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Spectral reflectance indices and their relationship with dry mater production in an association mapping population of white clover

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Introduction

Increase yield and yield-stability under abiotic stress conditions (cold, drought, among others) is a global challenge in the breeding of perennial forage species, due to the new climatic scenarios imposed by the global changes. The modern breeding offering tools that allow for manipulating the genotype/phenotype relationship (association mapping analysis) and to accelerate the genetic gain rates for traits with low heritability and high genotype x environment interaction. The next-generation sequencing methodologies allow for characterizing, in short time period and low cost, complex genomes as the white clover. Nowadays, modern breeding is demanding high-throughput phenotyping methodologies that allows for characterizing a high number of genotypes (>100), in short time periods (hours), and reasonable cost (Inostroza *et al*., 2015). During plant growth, canopy can absorb, reflect, or transmit energy reaching the surface due to interaction of incident radiation with the plant structure and photosynthetic elements. By determining the spectral signature of canopy and leaf reflectance with a spectroradiometer, it is possible to indirectly measure agronomic and physiological traits (Hernandez *et al*., 2015). Indices based on reflectance at different wavelengths, known as spectral reflectance indices (SRIs), have been used for this purpose. The objective of this work was to study the relationship between dry mater (DM) production and 11 SRIs evaluated in a white clover association mapping population.

Materials and Methods

A white clover association mapping populations (WCAMP) was established in three environments that represent a winter cold gradient associated with the altitude: Santa Rosa (SR), Atacalco (AT) and Puente Marchant (PM), located at 140, 659 and 1054 metres above sea level, respectively. In each location, a plant spaced experiment was arranged in an alpha lattice experimental design with two replicates and 24 incomplete blocks (IB). Each genotype was planted in a row (IB) with 1m spacing and the distance between rows was 1m. The establishment was carried out during October-November 2013 and the experiment will be evaluated during three growing seasons (2013, 2014/15 and 2015/16). In all locations, the plants were irrigated through a pressurized irrigation system with 2 Lh^{-1} drip emitters. Periodically the weeds were controlled in a manual form. The DM production was evaluated through cuts 2 cm above ground level with an electric shearing machine (Oster, SHOWMASTER™, USA). During the first growing season the DM production was evaluated in only one cut three month after planting (summer DM accumulation). During the second growing season DM production was evaluated in three cuts in SR and AT (10-07-2014, 11-24-2014 and 02-02-2015) and two cuts in PM (11-25-2014 and 02-03-2015). The fresh samples were dried in a forced air oven at 65°C until constant weight. One day before the November and February cuts (second season) the spectral reflectance of plant canopy was measured using a portable spectroradiometer (FieldSpec®, HandHeld 2™, ASD, CO, USA). The spectroradiometer was mounted on a tripod to measure at a height of 25 cm; equipment sensor covered a circle of 12.5 cm diameter. Three spectra per plant were taken and averaged. Full spectrums consist of 725 narrow channels with 1 nm interval between 350 and 1075 nm. Measurements were made on clear days from 11.00 to 16.00 h. Periodic radiometric calibration was performed against a field reference panel (Spectralon, ASD). Information from reflectance was used to calculate two group of spectral reflectance indices (SRIs). The structural-SRIs are associated to plant morphology and architecture and Chlorophyll-SRIs are associated to biochemical and physiological traits (Table 1). All statistical analyses were performed for second season dataset with R-project 3.1.2 software (http://www.r-project.org/). Correlation analysis was generated between each SRI and DM production with corrplot R package. Also the spectral curves were normalized through a noise removal and moving average procedure using prospectr R package and then a partial least squares (PLS) regression was performed between DM production and all spectral bands (350-1075 nm) using pls R package.

