

RELATIONSHIP BETWEEN SOYBEAN INDUSTRIAL-NUTRITIONAL QUALITY AND THE ASSIMILATE SOURCE UNDER HEAT AND WATER STRESS DURING SEED FILLING

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In crops, combination of heat stress (HS) with water stress (WS) modifies photosynthetic processes (RIZHYSKY et al., 2004) causing additive or multiplicative effects (PRASAD, 2004) that modify assimilate supply to seeds. The exposure of soybean to both abiotic stresses during the filling also affects final seed quality (DORNBOS & MULLEN, 1992; CARRERA et al., 2009). To our knowledge, quantitative relationships between seed chemical quality and photosynthetic markers describing the assimilate source are lacking for soybeans grown under heat and water stress during seed filling.

An experiment was conducted in the EEA INTA Manfredi (31° 49'S, 63° 46' W) during the 2012-2014 crop seasons using split-split plot with 2 replications, resulting in a three factorial arrangement: water level, genotype, and temperature level. Water levels were: i) non water stress (NWS), near field capacity, which was achieved by drip irrigation, and ii) water stress (WS), approximately 20% of available water content during 35 days from growth stage R5.5 (FEHR & CAVINESS, 1977). Temperature levels were: i) non heat stress (NHS), at environmental temperature (ET), and ii) heat stress (HS), comprising brief periods of exposure to temperature >32°C for 6 hours per day, during 21 days from R5.5. Field recorded variables were: quantum yield of photosystem II (Φ PSII) and photochemical efficiency of PSII (Fv/Fm) measured with a modulated pulse meter (Hansatech, FMS2 model); canopy temperature (CT) at 12:30 and 14.30h measured with a hand held infra-red thermometer (Testo 845, Spain); leaf chlorophyll levels (estimated with a SPAD chlorophyll meter, Minolta SPAD-502); and leaf relative water content (RWC). The laboratory variables determined were: ferric reducing ability of plasma (FRAP, $\mu\text{moles}/\text{m}^2$), malondialdehyde (MDA, $\mu\text{moles}/\text{m}^2$), total chlorophyll (TChl, mg/m^2), leaf total soluble sugars (LSS, g/m^2) and leaf total starch (LSt, g/m^2), leaf total proteins (LPr, g/m^2) and total ureides (TU, $\mu\text{moles}/\text{g}$ dry leaf). Seed chemical determinations comprises: protein and oil percentage (PrP and OP respectively) by NIRS; oleic, linoleic and linolenic acid concentrations (Ol, Li, Ln, respectively) by gas chromatography and total isoflavones (TI, $\text{mg}/100$ g dry defatted flour) by liquid chromatography. At harvest yield (Y) (g m^{-2}), seed weight (SW, g) and seed number (SN) were also determined.

A multivariate analysis was carried out to explore correlations between soybean seed components and chemical-physiological variables characterizing the photosynthetic source of assimilates during the seed filling. The biplot obtained from the first two principal components (PC1 and PC2) explained 90.8% of total variability in the data (Fig. 1) and revealed that higher levels of protein content (PrC), oil content (OC), OP, Li and TI in seeds were positively correlated with LSt, SPAD, Φ PSII, RWC, FRAP and Fv/Fm, and negatively correlated with PrP, Ol, LSS and CT. Figure 1 also shows that PrC, OC, OP, Li, TI, LSt, SPAD, Φ PSII, RWC, FRAP and Fv/Fm trait vectors were orientated towards irrigated plots, whereas PrP, Ol, LSS and CT trait vectors pointed towards heat and water stressed plots. In the latter, photosynthesis was modified by the overlapping of low levels of primary metabolism parameters (LSt, Φ PSII, Fv/Fm) with elevated levels of oxidative stress (low values of FRAP and high of MDA)

and high canopy temperature ($CT_{12:30}$). These changes in turn, affected final seed quality by decreasing protein and oil contents, Li and TI. The higher concentrations of protein in heat and water stress plots compared to the controls (Fig. 1) confirmed that, although, the response direction of protein and oil content to heat and water stress was the same (reduction), the magnitude of the response was more pronounced for oil. This resulted in a significant increase in protein concentration, in agreement with findings reported by ROTUNDO & WESTGATE (2008).

The observed correlations between seed quality components and the set of chemical-physiological variables were reflected in the fitted regression models. Highly significant explanations ($P < 0.05$) were obtained for protein and oil content from CT and SPAD (Table 1). Regressions indicated that protein and oil content linearly increased with decreasing CT and increasing SPAD. Alternatively, Fv/Fm, LSt, RWC were also significant predictors for both protein and oil content, but the fitted models were not as good as the multiple regressions including CT and SPAD (data not shown). Leaf SPAD provides an instantaneous assessment of leaf chlorophyll content and the values of Fv/Fm are directly proportional to the rate of CO₂ fixation producing starch (LSt), which is one of the main products of photosynthesis. Increases of SPAD (*i.e.* chlorophyll), Fv/Fm and LSt would result in an increase of oil content due to the dependence of seed oil synthesis on current photoassimilate production. These variables were also positively related to seed weight (Fig. 1), which could in turn explain their positive association with protein content. Canopy temperature is an indicator of water status of the crop and it is strongly associated with stomatal conductance (AMANI *et al.*, 1996). Warmer canopies (CT similar or higher than air temperature) may reflect constraints in plant water relationships that reduce CO₂ exchange, limiting oil and protein yield and seed weight.

