Insight of Transcriptional Regulators reveals the Tolerance Mechanism of Carpet-grass (*Axonopus compressus*) Against Drought

Mohsin Nawaz
Hainan University
Liao Li
Hainan University
Farrukh Azeem
Government College University Faisalabad
Samina Shabbir
Biogas Institute of Ministry of Agriculture
Ali Zohaib
adaptive research station gujjranwala
Umair Ashraf
University of Education Faisalabad Campus
Zhiyong Wang (wangzhiyong@hainanu.edu.cn)
Hainan University
Yang Hubiao
Chinese Academy of Tropical Agricultural Sciences

Research article

Keywords: Drought, Axonopus compressus, qRT-PCR, Enrichment analysis, lncRNAs, miRNAs, Transcriptomics

Posted Date: July 23rd, 2020

DOI: https://doi.org/10.21203/rs.3.rs-41830/v1

Version of Record: A version of this preprint was published at BMC Plant Biology on February 2nd, 2021. See the published version at https://doi.org/10.1186/s12870-021-02844-7.

Abstract

Highlights:

1. *Axonopus compressus* can stand severe drought stress by activating the potential defense mechanism.

2. We investigated the differential transcriptome of drought stressed and normal *Axonopus compressus* plants

3. New comers have been identified that involved in the drought response

4. Identified drought responsive genes which never known for other stresses.

5. The identified genes also respond to stress in *Arabidopsis thaliana* in different manners.

Background: Carpet grass [*Axonopus compressus* (L.)] is an important warm season perennial grass around the world and is renowned for its adaptability to varied environmental conditions. However, Carpet grass lacks enough data in public data bank, which confined our comprehension of the mechanism of environmental adaptations, gene discovery and development of molecular marker.

Methods: In current study, the DEGs (differentially expressed genes) in *Axonopus compressus* under drought stress (DS) were identified and compared with CK (control) by RNA-Seq.

Results: the 263,835 of total unigenes were identified in *Axonopus compressus*, and 201,303 (also add the numbers of remaining2 data bases) a sequence of unigenes significantly matched in at least one of the seven databases. A total of 153697 (58.25%) unigenes can be classified to 144 KEGG pathways, and 7,444 unigenes were expressed differentially between DS and CK, of which 4,249 were up-regulated and 3,195 were down-regulated unigenes. Of the 50 significantly enriched GO terms, 18, 6 and 14 items were related to BP, CC and MF, respectively. Analysis of KEGG enrichment showed 2569 DEGs involved in 143 different pathways, under drought stress 2,747 DEGs were up-regulated and 2,502 DEGs were down-regulated. Moreover, we identified 352 transcriptor factors (TFs) in *Axonopus compressus*, of which 270 were differentially expressed between CK and DS. The qRT-PCR validation experiment also support the transcriptional response of *Axonopus compressus* against drought.

Conclusions: The current findings provide the first framework for further investigation for the particular roles of these unigenes in *Axonopus compressus* in response to droughts.

Background

Axonopus is a genus of the grass family (Paniceae; Poaceae; Triticeae). Approximately 100 species of Axonopus have already been determined [1], and they are generally distributed along shoreline of the China and Africa, Alaska, central Asia, eastern Asia, North Sea, South America and Oceania [2, 3]. There are about more than 40% (55.5×106 km2) of Earth's surface occupied by Grasslands world-wide, Greenland and Antarctica are not included [4].

The social and ecological susceptibility to climate change are giant issues of modern era, the grassland gives an additional opinion about vulnerability of the ecosystem on account of alterations in policies, socio-economic elements, land use, and local climate [4]. Since the exposure to climatic hazards in various regions has become more apparent, the ecosystems are apt to have greater degrees of vulnerability. Globally, the number of socio-ecological attributes of the eco-systems changed drastically due to lack of enough soil moisture [5].

Plant growth, nutrient balance, photosynthetic dynamics have been influence by drought [6]. Tolerant plant species with drought responsive mechanisms at various levels such as, morphology as well as physiological and molecular basis can be used to cope up with water deficit. Since the important role of carpet-grass in ecosystem protection, number of scientists from a macro point of view have paid attention to how carpet-grass responds to global changes for instance drought, salinity, elevated temperature, and CO_2 exposure [7, 8]. However, little attention have been given to understand the genetic basis of its ecological diversifications, mainly because of the confined genomic resources in carpet-grass. Thus far, only limited ESTs and protein sequences from carpet-grass have been submitted in public online-databases [9]. The discovery of different Gene is also lagging, and just a few genes happen to be cloned and functionally authenticated [10, 11].

The high-throughput technologies, next generation sequencing (NGS) i.e., ABI/SOLiD, Roche/454, Illumina/Solexa has made it actually possible to produce genome resources at a sizable level and comparatively low cost [12, 13]. The next-generation sequencing have already been efficiently utilized to create large-scale transcriptome details in a number of plant species, i.e., Rice [14] Arabidopsis [15, 16], wheat [17], barley [18], and maize [19] to identify the candidate genes mechanisms.

The plants attempt to re-program their metabolism activities and growth, while confronting water deficit condition. It happens to be obvious that plants show distinct and extremely dynamic responses to limited water conditions [20]. These responses varied due to number of factors i.e., genotypes, experimental procedures, sampling technique and time [20, 21]. The molecular mechanisms in response to drought stress has been studied by many scientists and the genes associated with water uptake [22], transporter channels [23, 24], and transcription factors [25]. All these attributes are regarded as the regulators of drought tolerance in plants, although number of molecular elements and gene network of drought tolerance mechanism have been identified but still not fully elucidated [21].

Methods

Plant Materials and Drought Treatments

Axonopus compressus L. plants, with the age of thirty days old with uniform growth were acquired from the germplasm resource library maintained at the Hainan University, were used as experimental material. The germplasm resource library is maintained by Key Laboratory of Genetics and Germplasm Innovation of Tropical Special Forest Trees and Ornamental Plants, Ministry of Education, College of Forestry, Hainan University, Haikou 570228, P. R. China. The trial materials had been undertaken in growth chamber (27 ∘C, 16-h day length and 60% RH). A total of 90 Axonopus compressus (L.) plants were primarily divided into two groups CK (control group) and DS (drought experiment group), 5 cutings in each pot and 20 cuttings in one replicate group, this experiment was repeated 3 times named R1, R2 and R3. The drought treatments were induced by using the Polyethylene-glycol (PEG-8000). The PEG-8000 solution was replaced after 48 hours. At the 0th, 6th and 12th, 18th, 24th, 36th, 48th and 72th hours after induction of drought treatment the functional leaves (3rd to 5th mature leaf) were choose randomly from the plants of CK and DS for the purpose of physiological attributes. Samples were collected, instantly frozen in liquid nitrogen, stored at -80∘C, and finally sent for transcriptome sequencing to Metware Biotechnology Co., Ltd, Gaoxin Road, East Lake High-tech Zone, Wuhan, China.

Measurement of Physiological Traits

The physiological traits were recorded in different time interval of 0, 6th and 12th, 18th, 24th, 36th, 48th and 72th hour after induction of drought. At different time interval leaf water potential (LWP) in leaf was measured by Scholander chamber (SF-PRES-70, Solfranc Tecnologías SL, Vila-Seca, Spain). Electrolyte leakage (EL) was measured by estimating the electrical conductivity [26]. In the solution, the electrolyte leakage (S1) was measured using a conducti-meter after 22 h of floating at room temperature (Mettler-Toledo Instruments Co., Ltd, Shanghai, China). Total conductivity (S2) had been collected after the flasks were placed in a boiling water bath for 30 min. Relative water content (RWC) was also estimated, as described in Liu et al. [27]. For each plant, three independent replicates were collected for LWP, EL, and RWC. All measurements have been recorded at three biological replicates.

Extraction of RNA of Axonopus compressus, Library Preparation

Total RNA of *Axonopus compressus* was synthesized by employing the Trizol kit (Invitrogen, Carlsbad, CA, USA), and guarantee the obtained samples have been qualified for transcriptome sequencing. The RNA integrity and purity concentration were accessed by the Nanodrop method, (Qubit 2.0, Agilent 2100). Then, amount of three micro-gram of total-RNA per sample with three
biological replicates were introduced as raw materials for preparations for the RNA sample (working). The sequencing libraries of *Axonopus compressus* were created by NEBNext®Ultra™ RNA Library Prep Kit for Illumina® (New England Biolabs, Ipswich, MA, USA), as suggested by the manufacturer. The mRNAs had been extracted by total RNA by Oligo (dT) and were randomly divided into short length fragments. Random hexamer primer synthesized the first strand cDNA, then perhaps the buffer, DNA polymerase I, dNTPs and RNase H were used for processing the second strand cDNAs. The first strand of cDNA was extracted by random hexamer primer, afterword the dNTPs, buffer, DNA polymerase I and RNase H were used to generate cDNAs second strand. Finally, the AMPure XP beads were used to purify cDNAs and afterwards the end-repair and single nucleotide A (adenine) addition, the cDNA libraries (qualified) were developed by PCR technique. The Qubit 2.0 was used for primary quantitation, followed by Agilent 2100, which was employed to identify libraries insert size. Thereafter, the q-PCR technique had been used for accurate quantification of effective strength of the libraries (effective library concentration > 2 nM) to guarantee the library quality. After passing through series of screening steps, the high-throughput sequencing had been executed by Illumina Hiseq Xten.

Validation of RNA-seq data by qRT-PCR

The RNA were extracted from the CK and DS groups and used to develop the cDNA library. First-strand synthesis was carried out via MonScript (Monad) according to the suggested protocol by manufacturer. Biosystems-7500 (Thermofisher Scientific) was used to conduct the qRT-PCR and the primers pairs are listed in Table 1. The gene expression was quantified through CT-method, and the Actin1 used as references for normalization.

