Effects of Stress on the Medicinal Plant *Portulaca oleracea*: Focus on the Production of Betalain Pigments

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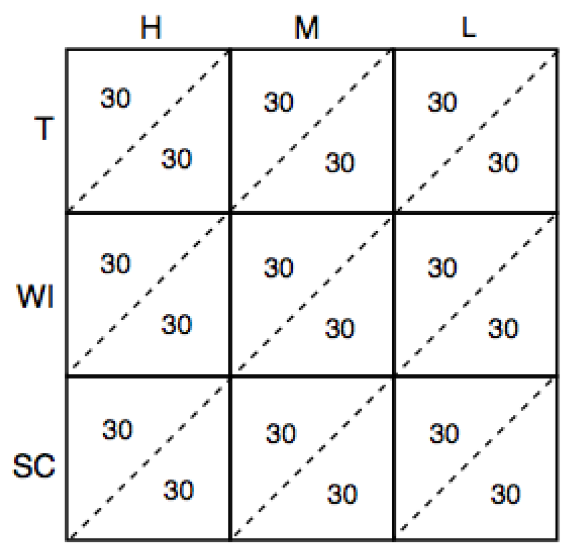
Introduction

Purslane is a weedy medicinal plant known to withstand extreme environmental conditions. It's production of chemically interesting secondary metabolites, such as betalains and other phenolic antioxidants, make it desirable to study. Under stress, plants will increase production of these secondary metabolites in order to survive. We have been investigating the response of purslane to both salinity and drought stress in order to chemically and morphologically quantify the effects that harsh environments have on the plant. Studying purslane provides new insight on the response of plants during environmental change, perhaps even global warming.

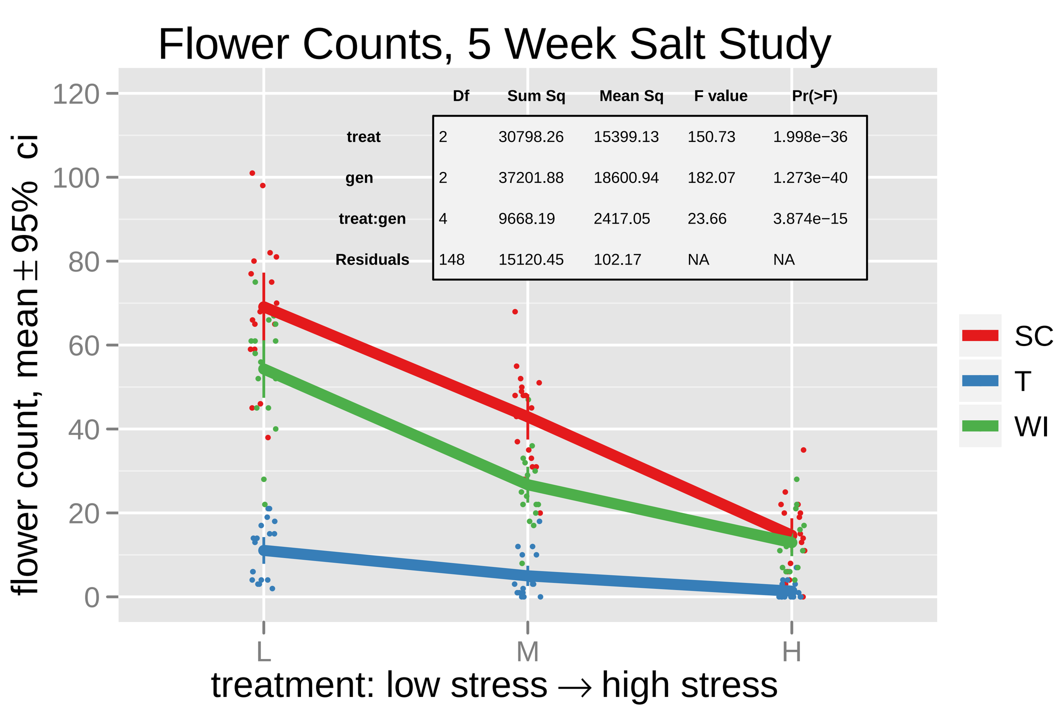
Experimental Design

1,080 plants of three purslane genotypes were planted at the beginning of the summer. Each genotype received three varying treatments (high, medium, and low) for salt or water. Samples were collected at an intermediate stage (weeks two and three), and a final stage (weeks five and six). The graphic below portrays the experimental design.



