

## Special Article - Drought Tolerance

# Recent Trend of Genome-Wide Multigene Family Analysis and Their Role in Plant Drought Tolerance

Kakar KU<sup>1</sup>, Akram Ali Baloch<sup>1</sup>, Nawaz Z<sup>2</sup> and Ahmed J<sup>1\*</sup>

<sup>1</sup>Department of Biotechnology, Faculty of Life Sciences & Informatics, Balochistan University of Information Technology, Engineering and Management Sciences, Quetta, Pakistan

<sup>2</sup>Department of Botany, University of Central Punjab, Rawalpindi, Pakistan

\*Corresponding author: Prof. Dr. Jamil Ahmed, Department of Biotechnology, Faculty of Life Sciences & Informatics, Balochistan University of Information Technology, Engineering and Management Sciences, Quetta, Pakistan. Tel: +923456789775; Email: jamil.ahmad@buitms.edu.pk

Received: July 24, 2019; Accepted: July 30, 2019;

Published: August 06, 2019

## Short Communication

Drought stress is a major abiotic stress that have devastating impact on world agriculture. Drought not only reduces the crop productivity and yield but also leads to lower incomes for farmers and weaken global food security [1]. Recent data from numerous independent studies have demonstrated that water stress reduced the average yield by 40% for Maize in 2016, 21% for Wheat in 2016 and 34-68% for cowpea in 2017 [2,3]. The speedy development and adoption of climate-resilient crop genotypes is imperative to ensure global food security [4].

Plants display stress resistance or stress tolerance through acclimation and adaptation mechanisms. To lessen the damages from drought stress, plant breeders, geneticists and farmers are exploring new approaches for resistance breeding in crops. The need to access and investigate genetic diversity on an unprecedented scale in plant genomes and discovery of new resistant genes has become inevitable. In this scenario, latest developments in plant genomics have provided additional resources to accelerate the pace of genetic improvement [5].

Current researches in plant genomics have largely focused on the genomewide identification, characterization and expression profiling of novel multigene families in sequenced organisms. These multigene families are being continuously reported in almost every known and important agricultural crop, including, rice, tomato, maize, citrus, papaya, Brassica, tobacco etc. To the best of our knowledge, more than 100 articles have been published in various journals this year, and the number is increasing gradually. The focus of such studies is to find one or more novel candidate gene(s) associated with plant growth, development and response to environmental stimuli. Examples of the newly identified gene families associated with drought stress response in plants include Mitogen-Activated Protein Kinases (MAPKs), Superoxide Dismutase (SOD), GRAS, Brassinazole-Resistant (BZR),

Auxin Response Factors (ARFs), Alternative Oxidase (AOX), WRKY transcription factors, Metacaspase (MC), Cyclic Nucleotide-Gated Ion Channels (CNGCs), Catalases (CATs), Heat shock transcription factors (Hsfs), Expansin (EXP), NAC family, Multiple Organellar RNA editing Factor (MORF), IQ67-domain (IQD), Teosinte-branched 1/Cycloidea/Proliferating (TCP), Aquaporins (AQPs), Calmodulin-binding Transcription Activators (CAMTAs), Late Embryogenesis Abundant (LEAs) and trihelix etc (Table 1). Many of these gene families have been systematically characterized in multiple plant species, while, others have been reported in particular species only. For example, BZR family of Transcription Factors (TFs), regulating the biosynthesis of brassinosteroids and drought resistance, is comprised of 16 genes in *Glycine max* (soybean), 11 genes in *Zea Mays* (Maize), 7 in *Medicago truncatula* (Barrel medic) and *Phaseolus vulgaris* (common bean), 6 in *Cajanus cajan* (pigeon pea) and *Cicer arietinum* (chickpea), and 5 genes in *Lotus japonicus* (Bridsfoot Trefoil) and *Vigna radiate* (mung bean) [6]. Similarly, the CNGC family genes involved in ion transport, calcium signaling, plant growth and stress response have been identified in Rice, tomato, Chinese cabbage and tobacco [7-9]. Depending on plant species and/or gene family, the proteins of each gene family have their own distinctive roles by using specific mechanism. For instance, MAPKs play central roles in hormone signaling transduction in cassava [10], while ARFs are critical components of the auxin-signaling pathway in chickpea, *Medicago* and *Arabidopsis* [11-13]. However, due to complex gene regulatory networks, crossed signaling pathways and protein-protein interactions, individual gene(s) or gene family contribute to various biological functions including plant growth, development, and biotic and abiotic stress resistance.