Table 1. Parameters of sequencing and assembly

Samples	Raw Reads	Clean Reads	Clean Base(G)	Error Rate(%)	Q20(%)	Q30(%)	GC Content(%)
Ck-1	59966928	59356354	8.90	0.02	97.91	94.02	55.66
Ck-2	62246876	60952544	9.14	0.02	97.75	93.69	55.13
Ck-3	61106902	60154449	9.02	0.02	97.83	93.86	55.39
DS-1	58110216	57318402	8.60	0.02	97.61	93.29	54.94
DS-2	56507047	55755291	8.37	0.02	97.67	93.45	55.11
DS-3	54903878	54192180	8.13	0.02	97.73	93.61	55.28
Total/Mean	352841847	347729220	52.16	0.02	97.75	93.65	55.25

Results

Effect of drought stress on physiological indicators in *Axonopus compressus*

In the Fig. 1, it was revealed that rate of H_2O_2 (Fig. 1a) production and MDA concentration (Fig. 1b) increased in drought treated plants. The MDA values and H_2O_2 production rate increase with the prolonged drought stress treatment, whereas MDA values and the production rate of H_2O_2 at 36 h after drought induction were significantly increased ($p < 0.01$). Similarly, the leaf water potential and relative water content decrease significantly under drought, contrarily the electrolyte leakage increased over the drought period in *Axonopus compressus*. As in case of MDA and H_2O_2 the values of EL were also increase as the time of drought stress increase. Whilst, the values of leaf water potential and relative water content decrease with the prolonged drought stress.

Fig. 1: Effect of drought on the physiologiacal traits of the *Axonopus compressus.*

RNA-Seq of *Axonopus compressus* and de novo assembly

RNA-Seq of the six libraries of cDNA (DS and CK with three repeats of each) resulted in 352.84 million raw reads, of which approximately 347.73 million clean reads de-novo compiled into contigs utilizing Trinity software, tend to range from 54.90 million to 62.25 million reads for each library and approximately 97% of reads with a quality score of Q20 (99% accuracy) (Table 1). The contigs were assembled into 278042 unigenes with an average length of 1066 bp and an N50 length of 1430 bp. All unigenes were over 200 bp long, of which 11.77% (32718 unigenes) were 2,000 bp long.

Functional annotation and classification of unigenes

A total of 135716 (48.81%), 188334 (67.64%), 120455 (43.32%), 189422 (68.13%), 100588 (36.18%), 153958 (55.37%) and 128626 (46.26%) unigenes would have significant levels (E-value <10−5) in KEGG, NR, Swissprot, Trembl, KOG, GO, and Pfam, respectively (Fig 2) (Additional file 1). Out of 278042 unique of high-quality sequences, 191893 (69.03%) unigenes complemented a sequence significantly in at least one of the seven public databases, The five key online databases (GO, KEGG, KOG, NR, and Trembl) were picked out of seven databases to draw a Venn diagram (Fig 3), the unigenes with significant values (Evalue10-5) are also noticed at each junctions of the Venn diagram, in which 94 unigenes corresponds to five databases. Based on the high-score BLASTx matches in the GO proteins database, The BLASTx high-score Predicated matches in the computer database of go proteins were confirmed, and a total of 153958 unigenes had been categorized with Blast2GO (E-value <10-5) and was designated at least once in GO. As shown in Fig 4, the unigenes referred to three main categories of GO and 59 subcategories, namely biological processes (BP), with 28 main sub-classes (405025 unigenes); cellular compartments (CC), with 18 main sub-classes (460094 unigenes); and molecular functions (MF), with 13 main sub-classes (194989 unigenes).

Fig 2. Unigenes matched in seven databases.

Fig 4. Functional classification of GO terms for *Axonopus compressus*.

Fig 3. Venn diagram of differential BLAST results for *Axonopus compressus*. The number of unigenes annotated with 5 databasesand the co-annotated gene number are presented

Cellular processes (21.43%) were the biggest subgroups in the category of biological processes, metabolic process (19.70%), biological regulation (9.24%), response to stimulus (9.22%) and regulation of biological process (8.36%). The largest subgroups in the cellular component category were cell (22.53 %) followed by cell part (22.47 %), organelle (17.22%), and membrane (11.72%) respectively. Similarly, the main subgroups in the molecular function category were binding and catalytic activity, that contribute 77.78% and 69.28% respectively, and 115199 unigenes associated to molecular function.

Within the *Axonopus compressus* unigenes, 112492 (42.64%) had been classified (E-value <10-5) into twenty six KOG clusters (Fig 5). The biggest groups which includes 1) general function prediction only (21955 genes, 19.52%); 2) posttranslational modification, protein turnover, and chaperones (11636 genes, 10.34%); 3) signal transduction mechanisms (11302 genes, 10.05%); 4) Function unknown (5785 genes, 5.14%); and 5) carbohydrate transport and metabolism (5616 genes, 4.99%).

Fig 5. KOG functional classification of the *Axonopus compressus* transcriptome.

Metabolic pathway analysis of *Axonopus compressus* by KEGG

A total of 135717 (51.44%) of the 263835 *Axonopus compressus* unigenes possessed significant correspondence in KO. All these unigenes, such unigenes have been reserved to 103 KEGG pathways and could be splited to 5 categories (Fig 5). The pathways of KEGG includes 4303 unigenes were the members of major group, metabolism (D), 921 associated to genetic information processing (C), 184 related to cellular processes (A), 347 involved to environmental information (B), and 151 related to category (E) of organism systems (Fig. 6) (Additional file 2).

Fig 6. KEGG pathway annotation for *Axonopus compressus*.

CDS prediction in *Axonopus compressus*

The BLASTx protein database (NR and SwissProt database) identified 278042 unigenous CDSs, of which 23652 unigenes were larger than 500 bp, 12520 unig In addition, the 43,713 unigenes were not linked to the NR and SwissProt database systems. The Estscan (Version; 3.0.3) software were used to interpret their ORF, frequency distribution, length, and related amino acid sequences of the unigene CDSs.

Differentially expressed genes (DEGs) analysis of *Axonopus compressus*

Amongst the differentially expressed unigenes, the expression of 7444 differs substantially between samples treated with drought-stress (DS) and control (CK) samples. Under drought treatment 4249 numbers were up-regulated and 3195 numbers were down-regulated ($p<0.05$) (Fig. 7) (Additional file 3).

Fig 7. Volcano plot of differentially expressed unigenes of *Axonopus compressus*.

***Axonopus compressus* GO enrichment analysis**

By employing the Gene ontology (GO) and the DEG enrichment analysis in *Axonopus compressus*, 23766 DEGs have been categorized to three GO groups and 4592 numbers associating the DS with CK (number of DEGs annotated in more than one terms), out of which 2766 items found associated to BP, 477 were linked with CC and 1349 were related to MF.

The key 50 DEGs dramatically enriched in three GO categories which are presented in Fig 8. In the top 50 significantly enriched identities (Corrected P-Value<0.05), the 18 found to be linked with BP [carotenoid biosynthetic process GO:0016117, carotenoid metabolic process GO:0016116, cellular amine metabolic process GO:0044106, cellular biogenic amine metabolic process GO:0006576, cellular response to heat GO:0034605, cellular transition metal ion homeostasis GO:0046916, glutamine family amino acid biosynthetic process GO:0009084, Group II intron splicing GO:0000373, heat acclimation GO:0010286, oligosaccharide catabolic process GO:0009313, raffinose catabolic process GO:0034484, raffinose metabolic process GO:0033530, regulation of seed germination GO:0010029, regulation of seedling development GO:1900140 , response to high light intensity GO:0009644, response to hydrogen peroxide GO:0042542, tetraterpenoid biosynthetic process GO:0016109, tetraterpenoid metabolic process GO:0016108], and 6 items were related to CC [chloroplast nucleoid GO:0042644, DNA packaging complex GO:0044815, Nucleoid GO:0009295, Nucleosome GO:0000786, plastid nucleoid GO:0042646, protein-DNA complex GO:0032993], and 14 items were related to MF [4-coumarate-CoA ligase activity GO:0016207, alpha-galactosidase activity GO:0004557, arogenate dehydratase activity GO:0047769, carbon-nitrogen ligase activity, with glutamine as amido-N-donor GO:0016884, ferric iron binding GO:0008199, ferroxidase activity GO:0004322, galactosidase activity GO:0015925, oxidoreductase activity, acting on single donors with incorporation of molecular oxygen GO:0016701, oxidoreductase response, acting on single donors with molecular oxygen incorporation, incorporation of two atoms of oxygen GO:0016702, oxidoreductase response, oxidizing metal ions, oxygen as acceptor GO:0016724, raffinose alpha-galactosidase activity GO:0052692, water channel activity GO:0015250, water transmembrane transporter activity GO:0005372](Additional file 4, 5).

Fig 8. GO-enriched DEGs in *Axonopus compressus*.

Axonopus compressus KEGG pathway enrichment analysis

The KEGG pathway enrichment analysis for DEGs revealed that 2569 DEGs participated in 143 various types of pathways in *Axonopus compressus.* By comparing DS with CK, the number of 2747 DEGs found up-regulated and 2502 DEGs have been identified as down-regulated in deficit water. The twenty key pathways which are found significantly enriched by comparing DS with CK are displayed in Fig 9. The pathways includes Valine, leucine and isoleucine biosynthesis (215 unigenes), Stilbenoid, diarylheptanoid and gingerol biosynthesis (330 unigenes), Starch and sucrose metabolism (1642 unigenes), RNA degradation (1536 unigenes), Protein processing in endoplasmic reticulum (1628 unigenes), Porphyrin and chlorophyll metabolism (559 unigenes), Plant hormone signal transduction (1672 unigenes), Phenylalanine, tyrosine and tryptophan biosynthesis (601 unigenes), Metabolic pathways (1659 unigenes), MAPK signaling pathway – plant (1661 unigenes), Linoleic acid metabolism (323 unigenes), Glyoxylate and dicarboxylate metabolism (1050 unigenes), Glycosphingolipid biosynthesis - globo and isoglobo series (103 unigenes), Galactose metabolism (735 unigenes), Carotenoid biosynthesis (506 unigenes), Carbon fixation in photosynthetic organisms (820 unigenes), Biosynthesis of secondary metabolites - unclassified (223 unigenes), Biosynthesis of secondary metabolites (1661 unigenes), Biosynthesis of amino acids (1655 unigenes), Alanine, aspartate and glutamate metabolism (639 unigenes) (Additional file 6).

Fig 9. KEGG pathway enrichment of DEGs in *Axonopus compressus*.