Morphological Parameters

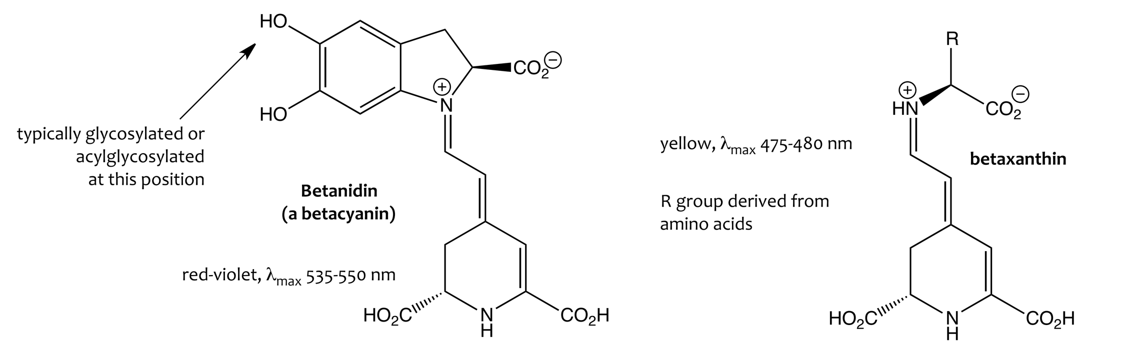
In all studies, numerous morphological parameters were measured each time plants were harvested. Among these, the flower count was the most reliable measure of plant fitness and stress. A typical set of data is shown below. Other parameters measured but not shown were node count, branch count, above ground mass, fraction of water in tissues and the number of dropped leaves.



	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treat	2	30798.26	15399.13	150.73	1.998e−36
gen	2	37201.88	18600.94	182.07	1.273e−40
treat:gen	4	9668.19	2417.05	23.66	3.874e−15
Residuals	148	15120.45	102.17	NA	NA

Betalains & Plants Under Stress

Betalains are colored pigments synthesized in purslane tissues. Betalains are classified in two main catagories : betaxanthins and betacyanins. Betaxanthins are yellow-orange pigments, usually present in flowers. Betacyanins are the red to violet pigments typically seen in purslane stems, especially when the plant is under stress. Typical betaxanthin and betacyanin structures are shown below.

