

Figure S1 ABA-insensitivity phenotype of ZMAD2 showed better drought tolerance. The phenotype of water loss in isolated alfalfa leaves and its water loss rate. **A** Phenotype of ZMAD3 before and 18 d after drought treatment and 3 d after rehydration treatment. Scale bars indicate 10 cm. **B**, **C** Leaf RWC and Leaf electrolyte leakage of ZMAD2 materials before and 18 d after drought treatment. The asterisk\* indicates that the difference between S1-0 and S1-50 is statistically significant ( $P < 0.05$ ).

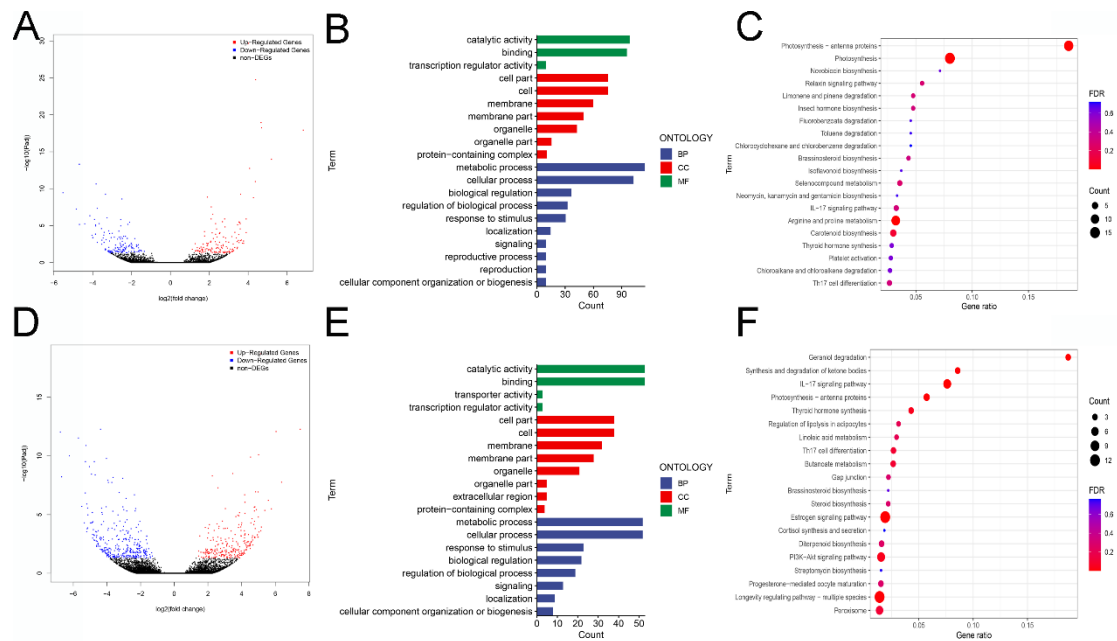


Figure S2 Leaf transcriptome analysis of S1-0 and S1-50 plant subjected to a drought stress treatment. **A** Volcano plots of up- and down-regulated DEGs in S1-0 before and after drought stress. **B** GO annotation of total DEGs in S1-0 before and after drought stress. **C** KEGG enrichment analysis of all DEGs in S1-0 before and after drought stress. **D** Volcano plots of up- and down-regulated DEGs of S1-50 before and after drought stress. **E** GO annotation of all DEGs in S1-50 before and after drought stress. **F** KEGG enrichment analysis of all DEGs in S1-50 before and after drought stress. The enrichment factor indicates the ratio of the proportion of DEGs in the pathway to the proportion of all genes annotated as the pathway. Blue dots in **a** represent genes with down-regulated expression, and red dots represent genes with up-regulated expression, the larger the enrichment factor, the darker the color, and the higher the confidence of the pathway results.

