

Introduction & Background

Portulaca oleracea, more commonly known as purslane, is a weedy medicinal plant which contains high levels of α -linolenic acid, a polyunsaturated fatty acid important in health and nutrition. We have been using purslane as a model for plants under the stress of climate change, which predicts increased temperatures, longer periods of drought, and more saline soils. Studying how plants respond to stress on a molecular level will enable us to focus on genotypes that have enhanced stress response capacities.

Experimental Design & Methods

We performed a $G \times E$ experiment in which we assessed the interactions between the genotype and environment. Two varieties of purslane, a commercially available Tall variety (T-16) as well as a wild strain collected in Wisconsin (WI-9), were studied. 120 plants of these varieties were planted and grown for 2 weeks with plain water. At that point, the plants of each genotype were divided into two groups receiving two salinity treatments: 200 mM NaCl solution and water containing no salt. Each combination of treatment and genotype contained 20 plants. After 3 weeks of treatment, their morphological and chemical changes based on the treatment they received were measured and analyzed.



WI-9 purslane stressed (right) and unstressed (left)

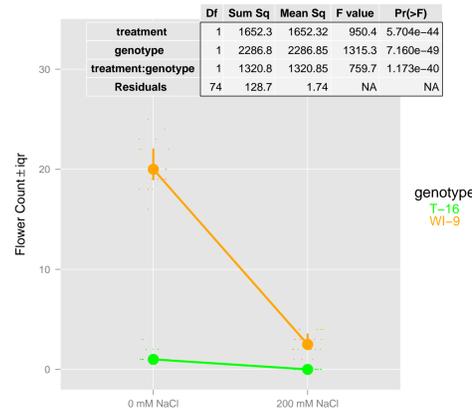


T-16 purslane stressed (left) and unstressed (right)

Flowers

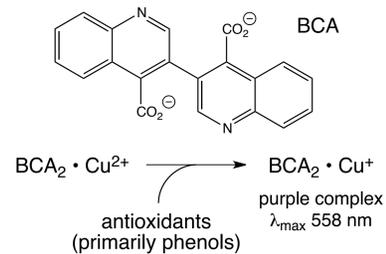
Morphological data were collected on a variety of factors including nodes, flowers, branches and dropped leaves as measures of reproductive fitness. The plants' production of flower structures gave the most dramatic response and clearly showed a $G \times E$ interaction.

Fitness As Measured By Flower Count

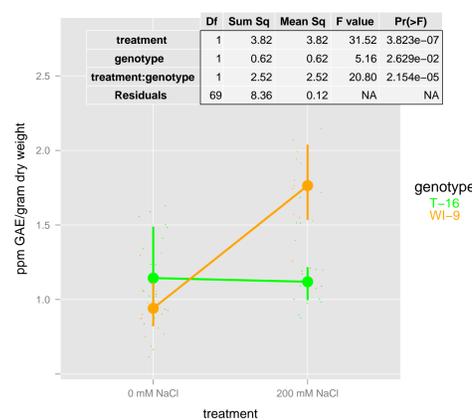


Total Antioxidants

Antioxidants present in each sample were found the BCA assay which measures the levels of antioxidants in a plant as gallic acid equivalents (GAE). Antioxidants are produced in response to stress. A $G \times E$ interaction was identified as the WI-9 genotype increased significantly while the T-16 genotype remained relatively unchanged.



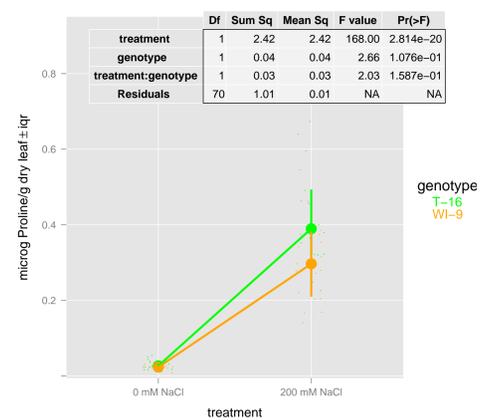
Total Antioxidants



Proline

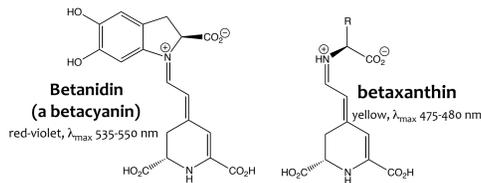
Proline, a common amino acid that acts as an antioxidant and osmoprotectant, is produced by plants when they are under stress. A calibration curve was used to calculate the amount of proline present in each sample. The proline assay involved a reaction with ninhydrin to produce a yellow adduct. For both genotypes, the levels of proline increased significantly when stressed, but the $G \times E$ interaction was not significant.

