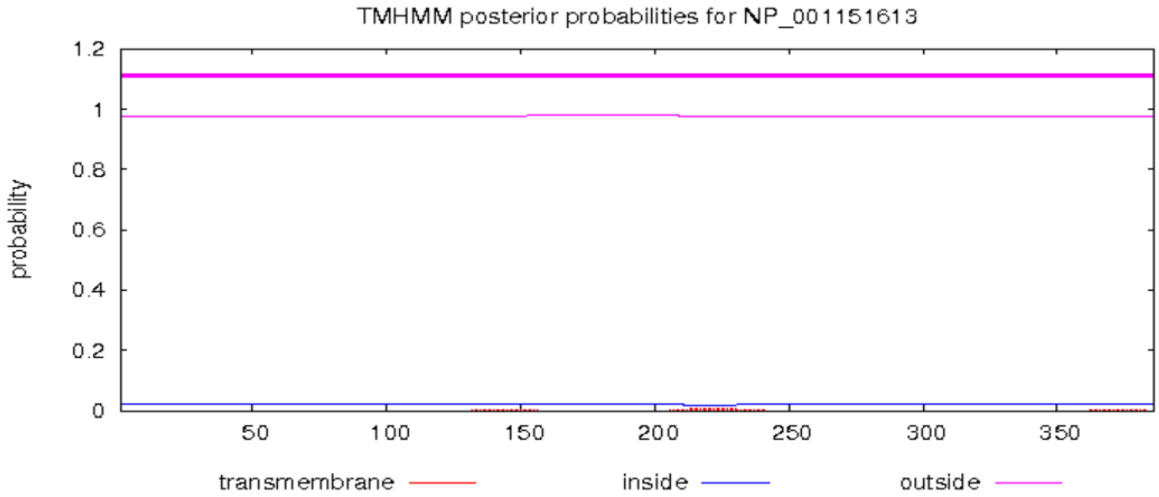

Fig. 3. Prediction of trans-membrane motifs in maize Zma1158.

Prediction of the *c*is-elements in the promter region of *Zma1158*

1.5 kb DNA sequences upstream of the start codon (ATG) of *Zma1158* were downloaded obtained from the NCBI database (http://www.ncbi.nlm.nih.gov/) and considered as the promoter region of *Zma1158*. *C*is-elements in the promoter region were analyzed with the PLACE web signal scan program (http://www.dna.affrc.go.jp/PLACE/signalup.html). Fig. 4 shows that a series of *c*is-elements including phytohormone ABA-responsive elements (ABREs) [6-7], drought-responsive elements and low temperature responsive elements (LTRE) were detected in the promoter region of *Zma1158*.

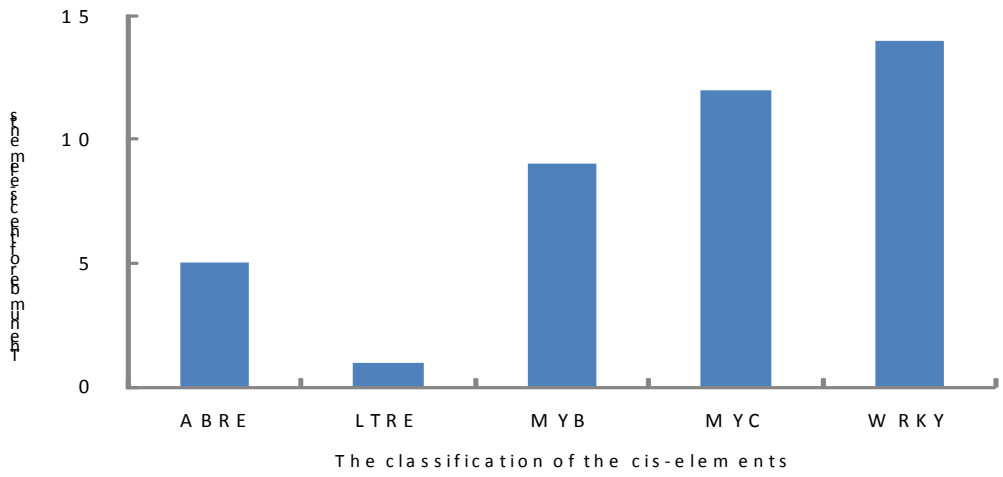

Fig. (4). Prediction of the *c*is-elements in the promoter region of Zma1158.

Conclusion

Zma1158, a member of NAC transcription factor super-family in maize, consisted of 386 amino acid residues. Protein sequence analysis indicated that Zma1158 contains one typical NAC domain and divergent transactivation domain in the C-terminal region. *Zma1158* have three exons and two introns according to the result of gene structure analysis. Bioinformatics analysis showed that Zma1158 has no trans-membrane motifs. The putative *c*is-elements detected in the promoter region of *Zma1158* showed that *Zma1158* may response to various abiotic stresses in ABA-dependent signaling pathway [8-9].

Materials and Methods

Cloning of full-length cDNA of Zma1158

Specfic primers were dignsed to amplify the full-length cDNA of Zma1158 using RT-PCR method. Zma1158 protein sequence was translated from the full-length cDNA of Zma1158 using Genscanonline software (http://genes.mit.edu/GEN-SCAN.html).

Sequence Analysis of Zma1158

NCBI database (http: //www.ncbi.nlm.nih.gov) was used to search for the conserved NAC domains of Zma1158. Mw tool (http://us.expasy.org/tools/ pi_tool.html) was used to measure the pI (isoelectric point) and Mw (molecular weight). Gene structure online software (http://gsds.cbi.pku.edu.cn/) was used to show the gene structure of Zma1158. TMHMM online software (http://www.cbs.dtu.dk/services/TMHMM) was used to identify the potential trans-membrane (TM) motifs. PLACE software (http://www.dna.affrc.go.jp/PLACE/signalup.html) was used to analyze the *c*is-acting elements in the promoter region.

Acknowledgements

This work was supported by State Key Laboratory of Crop Biology foundation (2015KF08), the Research Project of Beijing educational Committee (KM201510020003).

References

[1] N. Kazuo，I. Yusuke and Y. S. Kazuko, Plant Physiol, 149: 88-95(2009)

[2] K. Nakashima and K. Yamaguchi-Shinozaki, Plant Physiol, 126: 62-71(2005)

[3] A. N. Olsen，H. A. Ernst，L. L. Leggio and K. Skriver, 10: 79-87(2005)

[4] K. Kikuchi，M. Ueguchi-Tanaka，K. T. Yoshida，Y. Nagato and M. Matsusoka, Molecular and General Genetics, 262: 1047-1051(1999)

[5] R. Zimmermann and W. Werr, Plant molecular biology, 58: 669-685(2005)

[6] T. Yoshida，Y. Fujita，H. Sayama，S. Kidokoro and K. Maruyama, Plant Journal, 2010, 61 (4): 672–685

[7] H. D. Priest，S. A. Filichkin and T. C. Mockler, Current Opinion in Plant Biology, 2009, 12: 643-649

[8] Q. Feng，S. Kazuo and Y. S. Kazuko, Plant Cell Physiol, 52: 1569-1582(2011)

[9] T. Umezawa，K. Nakashima，T. Miyakawa，T. Kuromori and M. Tanokura, Plant Cell Physiol, 51: 1821-1839(2010)