Expression pattern analysis of transcription factors from *Aeluropus littoralis* in response to salt stress and recovery condition

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**ABSTRACT:** Salinity is one of the most important abiotic stresses that decrease crop production. Halophyte plant can tolerate high sodium chloride (NaCl) concentration and transcription factors (TFs) are prominent regulators in plant response to stress. In this study, the expression pattern of four salt-induced genes encoding transcription factors in *Aeluropus littoralis*, namely, *MYB*, *RF2*, *GTF*, and *ARID* was studied. Plants were exposed to salt stress (600 mM) and recovery condition and samples were collected at three timepoints (6, 12 and 24 hours). The results of quantitative real-time PCR (qPCR) showed that the effect of NaCl was statistically significant on genes expression. The expression level was increased after 6 hours in all genes and after that, it drastically decreased with promoting of stress duration in both tissues. The highest expression was observed for *MYB* gene in root (68.44) that was higher than shoot (38.57) after 6 hours. At the recovery condition, the genes expression was gradually decreased in different tissues and finally got a stable value. The result showed that the studied TFs play an important role in tolerance of *A. littoralis* to salinity and could be used as an informative resource in the breeding programs. Also, the response of *A. littoralis* to salt stress depends on the tissue type and plant exposure duration.

**KEYWORDS:** Abiotic stress, Halophyte, Recovery condition, Reference genes, Transcription factor.

**INTRODUCTION**

Salt stress is one of the most important problems in agriculture [1]. Approximately 25% of the world’s total area (including 15% of Iran’s area) has this problem [2]. Both grain yield and quality are downgraded by high salinity. Thus, improving level of abiotic stress tolerance has become a key objective in many crop breeding programs. Genetic variation for stress tolerance is rather limited in the domesticated gene pool, but a number of wild plant of Poaceae family have been proved to process notable high-level tolerance *Aeluropus littoralis*, a halophyte plant, belongs to Poaceae that can tolerate NaCl up to 600 mM [3] and grows in marshes, salty and drought lands. C4 photosynthesis system in *A. littoralis* is one of the reasons for being more flexible against salt and drought stresses. Therefore, *A. littoralis* may know as a great source of genes related to salt tolerance [4, 5]. Nowadays, stress responses understanding is one of the most important issues in plant research. Plants response
to abiotic stress with the change in many functional and structural mechanisms by altered metabolic pathways. Stress tolerance in plants can be enhanced via both ABA-dependent and ABA-independent pathways [6]. Various plant transcription factors (TFs) are up- or down-regulated in response to abiotic stress [7], and the response is typically quite rapid [8, 9]. TFs generally act as key regulators of gene expression and about 7% of the genes encoding the TFs in plants are specified to stress or defense responses. [10]. These TFs involve in responses to stress by encoding proteins with specific functions [11] and some of theme which plays the important roles in signaling and regulatory pathway could be used in genetic engineering [12]. Recently, the roles of some TF families in transgenic plants such as Arabidopsis, rice, maize and tobacco in abiotic stress have been reported in some literatures [13-21]. Most MYB proteins have domain repeats that able to bind DNA. In plants, most of the MYB proteins are involved in the responses to regulation mechanisms, such as abiotic and biotic stress [22], hormone signaling [23] and regulation of differentiation [24]. AtMYB60 and AtMYB96 are implicated in both biotic and abiotic stresses [25, 26]. AtMYB2 controls the ABA induction of salt and dehydration responsive genes [23]. RF2a is a bZIP transcription factor and control the promoter activity of rice in biotic stress. Studies in rice indicated that RF2a found in vascular tissues, leaves and stems cells, but in roots, the level of RF2a is very low [27]. bZIP proteins have been identified in all eukaryotes, including yeasts, vertebrates, and plants [28]. A family of transcription factors with basic region and Leu-zipper motif that defined by the conserved bZIP domain [29]. ABA activates bZIP proteins to bind to ABRE and initiate transcription of ABA-inducible genes [30] and modulates the signaling networks for a number of hormones and regulate the biosynthesis of starches, storage proteins and lipids in response to biotic and abiotic stresses [31]. The cells that respond to environmental changes typically induce the expression of different genes.

According to the crucial role of TFs in response to environmental stress and regulation of gene expression and the importance of A. littoralis as a salt tolerant wild member of Poaceae, this study was carried out to assess the expression pattern of four salt-induced genes encoding transcription factors. Moreover, the effect of different plant tissues (root and shoot) on gene expression was investigated.

