R2R3 MYB Transcription Factor, AhMYB94 Plays a Crucial Role in Stress adaptation of a Salt Susceptible Groundnut Cultivar-K6

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Morpho-physiological responses and expression analysis of MYB94 transcription factor gene in four groundnut cultivars under drought and salinity stress

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Abstract

Aim: The present study aimed to evaluate the impact of salinity stress on morpho-physiological parameters and relative expression of the MYB94 transcription factor gene among four widely cultivated groundnut cultivars.

Methodology: A comparative analysis of morpho-physiological parameters was studied in four groundnut cultivars (Kadiri-Amaravati, Kadiri-Lepakshi, Kadiri-6, and Kadiri-9) subjected to different concentrations of NaCl prepared in half-strength Arnon's nutrient growth medium. The relative expression analysis of a stress responsive transcription factor gene, MYB94 was studied in four $ground nut \, cultivars \, subjected \, to \, drought \, and \, NaCl \, stress \, by \, qRTPCR.$

Results: NaCl stress resulted in a significant increase in proline content, MDA content in cultivar Kadiri-9 compared to other cultivar studied. On the contrary, plant growth, shoot dry weight, root dry weight, leaf relative water content, cell membrane injury decreased to lesser extent in cultivar Kadiri-9 than Kadiri-Amaravati. Kadiri-Lepakshi. Kadiri-6 under salinity stress. qRT-PCR based relative expression analysis of the MYB94 gene revealed a significant increase of transcript expression levels in cultivar Kadiri-9 compared to other three cultivars under both drought and salinity stress.

Interpretation: Groundnut cultivar Kadiri-9 showed better tolerance to NaCl stress as evidenced from enhanced proline content, better relative water content and lesser extent of membrane damage when compared with other groundnut cultivars under given NaCl stress treatments. In addition. relative expression of MYB94 gene was significantly higher due to NaCl stress as compared to drought stress conditions in cultivar Kadiri-9 suggested that the MYB94 could be involved in salt stress tolerance.

Key words: Groundnut, MYB94 transcription factor, qRT-PCR, Salinity stress

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Effect of salinity stress and MYB94 expression analysis in four groundnut cultivars Expression analysis Comparative analysis Total RNA isolation Plant growth and Biomass, Synthesis of cDNA Relative water content, Proline content, Lipid peroxidation assay Cell membrane stability Groundnut cultivar Kadiri-9 showed high MYB94 gene expression levels and also tolerance towards salinity stress

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Co-expression of stress-responsive regulatory genes, MuNAC4, MuWRKY3 and MuMYB96 associated with resistant-traits improves drought adaptation in transgenic groundnut (Arachis hypogaea I.) plants

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Groundnut, cultivated under rain-fed conditions is prone to yield losses due to intermittent drought stress. Drought tolerance is a complex phenomenon and multiple gene expression required to maintain the cellular tolerance. Transcription factors (TFs) regulate many functional genes involved in tolerance mechanisms. In this study, three stress-responsive regulatory TFs cloned from horse gram, (Macrotyloma uniflorum (Lam) Verdc.), MuMYB96, involved in cuticular wax biosynthesis; MuWRKY3, associated with anti-oxidant defense mechanism and MuNAC4, tangled with lateral root development were simultaneously expressed to enhance drought stress resistance in groundnut (Arachis hypogaea L.). The multigene transgenic groundnut lines showed reduced ROS production, membrane damage, and increased superoxide dismutase (SOD) and ascorbate peroxidase (APX) enzyme activity, evidencing improved antioxidative defense mechanisms under drought stress. Multigene transgenic plants showed lower proline content, increased soluble sugars, epicuticular wax content and higher relative water content suggesting higher maintenance of tissue water status compared to wildype and mock plants. The scanning electron microscopy (SEM) analysis showed a substantial increase in deposition of cuticular waxes and variation in stomatal number in multigene transgenic lines compared to wild type and mock plants. The multigene transgenic plants showed increased growth of lateral roots, chlorophyll

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De novo Transcriptome Analysis of **Drought-Adapted Cluster Bean** (Cultivar RGC-1025) Reveals the Wax **Regulatory Genes Involved in Drought Resistance**

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Jagadeesh Kumar N. Venkatesh B. Javamma N. Pandurangaiah M and Sudhakar C (2022) De novo Transcriptome Analysis of Drought-Adapted Cluster Bean (Cultivar RGC-1025) Reveals the Wax Regulatory Genes Involved in Drought Resistance. Front. Plant Sci. 13:868142.

Reddy BM, Anthony Johnson AM.

Cluster bean (Cyamopsis tetragonoloba L.) is one of the multipurpose underexplored crops grown as green vegetable and for gum production in dryland areas. Cluster bean is known as relatively tolerant to drought and salinity stress. To elucidate the molecular mechanisms involved in the drought tolerance of cluster bean cultivar RGC-1025, RNA sequencing (RNA-seq) of the drought-stressed and control samples was performed. De novo assembly of the reads resulted in 66,838 transcripts involving 203 pathways. Among these transcripts, differentially expressed gene (DEG) analysis resulted in some of the drought-responsive genes expressing alpha dioxygenase 2, low temperatureinduced 65 kDa protein (LDI65), putative vacuolar amino acid transporter, and late embryogenesis abundant protein (LEA 3). The analysis also reported drought-responsive transcription factors (TFs). such as NAC, WRKY, GRAS, and MYB families. The relative expression of genes by qRT-PCR revealed consistency with the DEG analysis. Key genes involved in the wax biosynthesis pathway were mapped using the DEG data analysis. These results were positively correlated with epicuticular wax content and the wax depositions on the leaf surfaces, as evidenced by scanning electron microscope (SEM) image analysis. Further, these findings support the fact that enhanced wax deposits on the leaf surface had played a crucial role in combating the drought stress in cluster beans under drought stress conditions. In addition, this study provided a set of unknown genes and TFs that could be a source of engineering tolerance against drought stress in cluster beans.

Keywords: drought stress, transcriptome, wax genes, cluster bean (Cyamopsis tetragonoloba L.), differentailly expressed genes

INTRODUCTION

Cyamopsis tetragonoloba (L.) Taub. (Cluster bean) is a drought-adapted annual legume crop with lower water requirements than many other dryland legume crops. Cluster beans can grow in marginal soils because of their high water use efficiency, deep tap rooting system, etc. In India, cluster bean is cultivated for its green vegetables, foraging cattle, green manure, and dry pods for

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