

# Effects of Durum Wheat Breeding in Italy and Spain on the Flag Leaf Contribution to Grain Filling

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## Introduction

Grain filling in durum wheat (*Triticum turgidum* L. var *durum*) is supported by transient photosynthesis and the translocation of water soluble carbohydrates accumulated prior to anthesis. The flag leaf is considered to be a primary source of assimilates for grain filling and grain yield due to its short distance to the spike and the fact that it stays green for longer than the rest of the leaves. Positive correlations have been found between flag leaf size and yield (Briggs and Aytenfisu, 1980), between leaf area duration (LAD) and grain weight, and between LAD and grain filling duration (Gelang et al., 2000). This study was undertaken to assess the changes caused by durum wheat breeding in Italy and Spain on flag leaf attributes and their effect on the contribution of the flag leaf to grain filling.

## Materials and Methods

Two field experiments, including an historical series of 24 varieties released in Italy and Spain in different periods along the 20<sup>th</sup> century, were sown in 2004 and 2005 in north-eastern Spain following a RCB design. According to the year of release the varieties were grouped as: old (before 1945), intermediate (from 1950 to 1985) and modern (from 1988 to 2000). Grain filling curve coefficients (final grain weight, *W*; maximum rate of grain filling, *R*; and grain filling duration, *D*) were calculated from a logistic equation fitted to the mean grain weight data of spikes periodically sampled during grain filling (Darroch and Baker, 1990). Grain filling curve coefficients of intact plants (checks) were compared with those of plants whose flag leaf blade had been removed one week after anthesis.

**Pictures 1 and 2.** Application of the flag leaf removal treatment at anthesis. Sampling of intact spikes at maturity for determining grain weight and the translocation of pre-anthesis assimilates from the flag leaf to the grain.



Flag leaf blade attributes (length, area, relative area in relation to total leaf area per plant, weight, specific area and chlorophyll content in SPAD units) were measured at anthesis on a sample randomly taken of ten intact main stems. Dry matter translocation (DMT, mg) from the flag leaf was calculated as the difference between dry weight at anthesis and at physiological maturity. Dry matter translocation efficiency (DMTe, %) and the contribution of pre-anthesis assimilates to the grain (CPA, %) were assessed as:

$$\text{DMTe} = (\text{DMT} / \text{flag leaf dry weight anthesis}) \times 100$$

$$\text{CPA} = (\text{DMT} / \text{main spike grain weight}) \times 100$$

## Results and Discussion

Flag leaf removal significantly reduced *W* and *R* of Italian and Spanish varieties, but it had no effect on *D* (Table 1). Reductions in *W* due to flag leaf blade removal were larger in modern than in old varieties (Table 1), suggesting that the contribution of the flag leaf blade to grain filling increased over time. The most significant changes on flag leaf attributes of Italian varieties were recorded for chlorophyll content and leaf area duration (LAD), which increased 9.1% and 3.8% respectively (Table 2). According to a stepwise regression analysis (Table 3), the increase on the effect of flag leaf blade removal on final grain weight was mostly explained by the enlargement of the flag leaf area duration in Italian varieties ( $R^2=0.59$ ,  $P<0.05$ ; Fig. 2). Longer green flag leaf area duration has been related with the ability to maintain yield under drought (Foulkes et al., 2007), suggesting that Italian breeders enlarged LAD as a mechanism to increase durum wheat adaptation to Mediterranean conditions. In Spanish germplasm flag leaf length, area and weight were drastically reduced from old to modern varieties by 22%, 32% and 30%, respectively, while leaf area duration increased by 13% (Table 2). Changes on the effect of flag leaf removal to final grain weight were mostly explained by the reduction of flag leaf length, according to the stepwise regression analysis ( $R^2=0.61$ ,  $P<0.05$ ; Table 3 and Fig. 3). Changes in the flag leaf contribution to grain filling in the Spanish germplasm may be consequence of the introduction of the *Rht-B1* dwarfing gene during the 1970's (Royo and Briceño, 2008). The contrasting strategies followed to improve durum wheat yield in Italy and Spain (Royo et al., 2007) may have originated the differences between the attributes explaining the increases on the increases on the flag leaf contribution to grain filling.

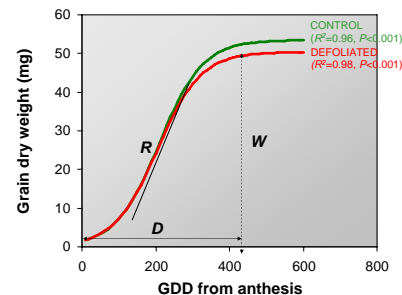
## Acknowledgements

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**Figure 1.** Grain filling curves fitted to the mean grain weight of intact and defoliated main stems averaged over 24 varieties.