### MATERIALS AND METHODS

#### Plant material

*A. littoralis* seeds were collected from Isfahan province and the sterilized seeds plated on full strength MS medium (pH 5.8). After germination, two weeks’ seedlings transferred to pots which containing Hoagland’s solution [32]. The 30 day-old seedlings were exposed to 500 mM of sodium chloride at five passages (received 100 mM sodium chloride per 48 hours (h) up to 500 mM) [33]. Leaves and roots were sampled in parallel. At the end of the fifth passage, samples were collected at 6, 12 and 24 h time-course. In order to plant recovery, the remained plants were transferred to a sodium chloride-free Hoagland’s solution, and then were collected after 6, 12 and 24 h. All samples and control plant were immediately frozen in liquid nitrogen and stored at -70 °C.

#### RNA extraction, cDNA synthesis, and qPCR

Shoot and root tissues were grounded in liquid nitrogen and total RNA was extracted using Trizol reagent (Invitrogen) according to the manufacturer’s protocol and treated with RNase-free DNaseI (Promega). The RNA quality and quantity were controlled using agarose gel electrophoresis and spectrophotometry, respectively. First strand cDNA synthesis and qPCR were performed according to Fermentas kit instructions. The cDNA used as a template for subsequent PCR. The ACTIN gene was used as internal reference gene and to calculate relative expression levels of specific genes. The studied genes were selected based on the search on NCBI databases (https://www.ncbi.nlm.nih.gov/) of *A. littoralis* and then they isolated from the extracted cDNAs through the specific primers designed by Primer3 and Oligo Analyzer 1.2 soft wares [34].

The quantitative PCR was performed in a 10-μl reaction containing SYBR Green I PCR Master Mix (Thermo Scientific, USA), 10 pmol each of forward and reverse gene-specific primers and 2 μl of diluted cDNA (1:9). PCR amplification was performed in C1000™ Thermal Cycler (Bio-Rad, USA) according to the company’s suggestions. The qPCR program was as follows: 95°C for 5 min, 15 S of denaturation, 30 S of annealing (depending on primer used) (Table 1) and 15 S of extension at 72°C for 40 cycles and a final extension at 72°C for 7 min(s). Melt curve analysis was performed to check the specificity of the amplified product. To minimize sample
Table 1. Sequences of primers used for real-time PCR amplification

<table>
<thead>
<tr>
<th>Gene</th>
<th>Description</th>
<th>Primer Sequence</th>
<th>Accession Number</th>
<th>Amplicon Size</th>
<th>Tm</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACT11</td>
<td>Actin-11</td>
<td>5'-GTATGGCAACATTGTCCCTCAG-3'</td>
<td>EE594539.1</td>
<td>118</td>
<td>59.4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5'-TGGACACGACCGTATGAGAT-3'</td>
<td></td>
<td></td>
<td>58.5</td>
</tr>
<tr>
<td>ARID</td>
<td>AT Rich Interactive Domain</td>
<td>5'-TCGCCATGCTTTGATGTCG-3'</td>
<td>JK671186</td>
<td>90</td>
<td>58.7</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5'-ATAGCCGGAAGGATGCATG-3'</td>
<td></td>
<td></td>
<td>58.4</td>
</tr>
<tr>
<td>MYB</td>
<td>Myeloblastosis</td>
<td>5'-GGAGGCGTACATGGAGGTG-3'</td>
<td>EF534705</td>
<td>166</td>
<td>60.1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5'-CGCCACCTTTCTCCAGTCC-3'</td>
<td></td>
<td></td>
<td>59.8</td>
</tr>
<tr>
<td>RF2</td>
<td>Transcription Factor RF2a-like</td>
<td>5'-GACATCACACCCCCAGAAG-3'</td>
<td>EE594580</td>
<td>184</td>
<td>58.85</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5'-TCACCTAGACACTGCGTACG-3'</td>
<td></td>
<td></td>
<td>58.95</td>
</tr>
<tr>
<td>GTF</td>
<td>General Transcription Factor 3C Polypeptide</td>
<td>5'-TTCAAGTGGCATCGGT-3'</td>
<td>JZ191082.1</td>
<td>108</td>
<td>60</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5'-AAAGGCGTCCCTGCGTCTT-3'</td>
<td></td>
<td></td>
<td>60.5</td>
</tr>
</tbody>
</table>

variation, the samples were normalized using actin expression mRNA expression. To reveal the absence of contamination or primer-dimers, a non-template control (NTC) reaction with each primer pair was run. The $2^{-\Delta\Delta CT}$ method was used for the quantitative analysis [35]. Data were analyzed by Microsoft Excel 2010. Following analysis of variance (ANOVA), Duncan’s multiple range tests was used to assess the significance of differences between mean values.