Drought stress associated differentially expressed transcription factors (TFs)

The transcription factors play a crucial role in growth and development of plants, and therefore can stimulate and/or suppress transcriptional gene expression to sustain normal physiological function in deficit water stress. In current study, the 352 TFs were observed in *Axonopus compressus* based on iTAK, including 270 transcription factors and 81 transcription regulatory factors that associated to 32 and 16 families, respectively (Additional file 7, 8). Differential analysis revealed 270 transcription factors were remained differentially expressed by comparing DS with CK, of which 216 transcription factors have been noticed up-regulated and 54 transcription factors were found down-regulated.

qRT-PCR Validation of Transcripts in *Axonopus compressus* under Drought

To check the validity of RNA-seq results, expression of six transcripts for the three biological replicates was analyzed by qRT-PCR (Figure 10). All the genes were identified as significantly different expression values (DEGs). The finding of qRT-PCR showed that the pattern of expression of these genes was identical to the shown in RNA-seq analysis. To validate the results, the six drought-responsive genes (NAC, MAP Kinase1, MYB2, PIP1, WRKY1, ABI5) were verified using qRT-PCR. The different genes were amplified to compare the gene expression and RNA-seq results, and the findings of this study showed that the expression of drought-responsive genes supporting the results of RNA-seq.

Figure 10: The qRT-PCR expression validation of drought-responsive genes in *A. compressus*. The bars showed relative expression of (A) NAC (B) MAP Kinase1, (C) MYB1, (D) PIP2, (E) WRKY1, (F) ABI5.

Discussion

Plant have a sessile lifestyle, and usually confront adverse environmental circumstances i.e., drought, cold, heat, salt, and floods. Deficit water is the most notable environmental determinant influencing crop production around the globe [28]. To improve potential tolerance mechanism against deficit water stress and also to develop drought-resistance in plants, many experts have attempted to elucidate the mechanisms of stress signaling in plants conferring the several perspectives [29, 30]. Plant defense mechanisms for abiotic stress are already being studied, but the response to drought has always been a complex event with several critical indicators to be examined.

Drought stress significantly increased ROS formation and imposes oxidative stress on the plant [26, 31]. Since lipid peroxidation is one of the earliest indicators of oxidative damage, MDA was estimated as an index for production of drought induced ROS [31]. A significant rise in MDA values and H_2O_2 concentration were noticed in *Axonopus compressus* leaves in drought condition (Fig. 1). Electrolyte leakage increased antagonistically with decline in water potential (Fig. 1), which proposed that plants system had been damaged by drought, as studied previously [32]. Drought may have an impact on the physical membranous structure of the lipid bilayer by inducing phase destruction [33].

To sustain under severe environmental stipulations, plants have developed an obscure regularity mechanism through a series of evolutionary advancement at varied levels to understand external signals and prompt transduction of stress signals, directing to a set of responses at different levels, i.e., physio-morphological, biochemical, and omic levels [34, 35]. As a strong drought-responsive plant, *Axonopus compressus* developed a series of morphological features to acclimate the deficit water conditions. Drought stress can drive the plant to change the metabolic status of cutin and wax, which is ultimately accomplished through the associated gene-expression regulation [36] In current study, the unigene responsible for the wax biosynthesis (Cluster-37496.124448) was enriched under DS in comparison with CK and also up-regulated at the transcript level. As a distinctive morphological feature of protection against drought stress, cutin and wax impersonate a critical function during water deficit condition by restricting loss of water by means of leaf epidermis (nonstomatal transpiration) [37, 38], therefore enhancing water use capability in plants. The current result revealed that all such changes associated with the series of adaptation processes in *Axonopus compressus* against water deficit. In our study, the transcription factors belong to the family AP2/ERF-ERF, MYB-related, zinc-finger, NF-X1 were found to be up-regulated. these transcription factors were known to regulate the biosynthesis of plant cutin and wax [39].

In water deficit, the plants going to receive stress signals first thereafter a number of pathways in a sequence triggered by phyto-hormones [40]. Subsequently, the plant hormones have an accountable part in response to scarce water conditions, and amongst the phyto-hormones, ABA is known to be a major homeostatic controller of abiotic stresses. Under water deficit conditions, the plants increase its ABA- content and will happen to closing of stomata in order to avoid the excessive water loss by transpiration across stomata [41], such high levels water loss would continue to persist sometimes even after transient recovery of leaf water status [42].

In current study, we observed 8 unigenes which encode NCED4 (9-cis-epoxycarotenoid dioxygenase) (Cluster-37496.127826) in *Axonopus compressus*, the crucial enzyme accountable for ABA biosynthesis (GO:0009688). Amongst 8 unigenes, 4 are up-regulated, and exhibited substantially different expression levels between DS and CK. Such behavior may possibly because of drought stress; *Axonopus compressus* plants had perceived signals firstly, which lead to ABA production, which observed to be translocated to the above-ground parts. The transportation mechanism system in the stem take into action and trigger short-term responses such as stomatal closure [40, 43], which will help *Axonopus compressus* to avert the dehydration and improve water withholding potential under drought stress. Accordingly, the understanding of stress signal and subsequent molecular mechanisms in *Axonopus compressus* needs to be elucidated in succeeding experiments to reveal the drought-resistance mechanism.

When plants respond to drought, the rise in amounts of ABA leads to binding with PYR/PYL, which perform a significant role in quantitative regulation of stomatal fissure and transcriptional response to ABA [44, 45]. The PYR/PYL reconstructs the PYR/PYL protein conformations, and this alteration enables the PYR/PYL to interplay with 2 C protein phosphatase (PP2C), negative regulator type, to construct a substitute complex (ABA-PYR/PYL-PP2C). The ABA-PYR/PYL-PP2C- complex may impede PP2C activity and trigger SNF1-related protein kinase 2 (SnRK2s), a positive regulator. While the PP2C also inhibits the activity of SnRK2s [46].

Such mechanism stimulate the stress-responsive genes to down-regulate its expression and support the plants to acclimate water deficit condition [47]. We found 715 PP2C in our experiments, from which 82 genes were remarkably down-regulated, and 17 SnRK2 were also identified. This symbolises that under water deficit, the low expression of PP2C in *Axonopus compressus* rescue the suppression of SnRK, and its expression promotes the closing of stomata. The SnRKexpression also trigger the stress-related genes towards downstream and support *Axonopus compressus* keeps normal growth and persistence in drought.

In plants, the stress responding genes can easily be divided into two groups i.e., functional and regulatory [48]: The functional proteins particularly with respect to small molecular osmolytes (i.e., proline, betaine, and soluble sugar), enzyme protectants (POD, SOD and CAT), later embryogenesis abundant (LEA), and aquaporins. The regulatory proteins include TFs, phosphatase, protein kinase and phospholipid metabolic enzymes, that can regulate the stress-related expression of genes under abiotic stresses.

This symbolises that under water deficit, the lowest expression of PP2C in *Axonopus compressus* might have rescue the SnRK suppression. The PP2C expression would have induce closing of stomata and trigger the downstream drought stress-related genes and assist the *Axonopus compressus* to sustain unhampered growth under drought. These proteins will prevent the plant cells from damaging encounters of stress by managing turgor pressure, including oxygen-free radicals scavenging and the structural intracellular bio-macromolecules protection [49, 50] the transcriptional expression of drought related genes were regulated by the TFs.

It has been confirmed that members of the TF families i.e., MYB, AP2, DREB, bHLH, PLATZ, bZIP, C2H2, NAC WRKY and HB are involved in plant stress-response mechanisms [51]. In our research, we found 270 DEGs in *Axonopus compressus* TFs, among them, 216 were up-regulated and 54 were down-regulated. it is obvious from the results that large numbers of TF expressions altered significantly under water deficit.

In the DEGs HARBI1,EEF1A□SUMO□ATXR3□SEY1□NLR□COMM□DHAR3□GUF1□SFH5□SHOC2 □MAPK's, FYVE□PIP, LSM14, PERQ□BRCA1□TIM22□STY46-like□SPN1□NEDD8□DDX21□Meis2□RAD50□MAG2□MON1□and AKT2 are all down-regulated, and RXW8, DHAR2-like, EFR3, MOCS2A, GDSL, ANP1-like, TBC1, ERCC-6, ZDHHC2, EXO84B, BAHD, SHOC2, XBAT32, RNF38, SPIRAL1, NDRG1, HESO1, NBR1, STY46, TNNI3K, PP2C26, HVA22 and DAD1 are all up-regulated. The TF families i.e., GATA, ATF/CREB, ABI, bZIP, WRKY, EREBP, AP2-like, PTI6, TRAF, ARF1, ATF/CREB, ABA responsive, RING, MYB, NAC, TUB, HSF, GRAS, C2H2, bZIP, SET, AP2/ERF-ERF, AUX/IAA, SNF2 and PTI6 some of them are up-regulated and some of them are down-regulated. Schematic diagram of potential defense mechanism and drought tolerance involving different gene related pathways in *Axonopus compressus* are shown in Fig. 11.

Several unigenes belong to similar family reported to have distinct expression models under water deficit conditions, that might symbolize various characteristic function in response to stress condition, and their behavior to drought [46, 52]. A particular transcription factor would have been associated with one and/or more than one members in the particular category and even other groups [53], which also reveals the complexity by which transcriptional

categories of genes that controls the drought responses of *Axonopus compressus*. The current study provides helpful data for additional functional reviews of the TFs' functions in order to develop tolerance in *Axonopus compressus*.

During signal transduction, the post-translational protein alterations are the essential phenomenon, of phosphorylation and de-phosphorylation in plants under stress condition [54]. Phosphorylation and dephosphorylation would trigger a number of proteins and enzymes with regulatory functions to control a wide range of cellular mechanisms or signal process. Therefore, likely to halt or initiate the enzyme activity in drought, unless adjusting the concentration of resultant proteins or intracellular enzymes activity, thus, it have been established that protein phosphorylation adjustment plays a significant function in the response to drought [54]. It is revealed that there are more than Arabidopsis have thousands of kinases [55], the CDPK (calcium-dependent protein kinase) and MAPK (Mitogen-activated protein kinase) are considered critical signaling mechanism in the plant under stress.

The stress signals were transduce into cellular processes by MAPK, succeeding phosphorylation processes of distinct downstream proteins to on and/or off their activities [56]. As a distinctive antenna, CDPKs can immediately transform upstream $Ca2^{+}$ signals to protein phosphorylation at downstream because of its structure of multifunctional protein, which couples calcium binding, and signalling capacities in product of single gene [57].