**Table 1.** Grain filling curve coefficients (*W*, final grain weight, *R* maximum rate of grain filling and *D*, grain filling duration) of control and flag leaf defoliated main stems for old, intermediate and modern Italian and Spanish varieties. In red, difference between treatments (%)

Country	Period	Treatment	<i>W</i> (mg)	<i>R</i> (mg GDD <sup>-1</sup> )	<i>D</i> (GDD)
Italian	Old	Control	56.1 a	0.235 a	395 a
		Defoliated	54.5 a	0.221 b	395 a
	Intermediate	Control	52.1 a	0.223 a	378 a
		Defoliated	47.9 b	0.210 b	371 a
	Modern	Control	53.4 a	0.218 a	395 a
		Defoliated	48.5 b	0.199 b	391 a
Spanish	Old	Control	53.4 a	0.235 a	374 a
		Defoliated	51.6 a	0.225 a	374 a
	Intermediate	Control	54.0 a	0.225 a	390 a
		Defoliated	51.0 b	0.214 a	385 a
	Modern	Control	50.0 a	0.228 a	365 a
		Defoliated	46.5 b	0.216 b	359 a

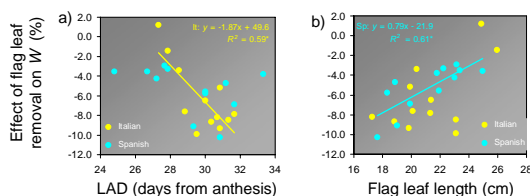
Means within a column and treatment followed by the same letter are not significantly different at 5% probability level.

**Table 2.** Means of the flag leaf attributes (LAD, leaf area duration; relLAP(%), flag leaf area in relation to the total leaf area per plant; DMT, dry matter translocation, DMTe, dry matter translocation efficiency; CPA, contribution of pre-anthesis assimilates to grain weight) for old, intermediate and modern Italian and Spanish varieties.

	Length (cm)	Area (cm <sup>2</sup> )	Weight (g)	Specific area (cm <sup>2</sup> g <sup>-1</sup> )	SPAD anthesis	LAD (d)	rel LAP (%)	DMT (mg)	DMTe (%)	CPA (%)
Italian										
Old	23.1 a	25.2 a	0.128 a	196 a	43.7 c	29.0 c	19.2 a	0.031 a	22.8 a	1.90 a
Intermediate	19.3 c	21.2 b	0.117 b	180 b	46.2 b	30.9 b	18.6 a	0.025 a	20.4 a	1.37 a
Modern	21.5 b	24.2 a	0.129 a	192 ab	47.7 a	30.1 a	19.1 a	0.031 a	21.6 a	1.53 a
Spanish										
Old	23.6 a	29.3 a	0.147 a	205 a	43.9 b	26.7 b	16.9 b	0.048 a	28.6 a	2.43 a
Intermediate	21.6 b	25.0 b	0.135 b	185 b	44.1 b	30.7 a	22.1 a	0.038 a	28.2 a	1.97 a
Modern	18.5 c	20.0 c	0.103 c	199 a	47.2 a	30.3 a	19.7 ab	0.019 b	17.0 b	0.99 b

Means within the same column and country followed by the same letter are not significantly different at 5% probability level.

**Figure 2.** Relationship between the effect of flag leaf removal on final grain weight (*W*, %) and (a) leaf area duration (LAD), and (b) flag leaf length, for 12 Italian and 12 Spanish varieties released in different periods during the 20<sup>th</sup> century.



**Table 3.** Stepwise regression equations ( $P<0.05$ ) for the relationships between the percentage of change of *W*, *R* and *D* as consequence of flag leaf blade removal as dependent variables, and flag leaf attributes as independent variables, for Italian and Spanish durum wheat varieties.

	Regression equation	Partial $R^2$	$R^2$	<i>P</i>
Italian	% of <i>W</i> change = $-1.87\text{LAD} + 49.6$	LAD: 0.59	0.59	0.004
	% of <i>R</i> change - No variable entered into the model			
	% of <i>D</i> change - No variable entered into the model			
Spanish	% of <i>W</i> change = $0.76\text{Length} - 21.6$	Length: 0.61	0.61	0.005
	% of <i>R</i> change = $-1.50\text{relLAP} + 24.4$	relLAP: 0.41	0.41	0.030
	% of <i>D</i> change = $1.60\text{relLAP} + 0.75\text{Length} - 47.6$	relLAP: 0.45	0.45	0.022
		Length: 0.24	0.69	0.037

LAD, Leaf area duration in days from anthesis; relLAP, flag leaf area in relation to total leaf area per plant (%)