Results and Discussion

In each location, the DM production showed a broad genetic variability (P<0.001). In SR, AT and PM the DM production varied between 25.8 to 557.3, 18.9 to 517.2 and 8.9 to 344.6 g plant^{-1}, respectively. The mean of total DM production was highest in SR (190.6 g plant^{-1}), followed for AT mean (175.6 g plant^{-1}) and PM mean (100.0 g plant^{-1}; Fig. 1). The correlation coefficient between DM and SRIs was highly significant (P<0.001; Table 1). The relationship varied between environments; In PM and SR the highest and lowest r values were observed, respectively. This was because of the differences in plant architecture, induced by the pattern of the irrigation water distribution of the drip irrigation system. In SR the soil had a higher natural superficial moisture that allows a higher ground level stolon elongation (prostrate growing pattern); whereas in AT and PM the soil moisture was limited to section cover by irrigation system (wet bulb) and the stolons showed a rather erect growing pattern. The PLS analysis showed a scarce relationship between DM production and the spectral reflectance matrix with R^2 values that varied between 0.15 and 0.20 (Table 1).

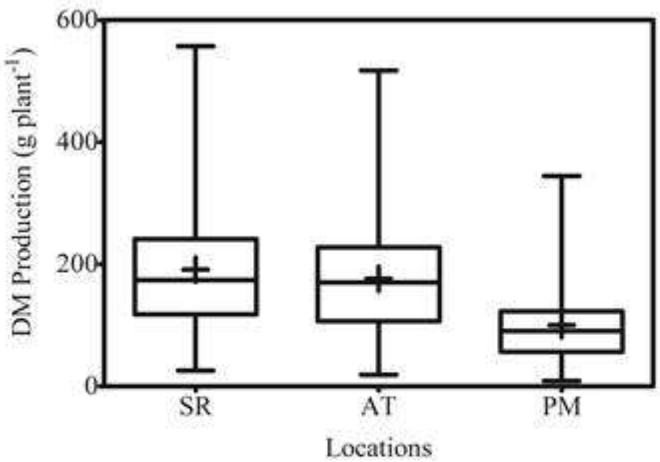

Fig. 1: Frequency distribution (box plot) for dry matter production of 192 white clover genotypes evaluated in three locations during 2014/15 growing season. Locations were Santa Rosa (SR), Atacalco (AT) and Puente Marchant (PM).

Table 1: Correlation coefficient (r value) of the relationship between dry mater production (DM production) and 11 spectral reflectance indices (SRIs) evaluated in a white clover association mapping population grown in three environments in Chile: Santa Rosa (SR), Atacalco (AT) and Puente Marchant (PM).

SRIs	Equation	DM production r value		
		SR	AT	PM
Chlorophyll Indices				
GM1	R750/R550	0.41***	0.57***	0.70***
GM2	R750/R700	0.39***	0.56***	0.69***
Lic1	(R800-R680)/(R800+R680)	0.29***	0.50***	0.52***
Vog1	R740/R720	0.44***	0.56***	0.70***
Vog2	(R734-R747)/(R715+R726)	-0.46***	-0.57***	-0.72***
Vog3	(R734-R747)/(R715+710)	-0.45***	-0.57***	-0.72***
WI	R970/R900	-0.34***	-0.56***	-0.71***
ZM	R750/R710	0.42***	0.56***	0.70***
Structural Indices				
NDVI	(R900-R680)/(R900 +R680)	0.34***	0.47***	0.42***
MCARI2	$\frac{1.5[2.5(R800-R670)-1.3(R800-550)]}{\sqrt{(2R800+1)^2-(6R800-5\sqrt{670})-0.5}}$	0.25***	0.37***	0.46***
MTCI	(R753-R708)/(R708-R681)	0.46***	0.51***	0.70***
PLS-All	DM production= Xβ + E	R^2=	R^2=	R^2=

**** For P<0.001 and ns= no significant. R is the reflectancia to a given wavelength.*

Conclusion

There is a statistically significant relationship between DM production and the 11 SRIs evaluated in the WCAM population. The relationship is highly influenced by environmental factors that induce changes in plant architecture.

References

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