In our study, we have observed 11089 protein kinases, Of which 344 are DEGs, out of which 182 unigenes have been up-regulated and 162 unigenes were found down-regulated, besides, we identified 16 and 8 DEGs encoding MAPK and CDPK respectively, and there are 9 and 2 DEGs up-regulated and 7 and 6 DEG were down-regulated, respectively. Whilst, it suggests that the MAPK and CDPK pathways engage in the signaling of drought stress as signal transduction variables and it may plays an important role against osmotic stress in *Axonopus compressus*. The plants faced water deficit conditions; osmo-tension would provoke ROS built-up in cells. Plants had previously been evolved the defense mechanism to extinguish the ROS and alleviate cellular damage. This defence mechanism comprises of SOD, POD and CAT. Current study showed 5 SOD, 3 CAT and 25 POD DEGs. In addition, proteins of small-molecule such as proline, betaine and LEA (late embryogenesis abundant) likewise play significant roles in restraining the cells from the damaging effect of drought.

Since LEA is a major osmotic regulator, LEA also belongs to large protein group that was observed to be first accumulated while dehydration under the lateral arena of developments. In current study, we observed 370 unigenes which can happen to encodes LEA. We observe 19 DEGs in *Axonopus compressus* (DS with CK), all of them were up-regulated. Therefore, the unigenes are crucial for the developing sustainability against the water shortage for *Axonopus compressus*. Additionally, the osmotic regulation ability of plants to hamper water loss is an additional fundamental feature to survive in drought. The bidirectional water channel AQP (aquaporin) in plants responsible for trans-membrane water mobility and long-distance water transportation. It is established that plants would have potential to defy the numerous stress circumstances through regulating the proteins related to water channel [58]; in *Axonopus compressus*, we observed 363 unigenes coding AQP, including 31 unigenes differentially expressed, 2 unigenes were found down-regulated and no one up-regulated. The AQP's showed reduced expression, demonstrated the decline in AQP's activity and/or ceased, the cessation of AQPs be able to limit moisture loss thus sustain water related homeostatic processes of plants,consequently, improving tolerance against drought in *Axonopus compressus*.

Fig 11. Schematic diagram of potential defense mechanisms for drought stress response and tolerance pathways in carpet-grass (*Axonopus compressu* L.).

Conclusion

Under drought condition, processing of stress signals, signal transduction, regulating gene expression and the subsequent downstream functional genes are essential variables in plant response to adversity. In current study, we observed 263835 unigenes in *Axonopus compressus* based on RNA-Seq, in which 7445 were differentially expressed unigenes (DS and CK). Overall 2747 were up-regulated and 2502 down-regulated unigenes. In addition, 352 (TFs) were found to be differentially expresed in *Axonopus compressus* (DS and CK). The findings indicate that these genes are involved in the campaign of resistance to drought stress and have a highly significant and complicated role. This current study provide valuable data for the molecular mechanisms underlying drought tolerance and qRT-PCR validation that sets the basis for further investigations of the gene regulatory networks under drought stress and other abiotic stress factors in *Axonopus compressus*.

Abbreviations

DEGs: Differentially expressed genes

DEPs: Differentially expressed proteins

FPKM: Fragments Per Kilobase Million

GO: Gene Ontology

KO: KEGG Orthology

GST: Glutathione S-transferase

DS: Drought Stress

CK: Control

NGS: Next Generation Sequencing

KEGG: Kyoto Encyclopedia of Genes and Genomes

MAPK: Mitogen-activated protein kinase

NDEGs: No significant difference in genes

RNA-seq: RNA sequencing

ROS: Reactive oxygen species

MDA: Malondialdehyde

EL: Electrolyte Leakage

cDNA: Complementary DNA

RNA-Seq: RNA sequencing

DEGs: Differentially expressed genes

CDSs: CoDing Sequences

ORF: Open Reading Frame

ncRNAs: Non-coding RNAs

DE: Differentially expressed

TFs: Transcription factors

BP: Biological processes

CC: Cellular components

MF: Molecular functions

POD: Peroxidase

SOD: Superoxide Dismutase

CAT: Catalase

LEA: Later Embryogenesis Abundant

ABA: Abscisic acid

PYR/PYL/RCAR: Pyrabactin Resistance1/Pyr1-Like/Regulatory Components of Aba Receptors

PP2C: Protein Phosphatase type 2C

SnRK2: Serine/Threonine Kinases2

SNF1: Sucrose Nonfermenting 1

PEG: Polyethylene glycol

LWP: Leaf water potential

dNTP: Deoxynucleoside triphosphate

qRT-PCR: Real-Time Quantitative Reverse Transcription PCR

Declarations

Competing Interests

The author(s) announced no potential conflicts of interest with regards to the research, authorship, and/or publication of this manuscript.

Author Contributions

Conceptualization: MN, ZW

Data curation: MN

Formal analysis: MN, FA, AZ

Funding acquisition: MN, ZW

Investigation: MN, LL

Methodology: MN

Writing – original draft: MN, FA, YH, LL

Writing – review & editing: MN, FA, ZW, SS, UA

All authors have read and approved the manuscript.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Funding

This work was supported by the National Natural Science Foundation of China (31060266), the Hainan Provincial Natural Science Foundation (310029). National Project on Sci-Tec Foundation Resources Survey (2017FY100600). The authors are grateful to the editors and referees for their valuable comments to improve our manuscript.

References

1. López A, Morrone O. Phylogenetic studies in Axonopus (Poaceae, Panicoideae, Paniceae) and related genera: morphology and molecular (nuclear and plastid) combined analyses. Syst Bot. 2012;37:671–6.
2. Ighovie ES, Ikechukwu EE. Phytoremediation of Crude Oil Contaminated Soil with Axonopus compressus in the Niger Delta Region of Nigeria. Nat Resour. 2014;5:59–67. doi:10.4236/nr.2014.52006.
3. Wang ZY, Hu HG, Bai CJ, Wang XL, Mo DQ, Li LF, et al. Genetic variation of asexual reproduction characteristics of Axonopus compressus (Sw.) P. Beauv. In: VI International Symposium on the Taxonomy of Cultivated Plants 1035. 2013. p. 189–203.
4. Ojima DS, Chuluun T, Galvin KA. Social-Ecological Vulnerability of Grassland Ecosystems. In: Climate Vulnerability: Understanding and Addressing Threats to Essential Resources. 2013. p. 151–62.
5. Knapp AK, Carroll CJW, Denton EM, La Pierre KJ, Collins SL, Smith MD. Differential sensitivity to regional-scale drought in six central US grasslands. Oecologia. 2015;177:949–57.
6. Farooq M, Wahid A, Kobayashi N, Fujita D, Basra SMA. Plant Drought Stress: Effects, Mechanisms and Management. In: Sustainable Agriculture. Dordrecht: Springer Netherlands; 2009. p. 153–88. doi:10.1007/978-90-481-2666-8_12.
7. Deng X, Liu K, Li M, Zhang W, Zhao X, Zhao Z, et al. Difference of selenium uptake and distribution in the plant and selenium form in the grains of rice with foliar spray of selenite or selenate at different stages. F Crop Res. 2017;211:165–71.
8. Lei T, Pang Z, Wang X, Li L, Fu J, Kan G, et al. Drought and carbon cycling of grassland ecosystems under global change: A review. Water. 2016;8:460.
9. Wang XL, Li Y, Liao L, Bai CJ, Wang ZY. Isolation and characterization of microsatellite markers for Axonopus compressus (Sw.) Beauv.(Poaceae) using 454 sequencing technology. Genet Mol Res. 2015;14:4696–702.
10. Porse A, Schou TS, Munck C, Ellabaan MMH, Sommer MOA. Biochemical mechanisms determine the functional compatibility of heterologous genes. Nat Commun. 2018;9:522.
11. Byeon Y, Lee H, Lee HY, Back K. Cloning and functional characterization of the Arabidopsis N-acetylserotonin O-methyltransferase responsible for melatonin synthesis. J Pineal Res. 2016;60:65–73.
12. Gierahn TM, Wadsworth II MH, Hughes TK, Bryson BD, Butler A, Satija R, et al. Seq-Well: portable, low-cost RNA sequencing of single cells at high throughput. Nat Methods. 2017;14:395.
13. Adams IP, Fox A, Boonham N, Massart S, De Jonghe K. The impact of high throughput sequencing on plant health diagnostics. Eur J plant Pathol. 2018;152:909–19.
14. Tang X, Liu G, Zhou J, Ren Q, You Q, Tian L, et al. A large-scale whole-genome sequencing analysis reveals highly specific genome editing by both Cas9 and Cpf1 (Cas12a) nucleases in rice. Genome Biol. 2018;19:84.
15. Srivastava D, Verma G, Chauhan AS, Pande V, Chakrabarty D. Rice (Oryza sativa L.) tau class glutathione S-transferase (OsGSTU30) overexpression in Arabidopsis thaliana modulates a regulatory network leading to heavy metal and drought stress tolerance. Metallomics. 2019;11:375–89.
16. Dubois M, Claeys H, Van den Broeck L, Inzé D. Time of day determines Arabidopsis transcriptome and growth dynamics under mild drought. Plant Cell Environ. 2017;40:180–9.
17. Zotova L, Kurishbayev A, Jatayev S, Khassanova G, Zhubatkanov A, Serikbay D, et al. Genes encoding transcription factors TaDREB5 and TaNFYC-A7 are differentially expressed in leaves of bread wheat in response to drought, dehydration and ABA. Front Plant Sci. 2018;9:1441.
18. Janiak A, Kwasniewski M, Sowa M, Kuczyńska A, Mikołajczak K, Ogrodowicz P, et al. Insights into Barley Root Transcriptome under Mild Drought Stress with an Emphasis on Gene Expression Regulatory Mechanisms. Int J Mol Sci. 2019;20:6139.

