

# Comparative Transcriptome Analysis of Resistant & Susceptible alfalfa (*Medicago sativa* L.) Cultivars After Pea Aphid (*Acyrtosiphon pisum*) Infestation

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The pea aphids (*Acyrtosiphon pisum*) are one of the most significant insect pests of alfalfa (*Medicago sativa* L.) and severely limit the alfalfa production. A possible solution is to use the resistant alfalfa cultivar. However, the underlying mechanisms of alfalfa resistance to the pea aphids are poorly understood. In this study, RNA-seq was used to conduct transcriptomics analysis on two alfalfa cultivars viz. a resistant (R) (Gannong-5) and a susceptible (S) (Zhongmu-1) after infested with pea aphids for 0, 12, 24 and 72h. Protein folding and regulation of biological quality were the prominent enriched GO terms in the resistant cultivar after aphid infestation. KEGG pathway analysis indicated that the differentially expressed genes (DEGs) in the resistant and susceptible cultivar comparison before aphid infestation are involved in linoleic acid metabolism which related to insect-resistant. In addition, glycolysis/gluconeogenesis, flavonoid biosynthesis and glutathione metabolism were significantly enriched in the 24h post-infested and photosynthesis-antenna proteins and starch and sucrose metabolism in the 72h post-infested, which were linked to plant resistance traits. Time course sequencing (TCseq) analysis identified nine and twelve clusters with similar expression change characteristics in resistant and susceptible cultivar respectively. The increased direct and indirect defense induced by pea aphid feeding were correlated with glutathione metabolism, linoleic acid metabolism, glycolysis/gluconeogenesis, etc. These findings will improve our understanding about the underlying mechanisms of alfalfa cultivar resistance response to pea aphids feeding and will provide the insights for further research on plant defense against aphids.

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