RESULTS

Salinity is an abiotic stress that causes various detrimental effects on plant growth and development [36]. The expression of 4 salt-induced genes encoding transcription factors, namely, MYB, RF2, GTF and ARID were screened in this study using qPCR method. The results displayed different expression patterns in A. littoralis under salt stress in different tissues and the significant effect of salinity on genes expression.

Expression pattern of MYB gene under salt stress

The result of expression analysis showed that the MYB gene expression was induced significantly by salt treatment in 6 h after stress but drastically decreased with promoting of stress duration in both root and shoot tissues, which reached to the lowest level of expression after 24 h after salt stress treatment (Fig 1). This expression in roots was higher than shoots. The Arabidopsis MYB transcription factor, AtMYB44, behaves in a very similar manner [37]. In the case of recovery, the MYB genes expression increased to 2.55, 5.2 and 5.92 folds in the shoot and 2.03, 7.97 and 3.66 folds in root respectively in comparison to control plants in a non-significant manner while the expression decreased significantly in both of tissues in comparison to stress condition (table 2 and table 3).

Expression pattern of RF2 gene under salt stress

Comparison of RF2 expression patterns in the different times showed significant differences in gene expression rates. The increasing in RF2 transcript levels was observed in shoots and roots of salt treated plants (Figure 2). After 6 h salt stress, the expression levels in shoots was in maximum (5.16-fold higher than control) and then decreased in the next times of exposure to stress treatment (table 2). In roots, the expression levels increased significantly until 12 h (2.04-fold higher than control plants) and then decreased in 24 h after salt stress (table 3). The result showed that the mRNA abundance of RF2 in shoots was higher than roots in all treatments. At the recovery stage (return of plants to non-stress condition), the expression of RF2 gene was gradually decreased compared to treated plants until reached to lowest value expression in both root and shoot samples in 24 h after recovery. This expression difference was significant statistically in shoots after 6 and 12 h recovery than control (table 2) but not significant with expression after 24 h recovery.

Expression pattern of GTF gene under salt stress

Sequence analysis indicated that JZ191082.1 EST refers to general transcription factor 3C polypeptide 5. As shown in Fig 3, the GTF expression in treated plant was induced by treatment with 500 mM NaCl. In the first 6 h,
**Table 2.** The means comparison of genes expression in shoot tissue

<table>
<thead>
<tr>
<th></th>
<th>cntr</th>
<th>6 h</th>
<th>12 h</th>
<th>24 h</th>
<th>6 h rec</th>
<th>12 h rec</th>
<th>24 h rec</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MYB</strong></td>
<td>1.0 c</td>
<td>38.57 a</td>
<td>9.94 b</td>
<td>5.73 bc</td>
<td>2.55 c</td>
<td>5.20 bc</td>
<td>5.92 bc</td>
</tr>
<tr>
<td><strong>RF2</strong></td>
<td>1.0 c</td>
<td>5.16 a</td>
<td>4.53 a</td>
<td>2.87 b</td>
<td>2.59 b</td>
<td>2.32 b</td>
<td>2.11 bc</td>
</tr>
<tr>
<td><strong>GTF</strong></td>
<td>1.0 b</td>
<td>3.85 a</td>
<td>2.14 b</td>
<td>1.76 b</td>
<td>1.83 b</td>
<td>1.23 b</td>
<td>1.38 b</td>
</tr>
<tr>
<td><strong>ARID</strong></td>
<td>1.0 d</td>
<td>5.93 a</td>
<td>4.91 ab</td>
<td>5.17 a</td>
<td>2.07 cd</td>
<td>3.04 bc</td>
<td>2.92 c</td>
</tr>
</tbody>
</table>

Control (Cntr), stress after 6 h, 12 h, 24 h, stress after 6 h recovery, 12 h recovery and 24 h recovery, respectively. Means with the same letters are not significantly different. (P > 0.05).