19. Liu X, Zhang X, Sun B, Hao L, Liu C, Zhang D, et al. Genome-wide identification and comparative analysis of drought-related microRNAs in two maize inbred lines with contrasting drought tolerance by deep sequencing. PLoS One. 2019;14.
20. Gosa SC, Lupo Y, Moshelion M. Quantitative and comparative analysis of whole-plant performance for functional physiological traits phenotyping: new tools to support pre-breeding and plant stress physiology studies. Plant Sci. 2019;282:49–59.
21. Min H, Chen C, Wei S, Shang X, Sun M, Xia R, et al. Identification of drought tolerant mechanisms in maize seedlings based on transcriptome analysis of recombination inbred lines. Front Plant Sci. 2016;7:1080.
22. Harris-Shultz KR, Hayes CM, Knoll JE. Mapping QTLs and Identification of Genes Associated with Drought Resistance in Sorghum. In: Sorghum. Springer; 2019. p. 11–40.
23. Scharwies JD, Dinneny JR. Water transport, perception, and response in plants. J Plant Res. 2019;132:311–24.
24. Li J, Yu G, Sun X, Liu Y, Liu J, Zhang X, et al. AcPIP2, a plasma membrane intrinsic protein from halophyte Atriplex canescens, enhances plant growth rate and abiotic stress tolerance when overexpressed in Arabidopsis thaliana. Plant Cell Rep. 2015;34:1401–15.
25. Fei X, Hou L, Shi J, Yang T, Liu Y, Wei A. Patterns of Drought Response of 38 WRKY Transcription Factors of Zanthoxylum bungeanum Maxim. Int J Mol Sci. 2019;20:68.
26. Nawaz M, Wang Z. Abscisic Acid and Glycine Betaine Mediated Tolerance Mechanisms under Drought Stress and Recovery in Axonopus compressus: A New Insight. Sci Rep. 2020;10:1–10.
27. Liu N, Lin S, Huang B. Differential Effects of Glycine Betaine and Spermidine on Osmotic Adjustment and Antioxidant Defense Contributing to Improved Drought Tolerance in Creeping Bentgrass. J Am Soc Hortic Sci. 2017;142:20–6.
28. Spokevicius A V, Tibbits J, Rigault P, Nolin M-A, Müller C, Merchant A. Medium term water deficit elicits distinct transcriptome responses in Eucalyptus species of contrasting environmental origin. BMC Genomics. 2017;18:284.
29. Saddhe AA, Malvankar MR, Karle SB, Kumar K. Reactive nitrogen species: paradigms of cellular signaling and regulation of salt stress in plants. Environ Exp Bot. 2019;161:86–97.
30. Hasanuzzaman M, Shabala L, Brodribb TJ, Zhou M, Shabala S. Understanding physiological and morphological traits contributing to drought tolerance in barley. J Agron Crop Sci. 2019;205:129–40.
31. Anjum SA, Ashraf U, Tanveer M, Khan I, Hussain S, Shahzad B, et al. Drought Induced Changes in Growth, Osmolyte Accumulation and Antioxidant Metabolism of Three Maize Hybrids. Front Plant Sci. 2017;08. doi:10.3389/fpls.2017.00069.
32. Jiang Y, Huang B. Protein Alterations in Tall Fescue in Response to Drought Stress and Abscisic Acid. Crop Sci. 2002;42:202–7. doi:doi:10.2135/cropsci2002.2020.
33. Gujjar RS, Akhtar M, Rai A, Singh M. Expression analysis of droughtinduced genes in wild tomato line (Solanum habrochaites). Curr Sci. 2014;107:496–501.
34. Cantalapiedra CP, García-Pereira MJ, Gracia MP, Igartua E, Casas AM, Contreras-Moreira B. Large Differences in Gene Expression Responses to Drought and Heat Stress between Elite Barley Cultivar Scarlett and a Spanish Landrace. Front Plant Sci. 2017;8 May.
35. Osakabe Y, Osakabe K, Shinozaki K, Tran L-SP. Response of plants to water stress. Front Plant Sci. 2014;5:86.
36. Yadav A, Stobdan T, Chauhan OP, Dwivedi SK, Chaurasia OP. Sea Buckthorn: A Multipurpose Medicinal Plant from Upper Himalayas. In: Medicinal Plants. Springer; 2019. p. 399–426.
37. Ding F, Wang G, Wang M, Zhang S. Exogenous melatonin improves tolerance to water deficit by promoting cuticle formation in tomato plants. Molecules. 2018;23:1605.
38. Bi H, Kovalchuk N, Langridge P, Tricker PJ, Lopato S, Borisjuk N. The impact of drought on wheat leaf cuticle properties. BMC Plant Biol. 2017;17:85.
39. Ziv C, Zhao Z, Gao YG, Xia Y. Multifunctional roles of plant cuticle during plant-pathogen interactions. Front Plant Sci. 2018;9:1088.
40. Nguyen D, Rieu I, Mariani C, van Dam NM. How plants handle multiple stresses: hormonal interactions underlying responses to abiotic stress and insect herbivory. Plant Mol Biol. 2016;91:727–40.
41. Tombesi S, Nardini A, Frioni T, Soccolini M, Zadra C, Farinelli D, et al. Stomatal closure is induced by hydraulic signals and maintained by ABA in drought-stressed grapevine. Sci Rep. 2015;5:1–12.
42. Martorell S, DIAZ-ESPEJO A, Medrano H, Ball MC, Choat B. Rapid hydraulic recovery in E ucalyptus pauciflora after drought: linkages between stem hydraulics and leaf gas exchange. Plant Cell Environ. 2014;37:617–26.
43. Dubois M, Inzé D. Plant growth under suboptimal water conditions: early responses and methods to study them. J Exp Bot. 2020.
44. Dittrich M, Mueller HM, Bauer H, Peirats-Llobet M, Rodriguez PL, Geilfus C-M, et al. The role of Arabidopsis ABA receptors from the PYR/PYL/RCAR family in stomatal acclimation and closure signal integration. Nat plants. 2019;5:1002–11.
45. Yu J, Yang L, Liu X, Tang R, Wang Y, Ge H, et al. Overexpression of poplar pyrabactin resistance-like abscisic acid receptors promotes abscisic acid sensitivity and drought resistance in transgenic Arabidopsis. PLoS One. 2016;11.
46. Ye G, Ma Y, Feng Z, Zhang X. Transcriptomic analysis of drought stress responses of sea buckthorn (Hippophae rhamnoidessubsp. sinensis) by RNA-Seq. PLoS One. 2018;13.
47. Sah SK, Reddy KR, Li J. Abscisic acid and abiotic stress tolerance in crop plants. Front Plant Sci. 2016;7:571.
48. Liu J-H, Peng T, Dai W. Critical cis-acting elements and interacting transcription factors: key players associated with abiotic stress responses in plants. Plant Mol Biol Report. 2014;32:303–17.

49. Qureshi MK, Munir S, Shahzad AN, Rasul S, Nouman W, Aslam K. Role of reactive oxygen species and contribution of new players in defense mechanism under drought stress in rice. Int J Agric Biol. 2018;20:1339–52.
50. Rokhzadi A. Response of chickpea (Cicer arietinum L.) to exogenous salicylic acid and ascorbic acid under vegetative and reproductive drought stress conditions. J Appl Bot Food Qual. 2014;87.
51. Gahlaut V, Jaiswal V, Kumar A, Gupta PK. Transcription factors involved in drought tolerance and their possible role in developing drought tolerant cultivars with emphasis on wheat (Triticum aestivum L.). Theor Appl Genet. 2016;129:2019–42.
52. Kulkarni M, Soolanayakanahally R, Ogawa S, Uga Y, Selvaraj MG, Kagale S. Drought response in wheat: key genes and regulatory mechanisms controlling root system architecture and transpiration efficiency. Front Chem. 2017;5:106.
53. Samad AFA, Sajad M, Nazaruddin N, Fauzi IA, Murad A, Zainal Z, et al. MicroRNA and transcription factor: key players in plant regulatory network. Front Plant Sci. 2017;8:565.
54. Sun H, Xia B, Wang X, Gao F, Zhou Y. Quantitative phosphoproteomic analysis provides insight into the response to short-term drought stress in Ammopiptanthus mongolicus roots. Int J Mol Sci. 2017;18:2158.
55. Bhaskara GB, Nguyen TT, Yang T-H, Verslues PE. Comparative analysis of phosphoproteome remodeling after short term water stress and ABA treatments versus longer term water stress acclimation. Front Plant Sci. 2017;8:523.
56. Ma H, Chen J, Zhang Z, Ma L, Yang Z, Zhang Q, et al. MAPK kinase 10.2 promotes disease resistance and drought tolerance by activating different MAPKs in rice. Plant J. 2017;92:557–70.
57. Hamel L-P, Sheen J, Séguin A. Ancient signals: comparative genomics of green plant CDPKs. Trends Plant Sci. 2014;19:79–89.
58. Quiroga G, Erice G, Aroca R, Zamarreño ÁM, García-Mina JM, Ruiz-Lozano JM. Radial water transport in arbuscular mycorrhizal maize plants under drought stress conditions is affected by indole-acetic acid (IAA) application. J Plant Physiol. 2020;:153115.