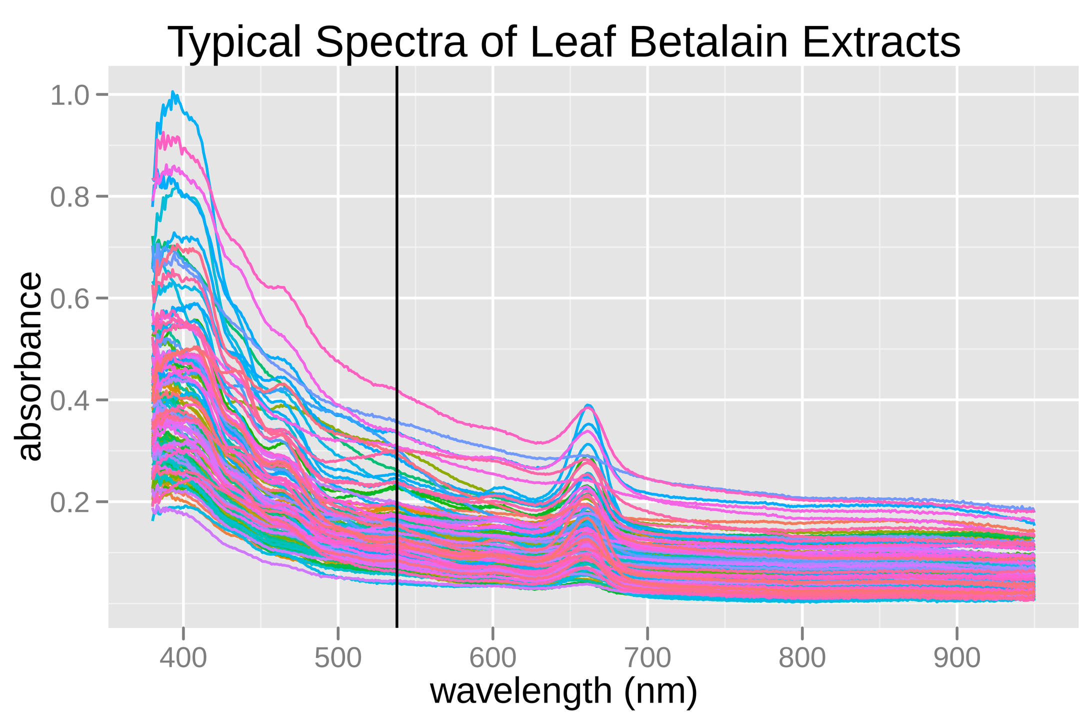


Below is a photo of a low stress (control) plant at 6 weeks (left). The picture on the right is a high stress drought treatment plant at six weeks. The increased production of betalains is apparent in the red color of the stem.


(a)

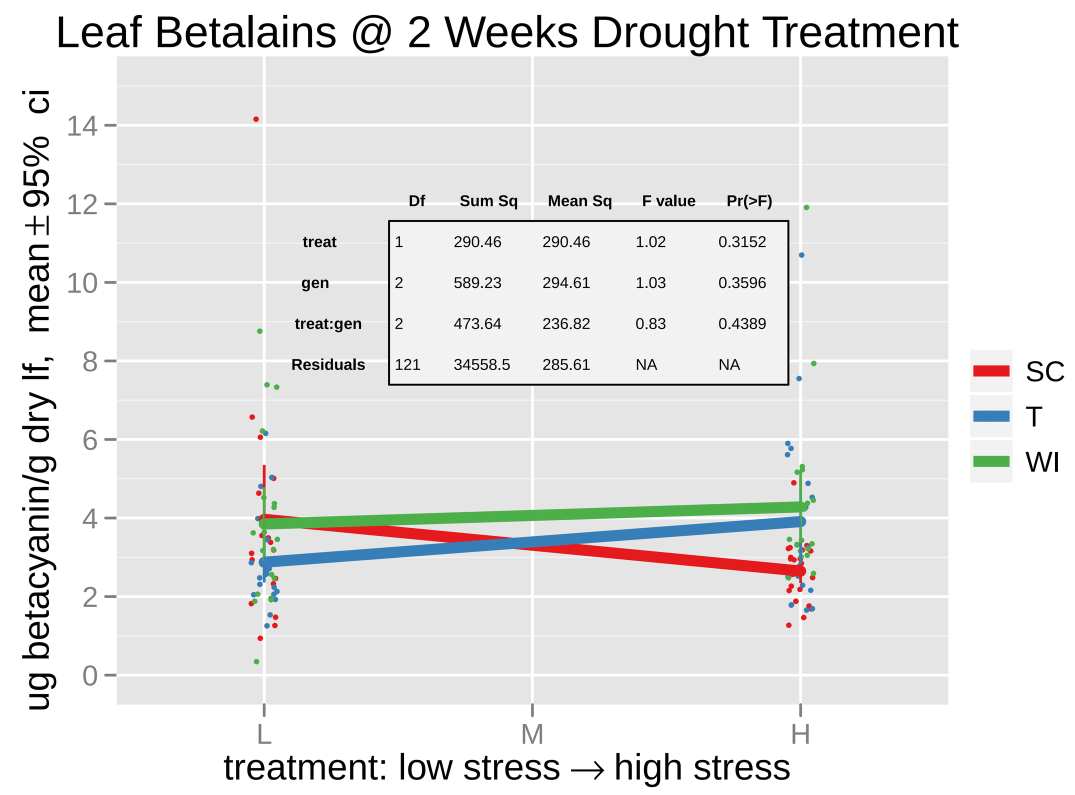

(b)

