Using gene expression patterns as signatures of a biological response reveals the rheostat-like nature of brassinosteroid signaling

Amanpreet Kaur^{1,2}, Norman Best³, Brian Dilkes^{1,2}

¹Department of Biochemistry, Purdue University, West Lafayette, IN; ²Center for Plant Biology, Purdue University, West Lafayette, IN; ³USDA-ARS, Columbia, MO

Plants respond to external or internal stimuli by reprogramming gene expression to elicit a physiological or biochemical response. Transcript profiling captures these changes as gene expression changes affected by a specific phenotype or treatment. The resulting data can be analyzed to uncover signaling mechanisms underlying the phenotype or treatment. However, identifying differentially expressed genes does not provide a comprehensive view of pathway level effects, quantify the response, or detail the phenotypic impacts. We propose that analyzing coordinated transcript abundance changes in a group of genes can provide this insight. Incorporating prior knowledge of known effects on gene sets provides biological context to gene expression consequences and facilitates the identification of links between gene expression and phenotype. Here, we demonstrate a gene set testing approach to assess the status of brassinosteroid (BR) hormone signaling. The transcriptional consequences of loss of BRs were analyzed by RNA-sequencing of the *brd1* mutant. Genes responsive to BR treatment were obtained from publicly available datasets of BR-treated maize seedlings. The expression of BR induced genes was lower in brd1 while BR suppressed genes were increased in brd1, consistent with loss of BR response in the mutant. To complement this gene set testing, we created a summary statistic called an "index" that aggregates the response of maize genes to BR excess and thus serves as a reporter of BR signaling status. This index calculation turned a set of differentially expressed genes responding to BR excess into a single reporter of BR signaling status, that was utilized to assess BR signaling in our RNA seq samples. BR indices revealed opposing effects on gene expression in BR-treated and BR-deficient samples and index values could assign function to a variety of BR signaling and biosynthetic mutants, demonstrating a rheostat-like nature of BR signaling in maize.