The primary and secondary functions of a newly identified gene(s) can be initially assessed by studying their functional domains followed by gene expression patterns or transcript abundances in plants against hormonal, biotic and abiotic stress through Real-Time Quantitative Polymerase Chain Reaction (qRT-PCR), Microarray or RNA-sequencing. Latest transcriptome-wide studies and expression profiling analyses have demonstrated the role of genome wide identified genes in plant drought tolerance. The qRT-PCR of cucumber (*Cucumis sativus*) Metacaspase (CsMC) family that encodes caspases like proteins involved in programmed cell death genes showed that the expression levels of CsMC2-CsMC5 were down-regulated by drought stress [14]. Huang et al. and Liu et al. noted similar responses for MC genes in rice and rubber tree (*Hevea brasiliensis*), indicating that some MC genes may play negative regulatory roles in drought stress and it's induced programmed cell death [15,16]. The overexpression of *Arabidopsis* catalase gene *AtCAT3*, and rice *OsCatA* and *OsCatC* improved drought stress tolerance in *Arabidopsis* [17] and conferred systematic resistance in transgenic rice [18]. The genome-wide expression profiles of *Hsfs* genes in *B. napus* revealed that many members of *BnaHsf* family respond to drought stress. The

**Table 1:** A summary of some of the recently identified drought stress responsive multigene families through genome-wide analysis.

Gene family	Plant Species	Common name	No. of genes	Reference
Mitogen-activated protein kinases (MAPKs)	<i>Cassava</i>			[10]
Superoxide dismutase (SOD)	<i>Vitis vinifera</i> (Grapevine)		10	[34]
GRAS genes	<i>Citrus sinensis</i>	Sweet orange	50	[33]
BRASSINAZOLE-RESISTANT (BZR)	<i>Cajanus cajan</i>	Pigeon pea	6	[6]
	<i>Cicer arietinum</i>	Chickpea	6	
	<i>Glycine max</i>	Soybean	16	
	<i>Lotus japonicus</i> (Bridsfoot Trefoil)		5	
	<i>Medicago truncatula</i>	Barrel medic	7	
	<i>Phaseolus vulgaris</i>	Common bean	7	
	<i>Vigna radiata</i>	Mung bean	5	
	<i>Zea Mays</i>	Maize	11	[43]
Auxin Response Factors (ARFs)	<i>Cicer arietinum</i>	Chickpea	24	[11]
ALTERNATIVE OXIDASE (AOX)	<i>Triticum aestivum</i>	Hexaploid wheat	20	[40]
WRKY	<i>Ananas comosus</i>	Pineapple	54	[45]
Metacaspase (MC)	<i>Cucumis sativus</i>	Cucumber	5	[14]
cyclic nucleotide-gated channels (CNGCs)	<i>Nicotiana tabacum</i>	Common tobacco	35	[9]
	<i>Nicotiana sylvestris</i>			
	<i>Nicotiana tomentosiformis</i>			
Catalases (CATs)	<i>Brassica oleracea</i>	Cabbage, cauliflower, broccoli, Brussels Sprout, kohlrabi and kale	26	
	<i>Gossypium hirsutum</i> L.	Cotton	7	[44]
Heat stress transcription factors (Hsfs)	<i>Brassica oleracea</i>	Cabbage, cauliflower, broccoli, Brussels sprout, kohlrabi and kale	35	[42]
Expansin (EXP)	<i>Triticum aestivum</i> L.	Common bread wheat	241	[19]
NAC family	<i>T. aestivum</i> L.	Common bread wheat	361	
Multiple organellar RNA editing factor (MORF)	<i>Populus trichocarpa</i>	Black cottonwood	9	[21]
IQD (IQ67-domain)	<i>Brassica rapa ssp. pekinensis</i>	Chinese cabbage	35	[20]
Teosinte-branched 1/Cycloidea/Proliferating (TCP)	<i>Zea mays</i>	Maize	46	[41]
Aquaporins (AQPs)	<i>Citrus sinensis</i>	Sweet orange	34	[30]
	<i>Vicia sativa sub sp. sativa</i>	Common vetch	21	[31]
Calmodulin-binding Transcription Activator CAMTA	<i>Musa acuminata</i>	Banana	5	[29]
Late Embryogenesis Abundant (LEA)	<i>Vitis vinifera</i>	Grape	60	[26]
Trihelix	<i>Gossypium hirsutum</i>		102	[27]
	<i>Gossypium arboreum</i>		51	
	<i>Gossypium raimondii</i>		51	