Figures

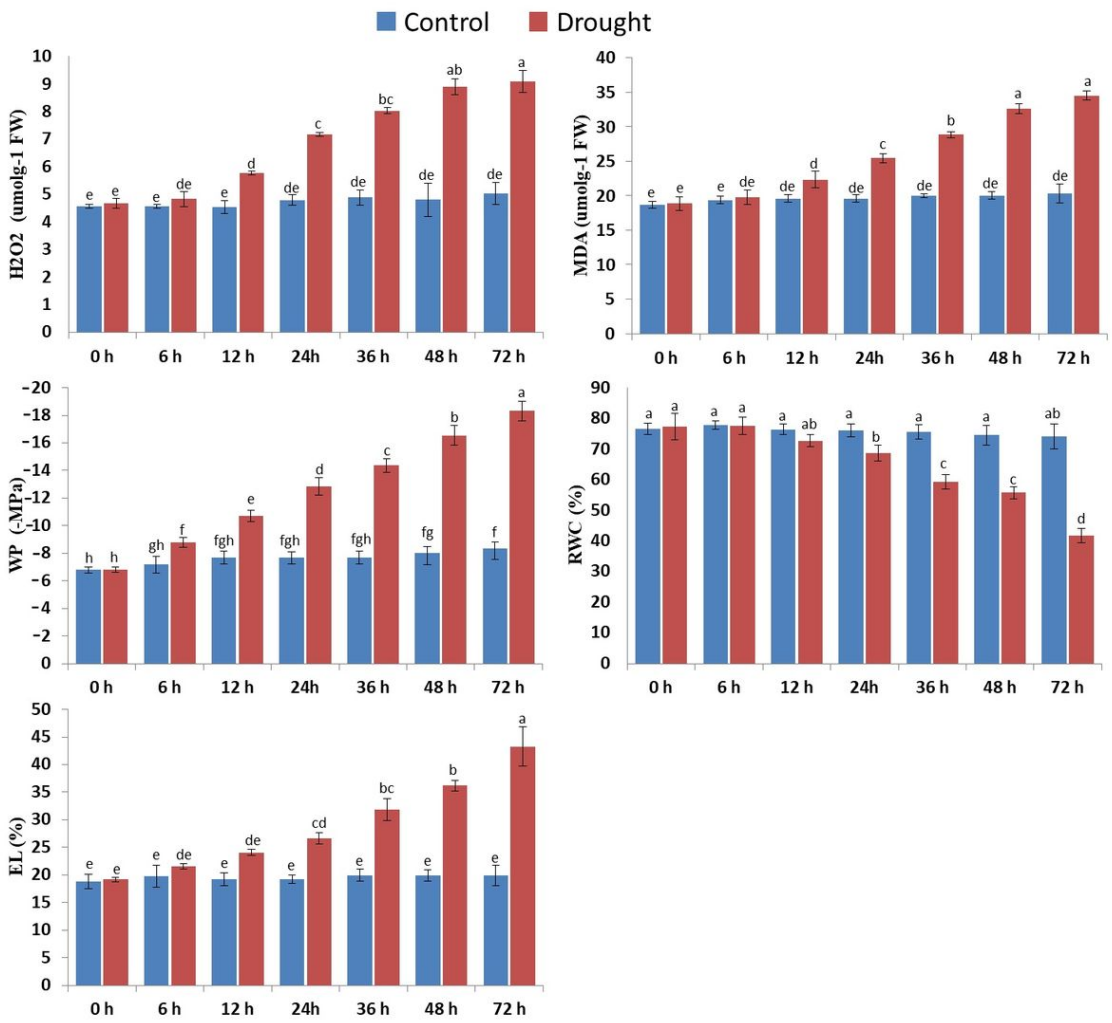

Figure 1

Effect of drought on the physiologiacal traits of the Axonopus compressus. RNA-Seq of Axonopus compressus and de novo assembly

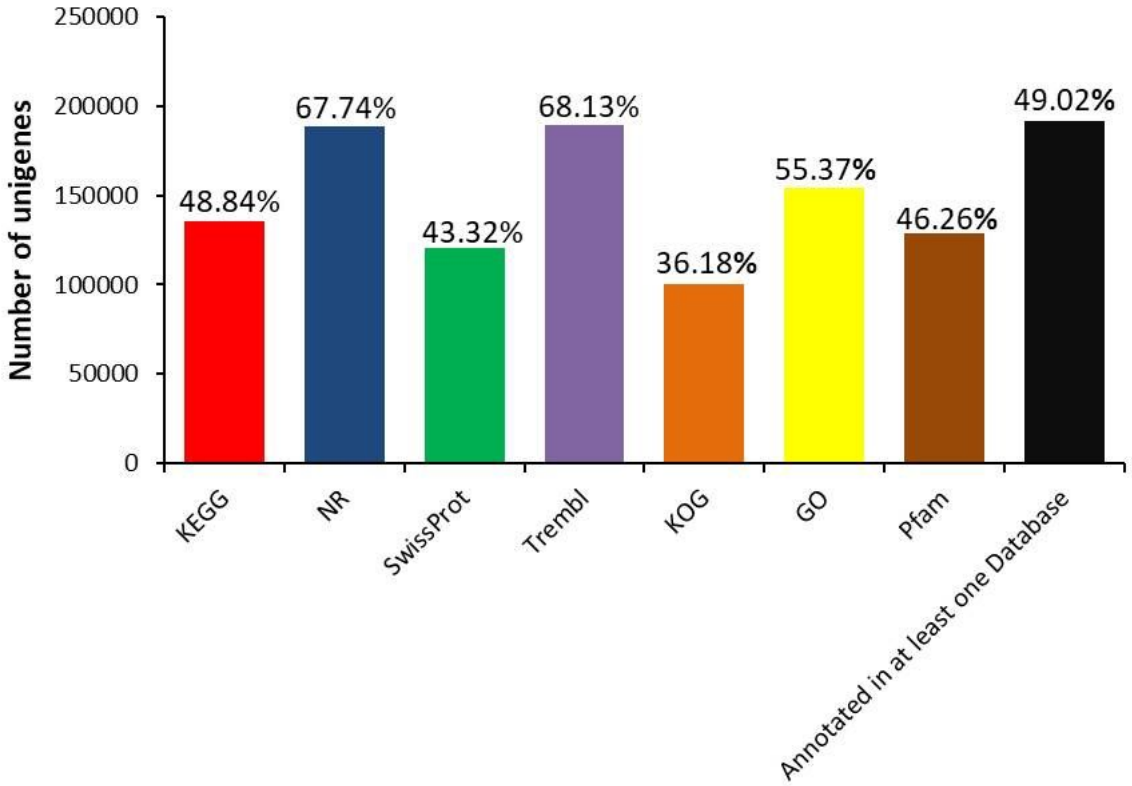

Figure 2

Unigenes matched in seven databases.

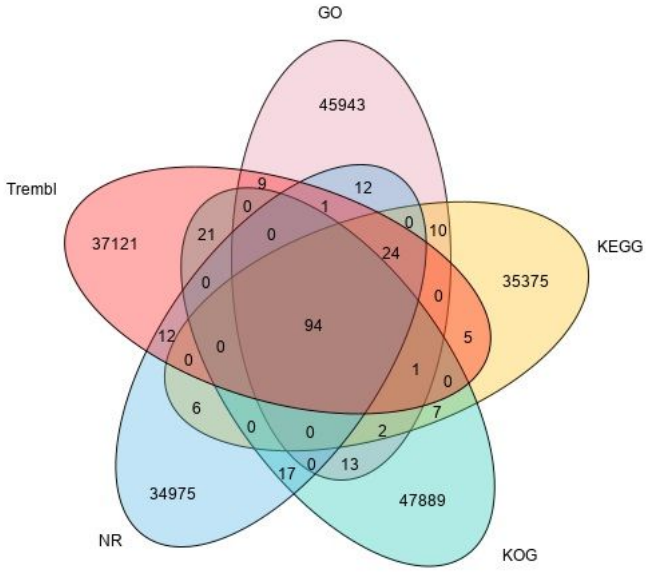

Figure 3

Venn diagram of differential BLAST results for Axonopus compressus. The number of unigenes annotated with 5 databasesand the co-annotated gene number are presented

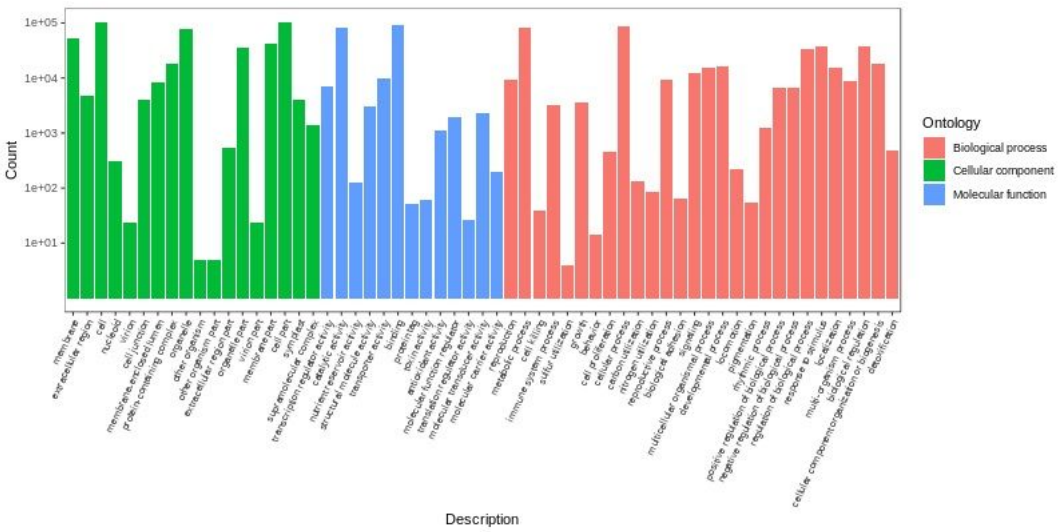

Figure 4

Functional classification of GO terms for Axonopus compressus.