**Table 3.** The means comparison of genes expression in root tissue

<table>
<thead>
<tr>
<th></th>
<th>cntr</th>
<th>6 h</th>
<th>12 h</th>
<th>24 h</th>
<th>6 h rec</th>
<th>12 h rec</th>
<th>24 h rec</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MYB</strong></td>
<td>1.0 c</td>
<td>68.44 a</td>
<td>18.58 b</td>
<td>3.79 c</td>
<td>2.03 c</td>
<td>7.97 bc</td>
<td>3.66 c</td>
</tr>
<tr>
<td><strong>RF2</strong></td>
<td>1.0 bc</td>
<td>1.45 b</td>
<td>2.04 a</td>
<td>1.16 bc</td>
<td>0.85 c</td>
<td>1.07 bc</td>
<td>0.87 c</td>
</tr>
<tr>
<td><strong>GTF</strong></td>
<td>1.0 a</td>
<td>0.98 a</td>
<td>0.58 a</td>
<td>0.82 a</td>
<td>0.89 a</td>
<td>1.18 a</td>
<td>1.18 a</td>
</tr>
<tr>
<td><strong>ARID</strong></td>
<td>1.0 b</td>
<td>0.83 b</td>
<td>0.26 b</td>
<td>0.56 b</td>
<td>2.49 a</td>
<td>1.24 b</td>
<td>2.66 a</td>
</tr>
</tbody>
</table>

Control (Cntr), stress after 6 h, 12 h, 24 h, stress after 6 h recovery, 12 h recovery and 24 h recovery, respectively. Means with the same letters are not significantly different. (P > 0.05).

there was a drastic increase in expression compared with the control in the shoot (3.85 folds) but the expression was significantly decreased 12 and 24 h after salt stress treatment (table 2). In root, transcript levels were decreased gradually to 0.98, 0.58 and 0.82 folds in response to 500 mM NaCl stress after 6, 12 and 24 h, respectively and the differences was not significant statistically (table 3). In recovery mode, changes in gene expression were different. In the shoot, GTF expression was decreased while in the root, expression of GTF increased in 6, 12 and 24 h after salt stress treatment, so that the expression in both tissues was closed to the expression level of control and in a non-significant manner. The result showed that GTF gene expression in shoots was higher than roots in all hours.

**Expression pattern of ARID under salt stress**

Bioinformatic analysis in NCBI database predicted that JK671186 EST refers to AT-Rich Interaction Domain protein. ARID proteins have an ancient DNA-binding domain that is conserved throughout higher eukaryotes and as a transcription factor, have a vital role in cell cycle and division [38]. The results indicated that ARID was highly expressed in shoots and the transcript levels of ARID were induced significantly by NaCl treatment (table 2). The expression levels of ARID in shoots increased 5.93, 4.91 and 5.17-fold relative to the control at 500 mM NaCl concentrations after 6, 12 and 24 h, respectively, while the level of transcripts in roots was 0.83, 0.26 and 0.56-fold lower than non-stressed controls (Fig 4) and the differences were not significant than control (table 3). Generally, higher transcript levels of ARID were detected in shoots compared to roots in all treatments (Fig. 4). The 5.93-fold expression of ARID in stress condition rather than non-stress one in the shoot, indicating a positive regulation role of ARID in salt stress. These results indicated that ARID may be involved in responding to salt stress in the plant. In recovery condition compared to stress one, ARID expression was increased significantly in roots and was roughly equal to gene expression in shoot tissue (Fig 4).

**DISCUSSION**

Plants have developed various strategies to respond to unfavorable environmental conditions [39]. To improve salt tolerant in plants, a proper understanding of molecular mechanisms and the gene expression of different factors that affect growth and developmental stage is essential. One of the most common used techniques for gene expression analysis is qPCR and has very advantages such as specificity, sensitivity, reproducibility, and accuracy. In this study, expression of genes was significantly induced by salt stress and reached
Figure 1. Expression pattern of MYB gene using q-PCR under salt stress and recovery condition in A. littoralis. Control (Cntr), stress after 6 h, 12 h, 24 h, stress after 6 h recovery, 12 h recovery and 24 h recovery, respectively. Values are Means and bars indicate Standard Error (SE).

Figure 2. Expression pattern of RF2 gene using q-PCR under salt stress and recovery condition in A. littoralis. Cntr, stress after 6 h, stress after 12 h, stress after 24 h, stress after 6 hours’ recovery, stress after 12 h recovery and stress after 24 h recovery, respectively. Values are Means and bars indicate SE.

Figure 3. Expression pattern of GTF gene using q-PCR under salt stress and recovery condition in A. littoralis. Cntr, stress after 6 h, 12 h, 24 h, stress after 6 h recovery, 12 h recovery and 24 h recovery, respectively. Values are Means and bars indicate SE.