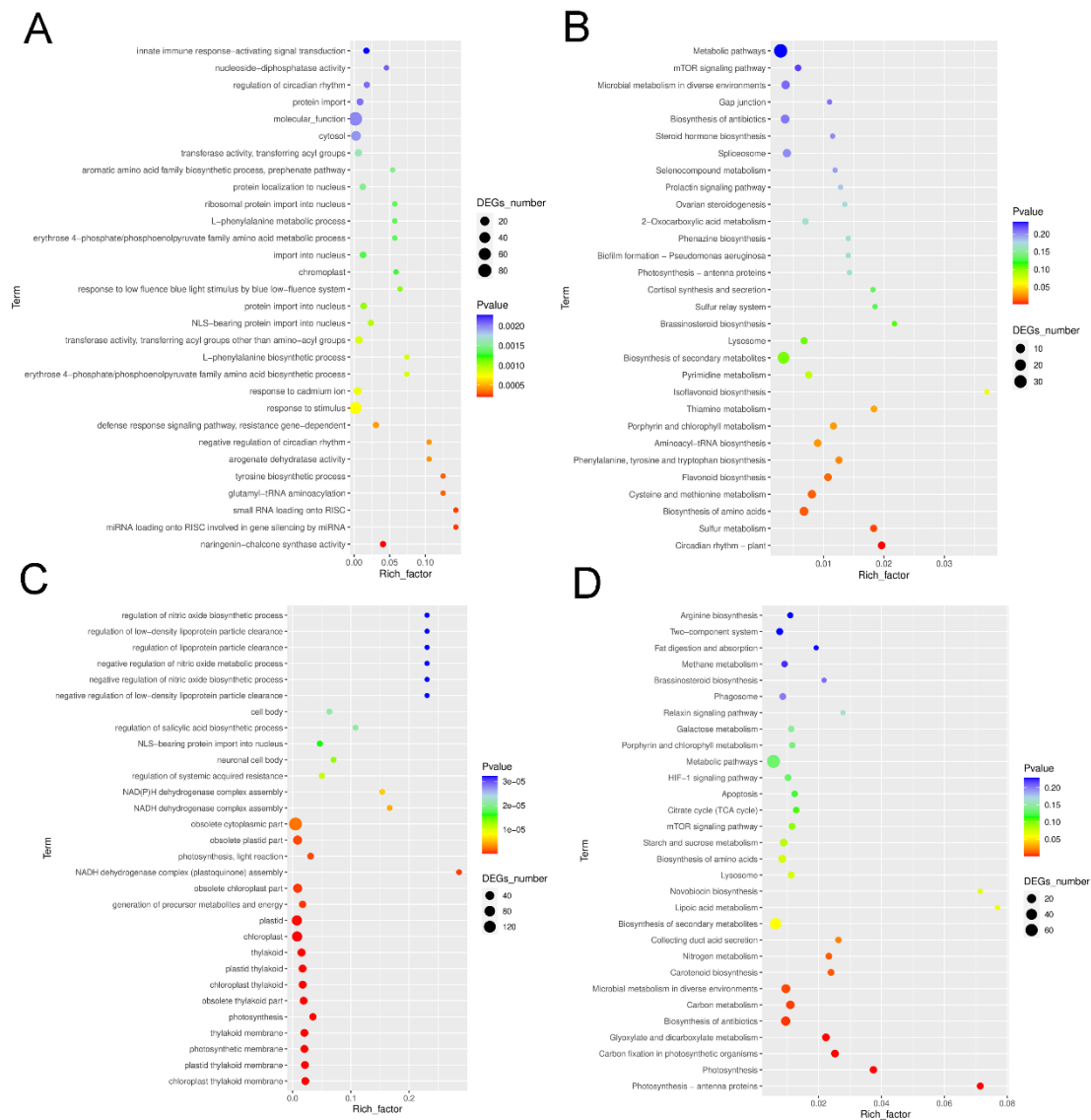


Figure S3 Transcriptome analysis before and after drought treatment of S1-0 compared to S1-50. **A** GO annotation of total DEGs in S1-0 compared to S1-50 before drought stress. **B** KEGG enrichment analysis of total DEGs in S1-0 versus S1-50 before drought stress. **C** GO annotation of total DEGs in S1-0 vs. S1-50 after drought stress. **D** KEGG enrichment analysis of total DEGs in S1-0 vs. S1-50 comparisons after drought stress.

Table S1 ABA Synthesis and Signal Transduction and Wax Synthesis Pathways Schematically Occurring Genes and their FPKM values.

Gene	Gene ID	S1-0	S1-50	DS1-0	DS1-50
CER	MS.gene046442	0.596667	5.573333333	0.1	8.883333
WSD	MS.gene92380	0	20.18333333	0	12.74
PYS	MS.gene69489	0.71	11.16333333	0	7.123333333
PYS	MS.gene22206	1.23	4.11	0.073333333	1.816666667
PYS	MS.gene46942	16.17	28.26	2.11	16.05
NCED	MS.gene062834	0.49	0.22	10.14	0.623333333
NCED	MS.gene063446	0.966666667	1.176666667	19.98	1.736666667
NCED	MS.gene039190	1.126666667	1.383333333	17.19333333	2.793333333
ABA2	MS.gene28847	79.87666667	67.8	5.886666667	30.25666667
AOG	MS.gene21268	0.653333333	2.913333333	0.873333333	6.6
PYR/PYL	MS.gene33451	16.70666667	4.97	0.266666667	0.516666667
PYR/PYL	MS.gene052795	7.68	3.303333333	0.496666667	0.943333333
PYR/PYL	MS.gene67442	7.403333333	14.3	0.48	0.493333333
PYR/PYL	MS.gene99506	16.95666667	16.24666667	2.03	4.416666667
PP2C	MS.gene046807	1.193333333	0.346666667	32.41333333	12.17666667
PP2C	MS.gene60356	1.38	0.713333333	65.64666667	12.61333333
PP2C	MS.gene049073	0.566666667	0.886666667	14.03666667	10.42
PP2C	MS.gene83625	4.303333333	0.64	0.19	0.176666667
PP2C	MS.gene07063	55.67	40.40666667	6.376666667	14.9
SnRK2	MS.gene89893	6.41	6.41	0.276666667	0.333333333
SnRK2	MS.gene041130	20.77333333	12.90333333	1.526666667	1.81
NADPH	MS.gene037818	5.75	0	6.11	0.743333333
NADPH	MS.gene38173	21.49333333	0.556666667	1.78	0
ABI5	MS.gene012975	0	0	4.61	2.776666667

Table S2 The primers used in the study.

Gene ID	Forward primer sequence	Reverse primer sequence
MS.gene062834	CTTCTTCGACGGAGACGGTATG	TGAAGCGGCAAGAGTAACTGAC
MS.gene33451	TACCGTTCCGTTACTACACTTCATC	CACCACATAGGACTCAACTATCACC
MS.gene99506	CGACAATCCACAAGCCTACAAAC	CGGAGATGACACGGACTTCAC
MS.gene049073	GCCGTATTTACTAATGGATTTGAGAGG	AACCAGATAGAACAACCACAGAGG
MS.gene83625	TATACATAATCAATCCTCGCAATCTTCTG	ATATTATCATCAACCGTGACAGTATTCG
MS.gene07063	ACGGTTGTGACTGTGAGGATTG	TGGATATGGAGAGCCTTTGATTGG
Actin	CAAAAGATGGCAGATGCTGAGGAT	CATGACACCAGTATGACGAGGTCG