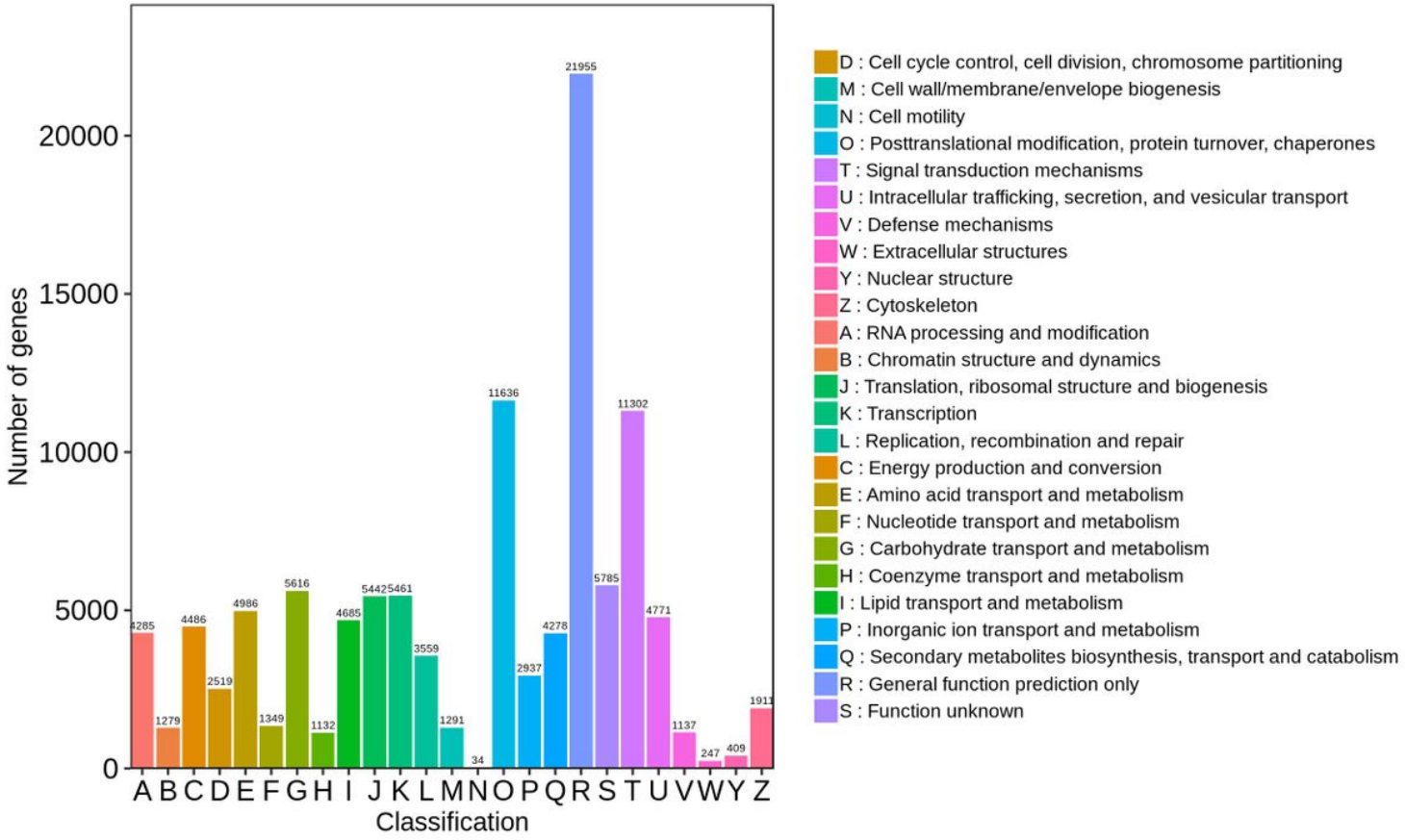

Figure 5

KOG functional classification of the Axonopus compressus transcriptome.

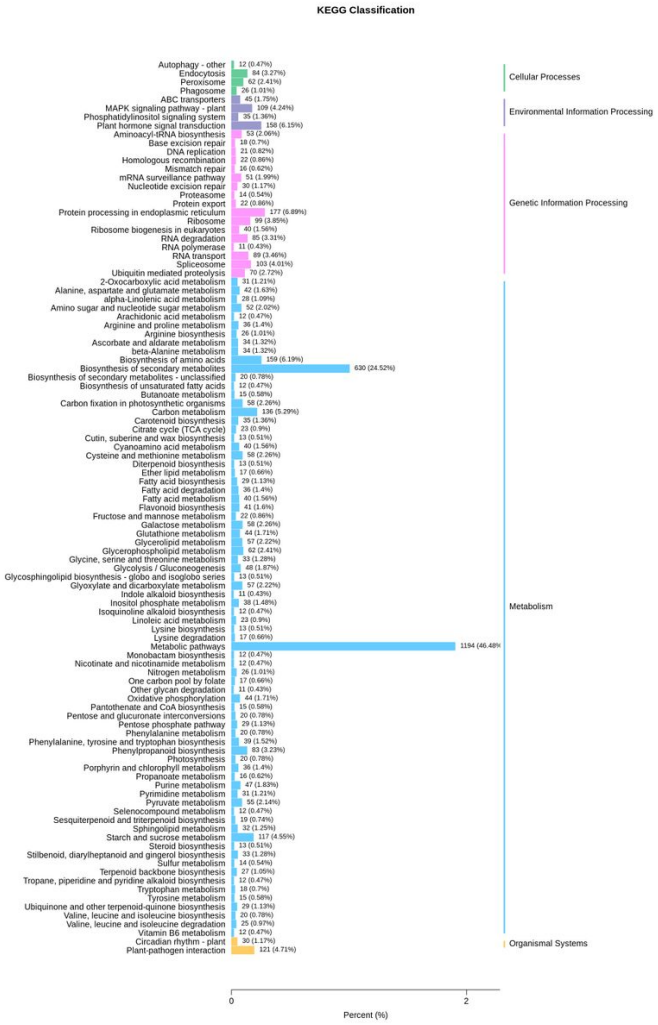

Figure 6

KEGG pathway annotation for Axonopus compressus.

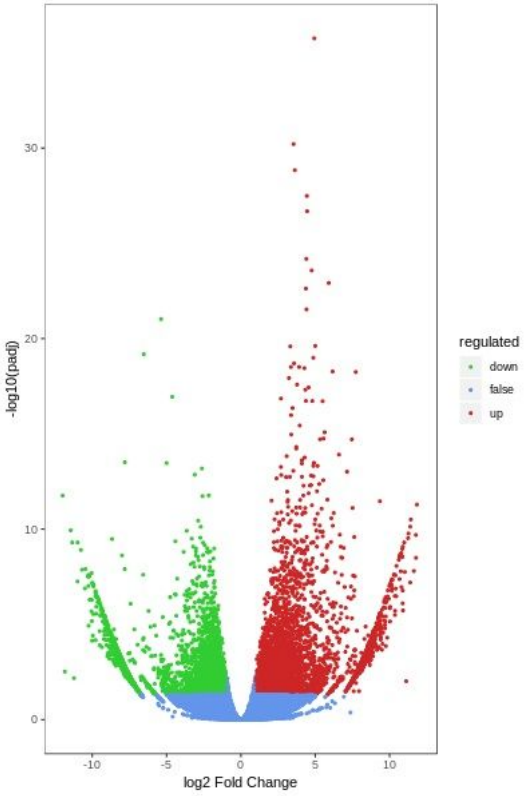

Figure 7

Volcano plot of differentially expressed unigenes of Axonopus compressus.

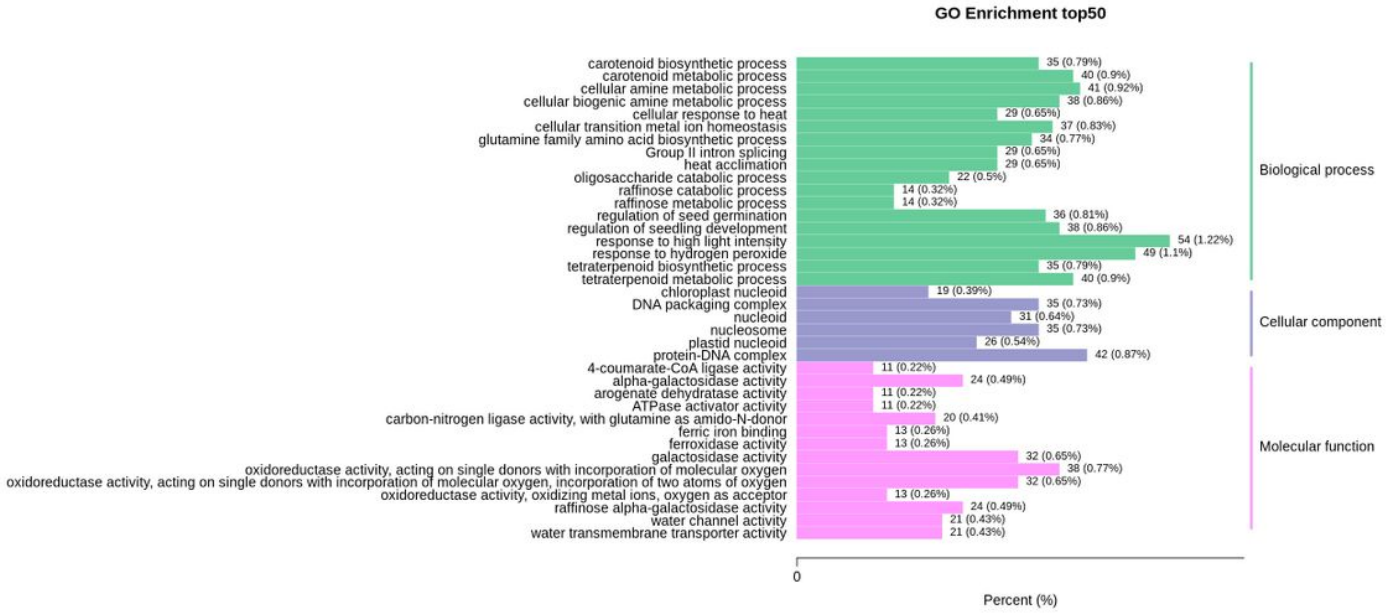

Figure 8

GO-enriched DEGs in Axonopus compressus.