artificially induced drought stress via PEG treatment induced greater response in wheat expansin (*TaEXPs*) genes [19] and *BrIQD5* in *B. rapa*, where *BrIQD5*-overexpressed plants showed more tolerance to drought stress than wild-type plants [20]. Wang et al. [21] identified nine drought responsive genes through expression profiling of water deficient black poplar (*Populus euramericana* cv. 'Neva'). The NAC family is the largest of the plant-specific transcription families comprising 488 genes in *Triticum aestivum* only [22]. The RNA-seq based expression data exhibited that 23 *TaNAC* genes were differentially expressed under drought stress treatment [22]. Moreover, NAC genes such as *MINAC9* (*Miscanthus lutarioriparius*) [23], *SINAC3* (*Solanum lycopersicum*) [23], *ONAC022* (*Oryza sativa*)

[24], *GmNAC3* and *GmNAC4* (*Soybean*) [25], have already been profiled as to be involved in drought stress tolerance. The microarray expression profiles of the grape LEA family genes (*VvLEAs*) exhibited strong response to drought stress [26]. Following RNA sequencing and qRT-PCR analysis, Magwanga et al. [27], overexpressed *GT2* gene from *Gossypium hirsutum* in cotton that enhanced the tolerance to maximum level. The transcriptome-wide profiles and functional analysis have also shown the regulatory roles of AQPs, CAMTAs, CNGCs in enhancing drought tolerance in rice, banana, citrus, vetches, tobacco and cabbages [28-31,7-9].

Characterization of the novel genes from genome wide analysis

have shown that the upstream regions of these genes harbored multiple drought responsive cis-acting regulatory elements i.e., eighty elements in 52 sugarcane *Expansin* genes [32]; thirty-one in orange *CsGRAS* genes [33]; four in cucumber *CsMC* genes [14]; and one in Grapevine *VvCSD6* gene [34]. The most widespread cis acting regulatory elements that are involved in drought inducibility include MYB binding sites, dehydration-responsive element (*DRE*; TACCGACAT), CRT-core motif (A/GCCGAC) and MYCATRD22 (CACATG) [35].

Additionally, studies focusing on genome-wide profiling and analysis of miRNAs have let the scientists identified numerous classes of drought stress responsive mRNAs in different crops [36]. Apart from cis-acting regulatory elements, many classes of microRNAs have been shown to be regulatory of gene expression by targeting endogenous and/or exogenous genes [37]. Growing evidence suggest that miRNA-guided regulation of ROS-related genes at the post-transcriptional level is essential for normal growth, development and adaptation to stress conditions including drought [38,39]. The novel drought responsive genes resulting from genomewide studies can serve as potential candidates for futuristic breeding programs to produce novel drought tolerant transgenic plant cultivars.