Raw Leaf Betalain Spectra



This figure depicts the raw leaf betalain spectra from the drought study plants at 3 weeks. For betacyanins, the λ_{max} is at 538 nm. One problem encountered during analysis is the appearance of chlorophyll in the spectra, which is co-extracted with the betalains. This interferes with a full analysis of all betalains.

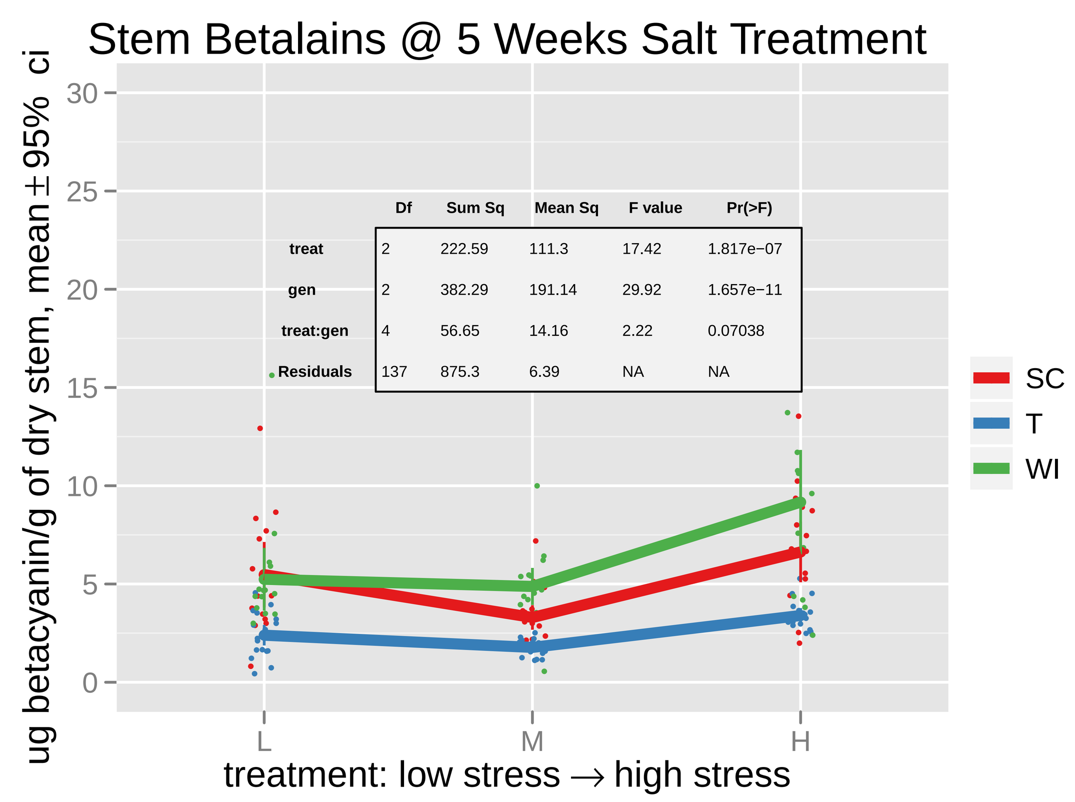
Leaf Betalains at Two Weeks Drought Treatment



	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treat	1	290.46	290.46	1.02	0.3152
gen	2	589.23	294.61	1.03	0.3596
treat:gen	2	473.64	236.82	0.83	0.4389
Residuals	121	34558.5	285.61	NA	NA

After two weeks, betalain levels in purslane leaves were unaffected by the lack of water. This may indicate that a threshold may be present and that a longer treatment period is required to show an effect. Further, this was a study of betacyanin in the leaves, but most of the obvious pigment change occurred in the stems, which were studied during the 5 and 6 week trials.