TI content was negative and linearly related to CT and positively associated with TChl (Table 1). Cooler canopies (CT lower than air temperature) with higher TChl could enhance photosynthetic rates promoting the synthesis of secondary metabolites such as isoflavones. In general, an increase in OI was associated with a reduction in Li and Ln (Fig. 1), which is expected since environments promoting oil with low oxidation capacity (higher OI) differ from those suitable for essential fatty acid production (higher Li, Ln) (CARRERA *et al.*, 2011). There was a significant and negative association between OI and $\phi PSII$, while Ln was positively associated with both $\phi PSII$ and TChl (Table 1). Photosynthesis effects ($\phi PSII$ and TChl) on fatty acid composition could be modulated by changes in the assimilate supply to the seeds. Increasing current photoassimilate production increases the amount of carbon supply to the seed and lipid synthesis (WILLIMS *et al.*, 1999) and extra carbon supply could be preferentially diverted to Li and Ln, which together represent 64% of soybean oil (WILSON, 2004).

Based on these results, management practices aimed at reducing exposure of crop to abiotic stresses during the filling period would allow increases of assimilate supply to the seed, improving quality components. High seed quality products mainly in terms of health promoting activities are desirable to meet niche market demands of soybean.

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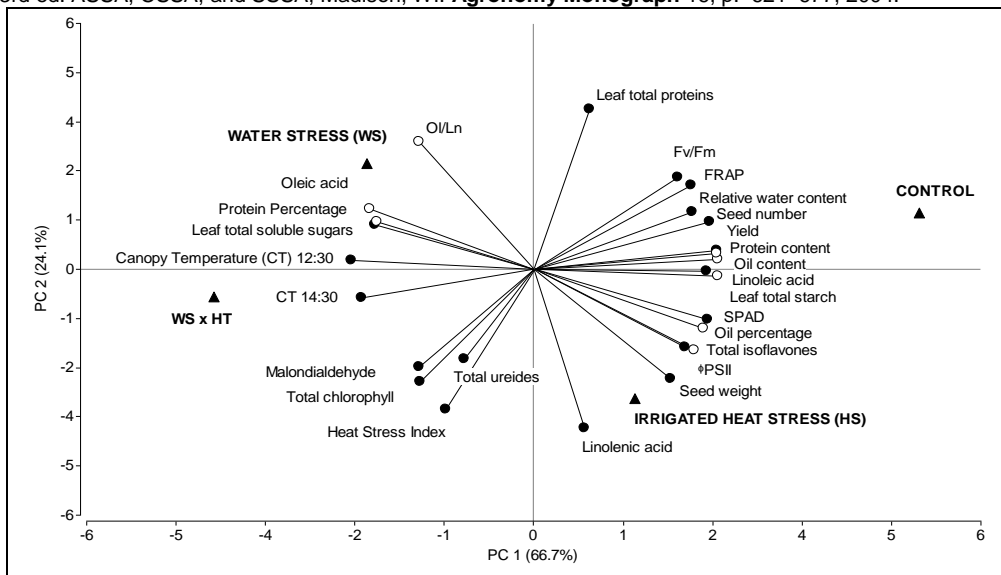


Figure 1. Biplot showing relations between soybean seed nutritional quality (white circles) and chemical-physiological variables of the photosynthetic source (black circles) under 4 conditions during seed filling: control, heat stress irrigated, water stress and heat x water stress (triangles). φPSII= quantum yield of photosystem II; Fv/Fm= Photochemical efficiency of PSII; SPAD= leaf chlorophyll meter; FRAP= ferric reducing ability of plasma; OI/Ln= oleic to linolenic acid ratio.

Table 1. Regression coefficients for soybean seed nutritional components and physiological variables of the photosynthetic source across the 4 treatments (control, heat stress irrigated, water stress and heat stress x water stress).

Chemical variable	Explanatory variable	Regression coefficient	Standard error	p value	Adjusted R ²
Protein content (g/m ²)	const	306.774	54.242	<0.0001	0.54
	CT ₁₂₃₀	-7.572	1.715	0.0001	
	SPAD	1.372	0.457	0.0057	
Oil content (g/m ²)	const	130.727	32.055	0.0004	0.42
	CT ₁₂₃₀	-2.981	1.033	0.0076	
	SPAD	0.874	0.266	0.0028	
Oleic acid (percentage of dry matter)	const	23.710	0.358	<0.0001	0.74
	φPSII	-6.543	0.715	<0.0001	
Linolenic acid (percentage of dry matter)	const	7.241	0.122	<0.0001	0.77
	φPSII	1.900	0.289	<0.0001	
	TChl	0.004	0.002	0.0183	
Total isoflavones (mg/100 g)	const	1155.31	246.01	0.0001	0.43
	CT ₁₂₃₀	-23.07	8.57	0.0125	
	TChl	2.513	0.654	0.0007	

CT₁₂₃₀= Canopy temperature measured at noon (12:30h); SPAD= leaf chlorophyll meter; φPSII= quantum yield of photosystem II; MDA= malondialdehyde; TChl= total chlorophyll.