# Comparative Gene Expression Profiles between Heterotic and Non-heterotic Hybrids of Tetraploid *Medicago sativa*

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We have previously shown that crosses between the sativa and falcata alfalfa germplasm pools can result in progeny that express heterosis for biomass yield. In order to begin to understand the genetic basis of heterosis, we conducted an experiment to test the hypotheses that hybrids expressing heterosis for biomass yield would show more nonadditive gene expression and more over dominance of expression patterns than a hybrid that does not exhibit heterosis. We tested these hypotheses with three *Medicago sativa* genotypes—one falcata and two sativa—and their three hybrid populations; the two sativa x sativa crosses produced heterosis for biomass yield and the sativa x sativa cross did not. We analyzed gene expression on Affymetrix *M. truncatula* GeneChip arrays, using RNA from plants grown in a growth chamber. Alfalfa hybridized to approximately 47% of the *M. truncatula* genes. Using the Affymetrix MAS5.0 software to analyze the data, the two heterotic hybrids performed similarly, with about 27% of genes showing differential expression compared to 12.5% for the non-heterotic hybrid. At a false discovery rate of 0.15, 4.7% of differentially expressed genes in hybrids (~300 genes) showed nonadditive expression compared to only 0.5% (16 genes) in the non-heterotic hybrid. Of the nonadditively expressed genes, approximately 50% showed over dominance expression in heterotic hybrids; only 1 of 16 showed a similar profile in the non-heterotic hybrid. Genes whose expression differed in the parents were three times more likely to show nonadditive expression than genes whose parental transcript levels were equal. The general pattern of results was the same when we used RMA software to analyze the data; however, the genes expressing overdominance were largely different between the two analysis methods. Based on the results, we suggest that nonadditive expression of transcript levels may contribute to heterosis for biomass yield in alfalfa.