High-Throughput Approach for Phenotyping Alfalfa (*Medicago sativa* L.) Yield

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INTRODUCTION

- Phenotyping plays a central role in plant breeding.
- High-throughput phenotyping (HTP) can be an alternative method to manual phenotyping to improve yield in alfalfa.

Fig. 1: A) Manual vs B) HTP phenotyping in alfalfa.

OBJECTIVES

i) Model herbage accumulation (HA) in alfalfa using vegetation indices (VIs)
ii) Fit univariate and bivariate models for HA and VIs to select alfalfa families with improved yield
iii) Estimate genetic gain for yield for univariate and bivariate models for HA and VIs

MATERIALS AND METHODS

- **Breeding population:** 145 full-sib and 36 half-sib families, and three controls 'Bulldog805' and 'FL99' and breeding line UF_AP_2015.
- **Experimental Design:** row-column with up to three replicates for each family.
- **Manual data collection:** HA harvested four times (Aug, Oct, Dec in 2018, and Jan 2019).
- **HTP data collection:** an UAV (DJI Matrice 100) equipped with a multispectral sensor (RedEdge, MicaSense) was used for HTP prior to each manual sampling.
- **Image processing:** Agisoft photoscan and QGIS software
- **Statistical analysis:** R software

$$NDVI = \frac{(RNIR - RRed)}{(RNIR + RRed)}$$

Fig. 2: Workflow of image processing and data acquisition for HTP.

0%	10%	20%	30%	40%	50%	60%	70%

HA data included   HA missing data

Fig. 3: Various levels of liming data of HA for bivariate and univariate analysis.

RESULTS AND DISCUSSION

- All VIs showed high correlation with HA, and VIs predicted HA with moderate accuracy (Fig. 4)
- The correlation and coincidence of selection and genetic gain were higher for the bivariate model than any univariate model with various levels (10%-90%) of limiting data of HA

Fig 4: A) Pearson correlation between HA (kg/ha) and VIs B) Linear regression for herbage accumulation (HA) of alfalfa and UAV-based vegetation indices (VIs): NDVI-normalized difference vegetation index, GNDVI-green normalized difference vegetation index, NDRE-normalized difference red edge, GRVI-Green and Red ratio Vegetation Index.

Fig 5: A Comparison of bivariate and univariate models (shape) for HA and NDVI for **coincidence of selection** after applying a 10% selection intensity (red), and **correlation among breeding values** for all families (green) B) Comparison of bivariate model for HA and NDVI (red), and univariate (HA, green, NDVI, blue) models for **genetic gain**

- The univariate model for HA provided higher correlations and % coincidence than univariate models for NDVI (Figure 5), and lower genetic gain for HA was obtained when the selection was performed using only NDVI data
- Coincidence of selection for improved genotypes and correlation among breeding values were increasing with the increased level of HA for univariate models
- NDVI was able to detect the genetic variation present in this breeding population and can be used to select breeding lines exhibiting higher NDVI values, which correspond to breeding lines with higher HA (R^2>0.66)

CONCLUSIONS

- HTP can be a faster way of phenotyping and an alternative of manual phenotyping
- Combining the HTP technology with manually collected HA data represents a reliable method to phenotype alfalfa breeding lines to improve genetic gain for HA in alfalfa

REFERENCE

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