Figure 4. Expression pattern of ARID gene using q-PCR under salt stress and recovery condition in A. littoralis. Cntr, stress after 6 h, 12 h, 24 h, stress after 6 h recovery, 12 h recovery and 24 h recovery, respectively. Values are Means and bars indicate SE.
the highest peaks at 6 h after salt stress. MYB transcription factors play important roles in plant growth and stress responses. The highest transcript level of MYB in *A. littoralis* was nearly 68-fold over that in the non-treated control, indicating a positive regulation role of MYB in salt stress. Most MYB genes were positive and negative regulators of transcription [40] and play important roles in the regulation of many secondary metabolites at the transcriptional level [41]. MYB genes are involved in the signal transduction pathways of salicylic acid [42], abscisic acid (ABA) [23], gibberellic acid [43] and jasmonic acid [44] as well. The phytohormone ABA, produced under water deficit conditions, caused stomatal closure and played an important role in the adaptation of vegetative tissues to abiotic environmental stresses, such as drought and high salinity [45, 46]. Recently, the role of AtMYB44 and AtMYB15 Over-expression in stress tolerance [37, 47], TaMYB2A role in making Arabidopsis tolerance to multiple abiotic stresses [48], AtMYB2 role in dehydration and salt stress tolerance at the transcriptional level [49] and TaMYBsd1 role in salt tolerance in wheat cultivars [50] were reported. In this study, transcriptional levels of *MYB* in the shoot and root of *A. littoralis* plants significantly increased in response to salinity after 6 h of salt stress which was consistent with the report that Tamyb2 was rapidly induced under osmotic stress for 1 h [51]. The result indicated the prominent roles of *MYB* family in quick response to stresses in plants as well as other TFs which regulated rapidly in response to biotic stress [7]. Because the roots are the first area which sense salt stress so the first response should occur in the root rather than the leaf. Furthermore, some MYB-encoding genes showed preferential accumulation of transcripts in tissues or under a specific condition. For example, some MYB genes in *Gossypium raimondii* were mainly expressed in leaves and roots under drought and saline conditions. A number of *GrMYB* genes such as *GrMYB050* and *GrMYB074* strongly and preferentially expressed in leaves under drought stress, whereas some of them were highly expressed in roots under drought and salt stress [52]. By contrast of *MYB*, in *RF2* gene expression pattern, whereas gene expression was increased in both tissue but it was in shoot higher than root and the peak level in the shoot was at 6 h and in root in 12 h after stress. *RF2a* was found in different tissues such as leave and stem in high level and low level in the root. *RF2* plays a critical role in leaf tissue differentiation and reducing the amount of it in transgenic rice plants severely disrupted shoot development. *RF2a* is a bZIP transcription factor that affects shoot development in *Oryza sativa*. With reducing of *RF2a* levels in transgenic rice line, some characteristics, such as severe stunting of shoots and leaf twisting and aberrant tissue organization were presented [53]. The RF2a protein has a bZIP domain which is similar to the domain from Arabidopsis, tobacco, tomato, and other plants [54]. Different studies indicated that the plants bZIP factors are vital regulators of plant specific processes including pathogen defense, light and stress signaling [55]. This group of bZIP proteins is involved in regulating gene expression in vascular tissues and shoot tissue organization in rice [27] and involved in ABA signaling pathway and Acts as positive regulator of the expression of abiotic stress-responsive genes through an ABA-dependent signaling pathway [56]. It Plays an important role in ABA and auxin responses by directly binding to the ABA-responsive element (ABRE)-containing genes, especially WRKY family genes. Suppresses auxin signaling by targeting ABRE-containing genes related to auxin metabolism or signaling [57]. Consistent with these studies, the expression of bZIP in Arabidopsis [58] and Rice [31] validated the role of bZIP in response to abiotic stresses. Some bZIP TFs such OsbZIP50 and OsbZIP39 are transcription factors that are involved in the activation of numerous chaperone genes [59, 60] and someone such an OsbZIP46 may be activated by post translational phosphorylation modification and Improves abiotic stress tolerance [61]. Although, the gene expression of *GTF* and *ARID* was increased in shoot tissue under 500 mM NaCl, but the expression level of these genes was decreased in root compared to the control. Recently reported that in *A. littoralis*, *GTF* expression level in leaf was higher and more stable than root tissue [62]. *GTFs* are involved in transcription initiation step and have the main role in RNA polymerase II regulation. Recent studies indicated that early event in the activation of genes is the binding of GTFs and Pol II to enhancer elements [63] and form the TATA-box complex [64] that involves in assembling of general transcription factors. Tfg3 (a small subunit of TFII1 general transcription factor) of *Schizosaccharomyces pombe* was characterized and proposed that Tfg3 had an important role in high temperatures stress as a gene expression regulator [65]. Some ARID protein members are expressed ubiquitously and others in a highly tissue-specific pattern [66] and exhibited some cellular functions, including transcriptional regulation, participation in cell differentiation and development,
embryonic development and cell lineage [67]. The expression of GTF and ARID genes in recovery stage reached nearly or higher than to the control. Furthermore, their expression level in the shoot was higher than root. Similar results have been reported about the role of transcription factor by other researchers in plants under salt stress [68-71, 47]. This expression pattern in A. littoralis was indicated that timing and tissue type is an important factor in stress-related gene response in salt stress condition. The results showed that when plants were transferred to a NaCl-free Hoagland’s solution after salt stress duration as a recovery, the expression of up-regulated genes under salt stress was decreased gradually in different tissues and finally depending on the function of genes such as plant growth, cell cycle and division, developmental processes and et cetera was placed in a constant value. Quantitative differences in the expression of genes involved in tolerance induction during abiotic stress could also be the result of the timing and intensity of transcriptional activation of involved genes rather than their presence/absence or other mechanisms [72]. Therefore, to make a plant with multiple resistances to abiotic stresses, it’s important to increase the capability of genetical control of one or several key regulatory factor(s) [73]. Furthermore, considering transcription factors as candidate genes in breeding programs will give us a clear understanding the net of abiotic stress events and will finally lead us to improve crop varieties with higher stress tolerance. In the present study, the results showed that in A. littoralis the expression patterns of genes encoding transcription factors changes in response to salt stress and it is depending on the tissue type and treatment exposure duration. So it could be mentioned in designing breeding and biotechnology programs of salt tolerance plants.