Proline Levels



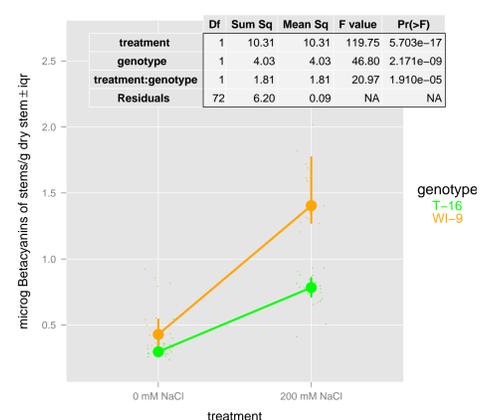
Stem Betacyanins

Betalain pigments, betaxanthins and betacyanins, are produced by purslane. Betaxanthins, typically found in flowers, are yellow while betacyanins, usually found in stems, are a red-purple color.



The amount of both betalain pigments were found for leaves and stems, respectively. Stem betacyanin data are shown below. The distinctive slopes indicate another $G \times E$ interaction.

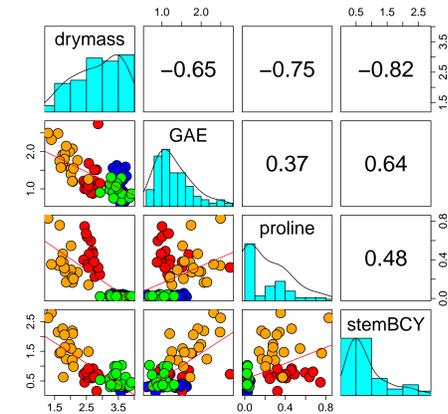
Betacyanins of Stems



Correlation Amongst Variables

The plots below show the correlations between independent variables with respect to salinity stress.

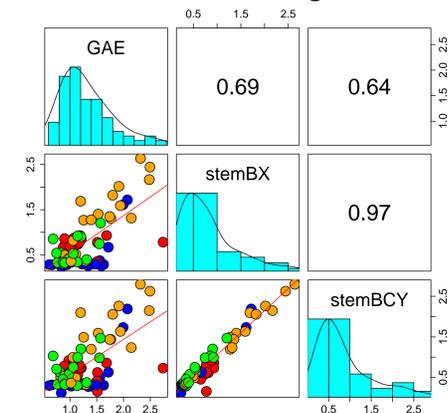
Fitness vs Antioxidants



Key: red = T High Salt, orange = WI High Salt, blue = T Low Salt, green = WI Low Salt
stemBCY = stem betacyanins
stemBX = stem betaxanthins

As stress increased, we observed a decrease in above ground dry mass (shown above). A similar trend was observed with flowers (not shown). All other variables correlated positively with increased stress.

GAE vs Betalain Pigments

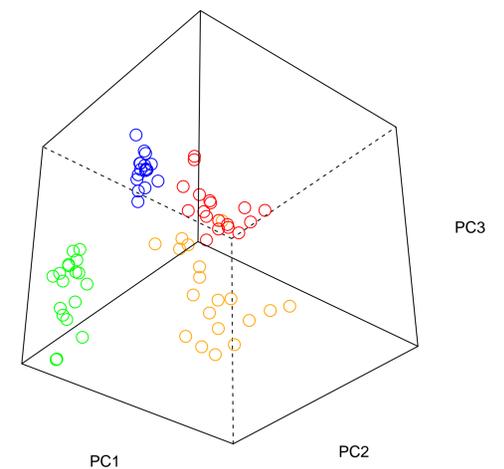


key as above

We observed a strong positive correlation between the betaxanthins and betacyanins in the stems (see above), as these betalain pigments have a common biosynthetic pathway. Additionally, there is a positive correlation between levels of antioxidants and betalain pigments in the stems of purslane.

Principal Component Analysis

Principal Component Analysis (PCA) is a dimensional reduction method that merges correlating variables into new independent variables, the principle components (PCs). These PCs describe as much variance as possible, the essence of the data, without noise or correlation with other PCs.



The separation on the PCA plot of the four different combinations of treatment and genotype shows the significance of the combination of treatment and genotype. This implies that the genotypes have different molecular pathways and respond differently to salinity stress on a molecular level.

References

- L. S. Bates, R. P. Waldren, and I. D. Teare, "Rapid determination of free proline for water-stress studies," *Plant and Soil*, vol. 39, pp. 205 - 207, 1973.
D.C Marino, L. Z. L. Sabino, J. A. Jr., A. D. A. Ruggero, and H. D. Moya, "Analysis of the polyphenols content in medicinal plants based on the reduction of Cu(II)/bichononic complexes," *Journal of Agricultural and Food Chemistry*, vol. 57, pp. 11061-11066, 2009.

Acknowledgments

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