## References

- Lamaoui M, Jemo M, Datla R, Bekkaoui F. Heat and Drought Stresses in Crops and Approaches for Their Mitigation. *Frontiers in chemistry*. 2018; 6: 26.
- Daryanto S, Wang L, Jacinthe PA. Global synthesis of drought effects on maize and wheat production. *PLoS one*. 2016.
- Farooq M, Gogoi N, Barthakur S. Drought stress in grain legumes during reproduction and grain filling. *Journal of Agronomy and Crop Science*. 2017; 203: 81-102.
- Mickelbart MV, Hasegawa PM, Bailey-Serres J. Genetic mechanisms of abiotic stress tolerance that translate to crop yield stability. *Nature Reviews Genetics*. 2015; 16: 237-251.
- Muthamilarsan M, Suresh BV, Pandey G, Kumari K, Parida SK, Prasad M. Development of 5123 intron-length polymorphic markers for large-scale genotyping applications in foxtail millet. *DNA*. 2014; 21: 41-52.
- Li Y, He L, Li J, Chen J, Liu C. Genome-Wide Identification, Characterization, and Expression Profiling of the Legume BZR Transcription Factor Gene Family. *Frontiers in Plant Science*. 2018; 9: 1332.
- Kakar KU, Nawaz Z, Kakar K. Comprehensive genomic analysis of the CNGC gene family in Brassica oleracea: novel insights into synteny, structures, and transcript profiles. *BMC Genomics*. 2017; 18: 869.
- Nawaz Z, Kakar KU, Saand MA, Shu QY. Cyclic nucleotide-gated ion channel gene family in rice, identification, characterization and experimental analysis of expression response to plant hormones, biotic and abiotic stresses. *BMC Genomics*. 2014; 15: 853.
- Nawaz Z, Kakar KU, Ullah R. Genome-wide identification, evolution and expression analysis of cyclic nucleotide-gated channels in tobacco (*Nicotiana tabacum L.*). *Genomics*. 2019; 111: 142-158.
- Yan Y, Wang L, Ding Z. Genome-Wide Identification and Expression Analysis of the Mitogen-Activated Protein Kinase Gene Family in Cassava. *Frontiers in Plant Science*. 2016; 7: 1294.
- Die JV, Gil J, Millan T. Genome-wide identification of the auxin response factor gene family in *Cicer arietinum*. *BMC Genomics*. 2018; 19: 301.
- Shen C, Yue R, Sun T. Genome-wide identification and expression analysis of auxin response factor gene family in *Medicago truncatula*. *Frontiers in plant science*. 2015; 6: 73.
- Okushima Y, Overvoorde PJ, Arima K. Functional genomic analysis of the AUXIN RESPONSE FACTOR gene family members in *Arabidopsis thaliana*: unique and overlapping functions of ARF7 and ARF19. *The Plant*. 2005; 17: 444-463.
- Zhou Y, Hu L, Jiang L, Liu S. Genome-wide identification, characterization, and transcriptional analysis of the metacaspase gene family in cucumber (*Cucumis sativus*). *Genome*. 2018; 61: 187-194.
- Huang L, Zhang H, Hong Y, Liu S, Li D, Song F. Stress-responsive expression, subcellular localization and protein-protein interactions of the rice metacaspase family. *International journal of molecular sciences*. 2015; 16: 16216-16241.
- Liu H, Deng Z, Chen J, Wang S, Hao L, Li D. Genome-wide identification and expression analysis of the metacaspase gene family in *Hevea brasiliensis*. *Plant Physiol Biochem*. 2016; 105: 90-101.
- Zou JJ, Li XD, Ratnasekera D. Arabidopsis CALCIUM-DEPENDENT PROTEIN KINASE8 and CATALASE3 function in abscisic acid-mediated signaling and H<sub>2</sub>O<sub>2</sub> homeostasis in stomatal guard cells under drought stress. *The Plant Cell*. 2015; 27: 1445-1460.
- Joo J, Lee YH, Song SI. Rice CatA, CatB, and CatC are involved in environmental stress response, root growth, and photorespiration, respectively. *Journal of Plant Biology*. 2014; 57: 375-382.
- Han Z, Liu Y, Deng X. Genome-wide identification and expression analysis of expansin gene family in common wheat (*Triticum aestivum L.*). *BMC Genomics*. 2019; 20: 101.
- Yuan J, Liu T, Yu Z. Genome-wide analysis of the Chinese cabbage IQD gene family and the response of BrIQD5 in drought resistance. *Plant Mol Biol*. 2019; 99: 603-620.
- Wang D, Meng S, Su W. Genome-Wide Analysis of Multiple Organellar RNA Editing Factor Family in Poplar Reveals Evolution and Roles in Drought Stress. *International Journal of Molecular Sciences*. 2019a; 20: 1425.
- Guérin C, Roche J, Allard V, Ravel C, Mouzeyar S, Bouzidi MF. Genome-wide analysis, expansion and expression of the NAC family under drought and heat stresses in bread wheat (*T. aestivum L.*). *PLOS ONE*. 2019; 14: e0213390.
- Zhao X, Yang X, Pei S. The Miscanthus NAC transcription factor MINAC9 enhances abiotic stress tolerance in transgenic Arabidopsis. *Gene*. 2016; 586: 158-169.
- Hong Y, Zhang H, Huang L, Li D, Song F. Overexpression of a stress-responsive NAC transcription factor gene ONAC022 improves drought and salt tolerance in rice. *Frontiers in plant science*. 2016; 7: 4.
- Pinheiro GL, Marques CS, Costa MD. Complete inventory of soybean NAC transcription factors: sequence conservation and expression analysis uncover their distinct roles in stress response. *Gene*. 2009; 444: 10-23.
- Ibrahim M, Kibar U, Kazan K. Genome-wide identification of the LEA protein gene family in grapevine (*Vitis vinifera L.*). *Tree Genet Genom*. 2019; 15: 55.
- Magwanga RO, Kirungu JN, Lu P. Genome wide identification of the Trihelix transcription factors and overexpression of Gh\_A05G2067 (GT□2), a novel gene contributing to increased drought and salt stresses tolerance in cotton. *Physiol Plant*. 2019.
- Kakar KU, Nawaz Z, Cui Z. Evolutionary and expression analysis of CAMTA gene family in *Nicotiana tabacum* yielded insights into their origin, expansion and stress responses. *Scientific reports*. 2018; 8: 10322.
- Meer L, Mumtaz S, Labbo AM, Khan MJ, Sadiq I. Genome-wide identification and expression analysis of calmodulin-binding transcription activator genes in banana under drought stress. *Scientia Horticulturae*. 2019; 244: 10-14.
- Wei Q, Ma Q, Ma Z. Genome-wide identification and characterization of sweet orange (*Citrus sinensis*) aquaporin genes and their expression in two citrus cultivars differing in drought tolerance. *Tree Genet Genom*. 2019a; 15: 17.
- Wei X, Jin X, Ndayambaza B. Transcriptome-Wide Characterization and Functional Identification of the Aquaporin Gene Family During Drought Stress in Common Vetch. *DNA Cell Biol*. 2019b; 38: 374-384.
- Santiago TR, Pereira VM, De Souza WR. Genome-wide identification,