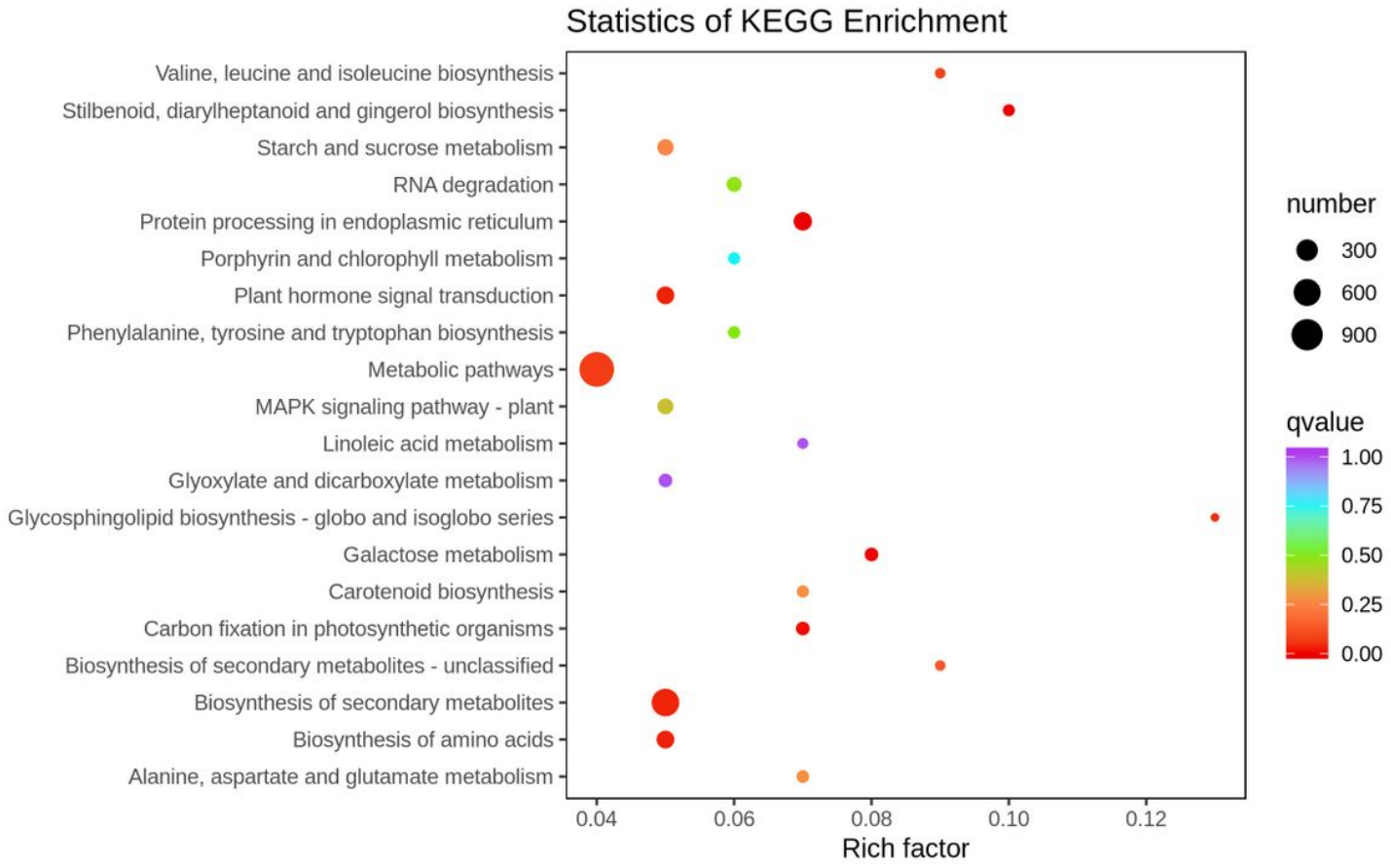

Figure 9

KEGG pathway enrichment of DEGs in Axonopus compressus.

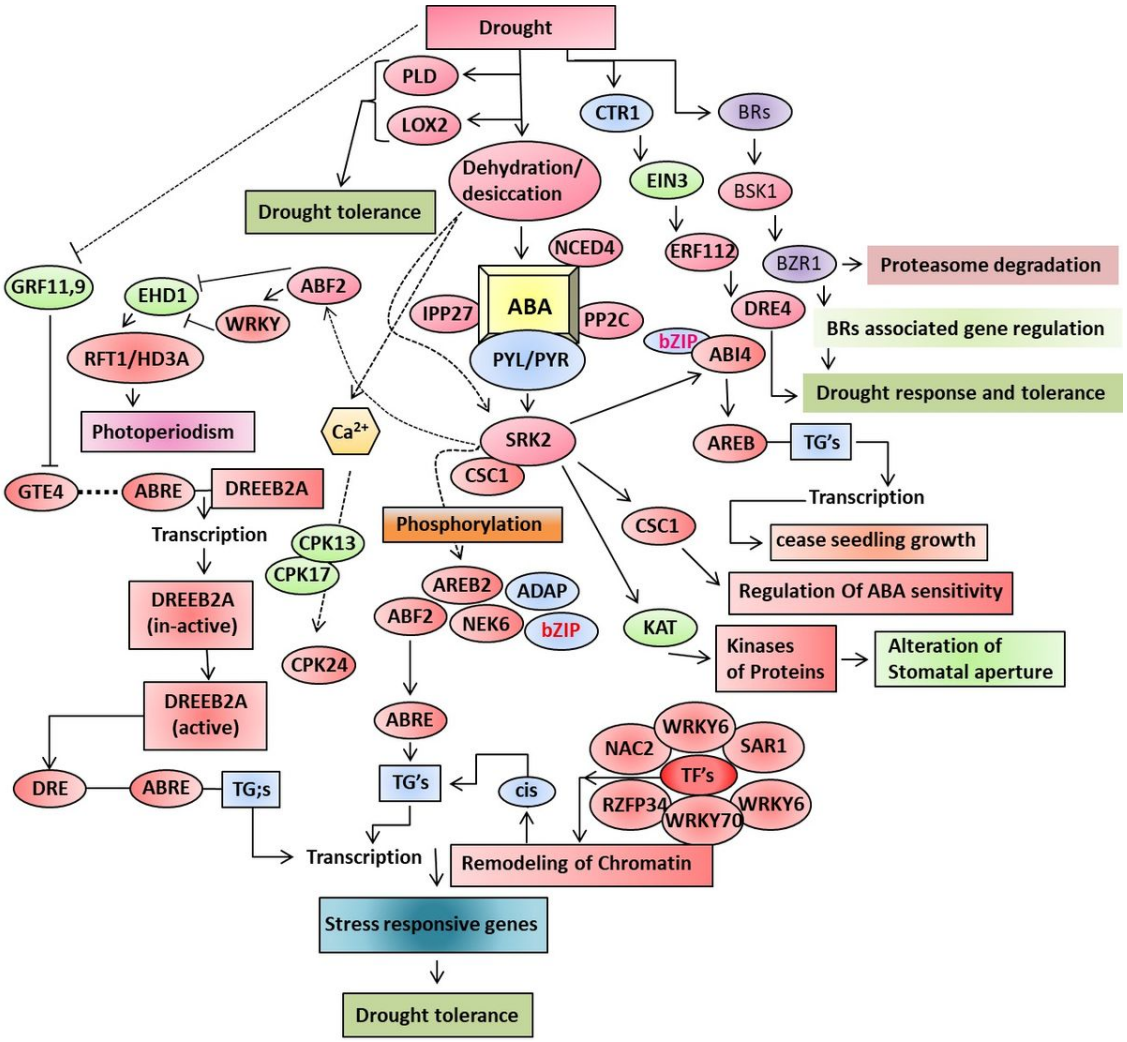

Figure 10

The qRT-PCR expression validation of drought-responsive genes in A. compressus. The bars showed relative expression of (A) NAC (B) MAP Kinase1, (C) MYB1, (D) PIP2, (E) WRKY1, (F) ABI5.

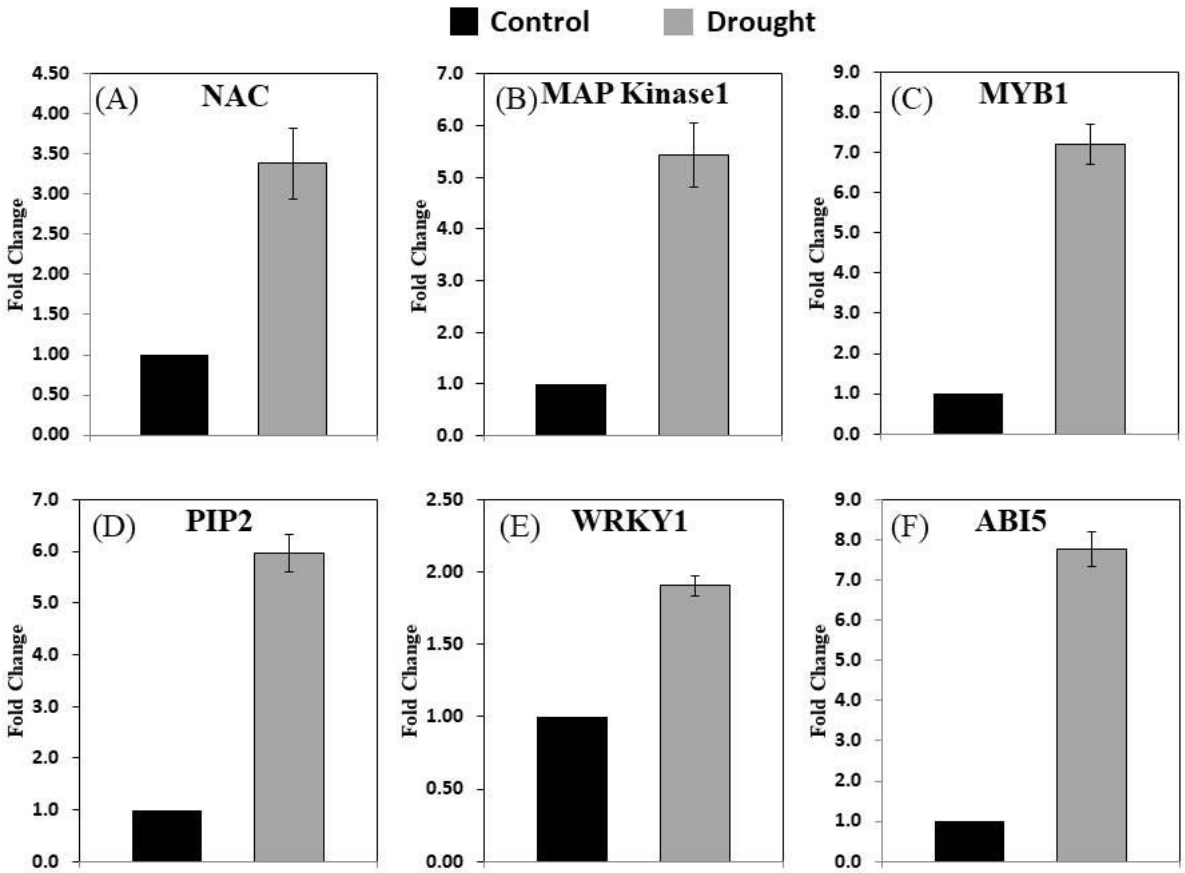

Figure 11

Schematic diagram of potential defense mechanisms for drought stress response and tolerance pathways in carpet-grass (Axonopus compressu L.).

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Additionalfile1.Metabolicpathwayanalysiskegg.xlsx
- Additionalfile2.Differentially.xlsx
- Additionalfile3.GOclassification.xlsx
- Additionalfile4.GOenrichment.xlsx
- Additionalfile5.KEGGpathway.xlsx
- Additionalfile6.Transcriptionfactors.docx
- Additionalfile7.Transcription.docx
- Additionalfile8.Listoftheprimersused.docx