Stem Betalains at Five Weeks Salt Treatment



	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treat	2	222.59	111.3	17.42	1.817e−07
gen	2	382.29	191.14	29.92	1.657e−11
treat:gen	4	56.65	14.16	2.22	0.07038
Residuals	137	875.3	6.39	NA	NA

At five weeks, the betalain content of stems was analyzed. The figure above indicates that purslane responded to salinity stress by increasing betalain production in the stem tissue in a manner consistent with the idea of a threshold effect. Both genotype and treatment factors were statistically significant.

Acknowledgements

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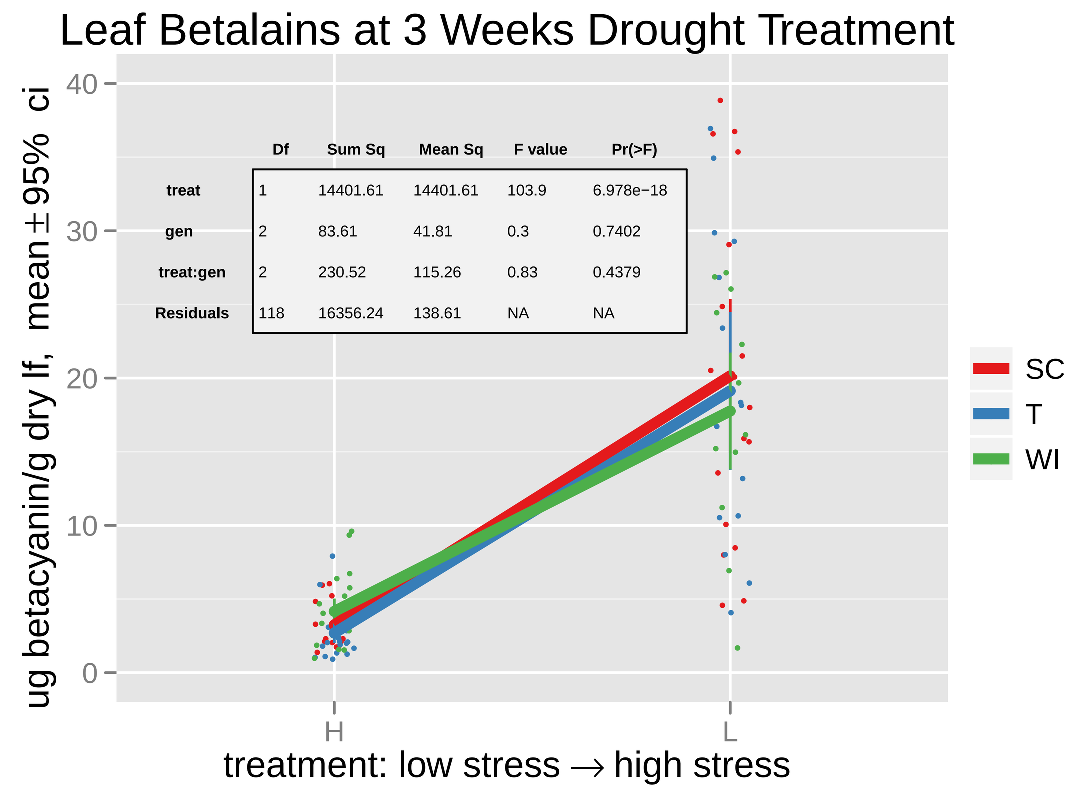
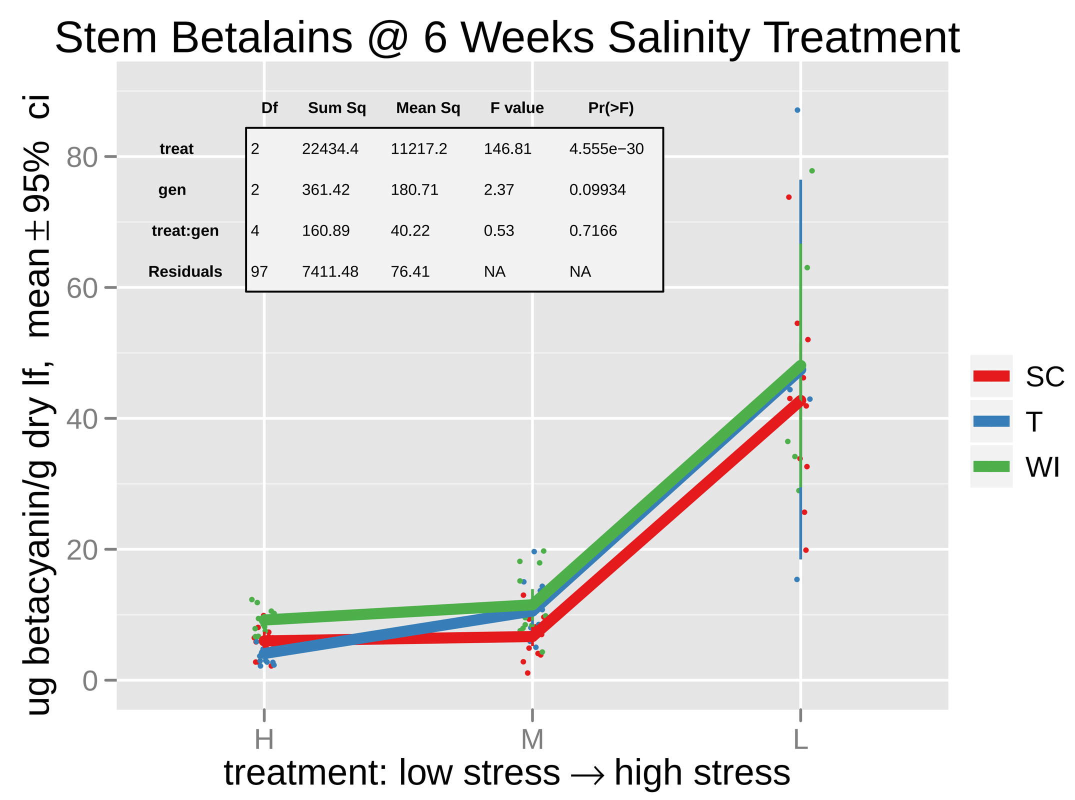
Leaf Betalains at Three Weeks Drought Treatment



	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treat	1	14401.61	14401.61	103.9	6.978e−18
gen	2	83.61	41.81	0.3	0.7402
treat:gen	2	230.52	115.26	0.83	0.4379
Residuals	118	16356.24	138.61	NA	NA

After three weeks, the plants in the drought study showed a dramatic response to the drought stress across all genotypes, but statistical significance was limited the the treatment factor.

Stem Betacyanins at Six Weeks Salt Treatment



	Df	Sum Sq	Mean Sq	F value	Pr(>F)
treat	2	22434.4	11217.2	146.81	4.555e−30
gen	2	361.42	180.71	2.37	0.09934
treat:gen	4	160.89	40.22	0.53	0.7166
Residuals	97	7411.48	76.41	NA	NA

At six weeks, the stems were again analyzed. The figure above shows a similar but amplified trend compared to the short salinity study. The treatment factor is statistically significant.

Conclusions

In response to stress, all three purslane genotypes increase the production of betalains. The increased betalain production was most clear in the five and six week trials. Although there was little response in the low and medium treatments, plants in high treatments responded by producing high levels of betacyanins, especially at longer treatment periods.