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بررسی الگوی بیان فاکتورهای روتوپسی در شرایط تنش شوری و محيط بازیابی در گیاه هالوفیت

Aeluropus littoralis

چکیده

تنش شوری یکی از مهم‌ترین شرایط غیر روتینی است که رشد و تولید محصولات زراعی را محدود می‌کند. گیاهان هالوفیت غلظت های بالای کلرید سدیم را تحمل کرده و فاکتورهای روتوپسی، عوامل تنظیم کننده مهمی در پاسخ گیاه به تنش ها می‌باشند. در این تحقیق، الگوی بینایی آن که کننده فاکتورهای روتوپسی در گیاه آلوروپوس لیتورالیس با نام‌های MYB، RF2، GTF و ARID در پاسخ به تنها تنش شوری بررسی شد. به‌دست آمده که گیاهان در معرض تنش شوری ۶۰۰ میلی‌مولار کلرید روتوپسی قرار گرفته و نمونه برداری از گیاهان در زمان‌های ۶، ۱۲ و ۲۴ ساعت انجام گرفت. نتایج داده‌های حاصل از qPCR نشان داد که کلرید سدیم تأثیر معنی‌داری از لحاظ آماری بر اندازه‌بندی تولید پرورشی و افزایش زمان تنش میزان آن در بافت‌های ریشه و اندازه‌بندی کاهش یافته بالاترین میزان این پرورش در ریشه‌های گیاه (۶۷/۴۴) در ۶ ساعت ابتدا و در ریشه‌های گیاه (۶۷/۴۴) مشاهده شد که بیشتر از بین‌رود در اندازه‌بندی گیاه (۶۷/۴۴) بود. در شرایط روتوپسی، گیاه زن‌ها در پاسخ‌های مختلف به تغییر کاهش و زمان‌های نهایی به مقدار ثانوی سریال. نتایج این تحقیق نشان داد که فاکتورهای روتوپسی مورد مطالعه نقش مهمی در تحمل گیاه آلورپوس لیتورالیس به تنش شوری داشته و می‌تواند به عنوان منبع اطلاعاتی مهمی در برنامه‌های اصلاحی معرفی شوند. همچنین، پاسخ گیاه آلورپوس لیتورالیس به تنش شوری به نوع بافت و مدت زمان در معرض تنش پردیدن گیاه نیز بستگی دارد.

کلمات کلیدی: آلوروپوس لیتورالیس، تنش شوری، زن‌مرجع، فاکتور روتوپسی، محيط روتوپسی