- characterization and expression profile analysis of expansins gene family in sugarcane (*Saccharum spp.*). *PLoS one*. 2018; 13: e0191081.
33. Zhang H, Mi L, Xu L, Yu C, Li C, Chen C. Genome-wide identification, characterization, interaction network and expression profile of GRAS gene family in sweet orange (*Citrus sinensis*). *Scientific Reports*. 2019; 9: 2156.
34. Hu X, Hao C, Cheng ZM, Zhong Y. Genome-Wide Identification, Characterization, and Expression Analysis of the Grapevine Superoxide Dismutase (SOD) Family. *International Journal of Genomics*. 2019; 13.
35. Zhu X, Huang C, Zhang L. Systematic Analysis of Hsf Family Genes in the Brassica napus Genome Reveals Novel Responses to Heat, Drought and High CO<sub>2</sub> Stresses. *Frontiers in Plant Science*. 2017; 8.
36. Zhou L, Liu Y, Liu Z, Kong D, Duan M, Luo L. Genome-wide identification and analysis of drought-responsive microRNAs in *Oryza sativa*. *J Exp Bot*. 2010; 61: 4157-4168.
37. Wang W, Liu D, Zhang X, Chen D, Cheng Y, Shen F. Plant microRNAs in cross-kingdom regulation of gene expression. *International journal of molecular sciences*. 2018; 19: 2007.
38. Zhang Y, Wang W, Chen J, Liu J, Xia M, Shen F. Identification of miRNAs and their targets in cotton inoculated with *Verticillium dahliae* by high-throughput sequencing and degradome analysis. *International journal of molecular sciences*. 2015; 16: 14749-14768.
39. Ma C, Burd S, Lers A. mi R 408 is involved in abiotic stress responses in *A. rabiidopsis*. *The Plant Journal*. 2015; 84: 169-187.
40. Brew-Appiah RA, York ZB, Krishnan V, Roalson EH, Sanguinet KA. Genome-wide identification and analysis of the ALTERNATIVE OXIDASE gene family in diploid and hexaploid wheat. *PLoS one*. 2018; 13: e0201439.
41. Ding S, Cai Z, Du H, Wang H. Genome-Wide Analysis of TCP Family Genes in *Zea mays* L. Identified a Role for ZmTCP42 in Drought Tolerance. *International Journal of Molecular Sciences*. 2019; 20: 2762.
42. Lohani N, Golicz AA, Singh MB, Bhalla PL. Genome-wide analysis of the Hsf gene family in *Brassica oleracea* and a comparative analysis of the Hsf gene family in *B. oleracea*, *B. rapa* and *B. napus*. *Functional & Integrative Genomics*. 2019; 19: 515-531.
43. Manoli A, Trevisan S, Quaggiotti S, Varotto S. Identification and characterization of the BZR transcription factor family and its expression in response to abiotic stresses in *Zea mays* L. *Plant growth regulation*. 2018; 84: 423-436.
44. Wang W, Cheng Y, Chen D. The Catalase Gene Family in Cotton: Genome-Wide Characterization and Bioinformatics Analysis. *Cells*. 2019b; 8: 86.
45. Xie T, Chen C, Li C, Liu J, Liu C, He Y. Genome-wide investigation of WRKY gene family in pineapple: evolution and expression profiles during development and stress. *BMC Genomics*. 2018; 19: 490.