# Determination of maximum achene size in sunflower

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#### ABSTRACT

It has been suggested that the maximum volume of grain could determine its potential dry weight, and that grain water content can be used as a surrogate of grain volume, but this issue has not been investigated in sunflower. The aim of this work was to examine the relationships between final achene weight and the dynamics of achene volume and achene, pericarp and embryo water contents. Seven experiments were conducted between 2002-6 in Argentina, testing a total of 10 sunflower genotypes. Achenes were collected every 2-3 days, and the fresh and dry weights of the achene, pericarp and embryo, achene dimensions (length, breadth, width) and volume were determined. The pericarp played a dominant role in achene water content throughout grain filling. Maximum achene and pericarp water contents were reached early in development, and these maxima were good predictors of final achene and embryo dry weights across genotypes. Achene water content and volume showed parallel increases between anthesis and peak achene water content, but achene volume and dimensions continued to increase for a few days after the time of peak achene water content. We have concluded that the relationship between final achene dry weight and peak values for achene water content is not causal, and that the strong correlation between these variables arises because of the correlation between achene water content and volume during the phase where both variables increase together and the fact that, at the time of peak achene water content, the achene has reached close to 90% of its final volume.

Key words: achene dimensions – achene size – pericarp – sunflower – water content.

# INTRODUCTION

In sunflower, as in other grain species, dry matter accumulation in the achene is related to achene moisture dynamics, and physiological maturity for sunflower is achieved with a grain water concentration of 38% (Rondanini et al., 2007). It has been suggested that maximum grain water content may be used as surrogate for maximum grain volume and that maximum grain volume may limit maximum final grain dry weight (Borrás et al., 2004). The relationship between maximum achene water content and achene final dry weight has not been explored for sunflower. Because pericarp maximum dry weight in sunflower is achieved well before maximum embryo dry weight during grain filling (Mantese et al., 2006), and if the pericarp represents a physical restriction to the growth of whole achene, maximum pericarp volume could determine the final achene size across different sunflower genotypes. The ease (relative to cereal grains) with which pericarp of the growing sunflower achenes can be separated from its contents increases the possibility of studying this issue.

The aim of this work was to relate final sunflower achene weight to the dynamics of achene volume, achene dimensions, and achene, pericarp and embryo water contents.

### MATERIALS AND METHODS

Seven experiments were conducted between 2002-6 at FAUBA (Buenos Aires) and the Advanta Semillas Biotechnology Centre (Balcarce). Ten genotypes were evaluated, having contrasting final achene size (30-105 mg achene<sup>-1</sup>) and pericarp proportion (17-35 %). They included inbred lines (HA89, IM9), white striped hybrids (Paraiso30, M734, CF19, CF29) and black striped hybrids (Paraiso20, VDH488, Aguara, DK4050). Experimental units were individual plants, and each experiment had three replicates. Starting at anthesis, achenes were collected every 2-3 days, and their volume (in genotypes DK4050 and CF29 only), dimensions (length, breadth, width, in genotypes Paraiso30, HA89, DK4050, and CF29 only), fresh weight and dry weight (all genotypes) were determined. Water content in achene, pericarp and embryo was calculated as the difference between the respective fresh and dry weights.

### **RESULTS AND DISCUSSION**

Pericarp and embryo contributions to whole achene water content dynamics

The dynamics of dry and fresh weight, as well as those of water content and concentration for the whole achene, pericarp and embryo exhibited consistent patterns across genotypes and environments, and are illustrated for the genotype CF19 in Fig. 1. Achene and pericarp fresh weight increased rapidly during early grain-filling, to fall after achieving peak values, while embryo fresh weight increased until physiological maturity (Fig. 1B). The pericarp was the dominant component of whole achene water content right up to physiological maturity (Fig. 1C), and exhibited a higher water concentration than the embryo throughout, even after achene water concentration had fallen to harvest maturity values (i.e., ca. 17%, Fig. 1D). Sunflower achene water dynamics (Fig. 1 C) contrast with those of wheat (which show an extended plateau) and soybean (which achieve maximum water content very close to physiological maturity) grains (Egli, 1998).



**Fig. 1.** Post-anthesis dynamics of whole achene, pericarp, and embryo dry weights (A), fresh weights (B), absolute water contents (C), and water concentration (fresh weight basis, D) for achenes from the peripheral position on the capitulum of hybrid CF19 (Exp. 1). Fitted functions are bi-linear (A) and cubic polynomials (D). Arrows on the "x" axis indicate PM. Each datum point is the mean of three replicates.

Averaged across genotypes, maximum pericarp water content was 1.9 times greater than maximum embryo water content, in spite of the fact that average embryo final dry weight was 3 times greater than average pericarp final dry weight (data not shown). Across genotypes, 62-78 %of the water in the achene was contained within the pericarp at the time of whole achene maximum water content (Table 1). This was unexpected, given that the pericarp constituted between 17-35 % of the final achene dry weight and, even at achene maximum water content, embryo dry weight was beginning to approach that of the pericarp.

Recently it has been shown that the achene dry weight/water concentration relationship (in both absolute and relative to maximum dry weight terms) for sunflower can be described using a trilinear relationship, in which the first section (for achene water concentrations in the 90-80% range) has a much steeper slope than the subsequent section (Rondanini et al., 2007). The slopes of the first two sections of the corresponding trilinear relationship for maize show the opposite behaviour (Borrás and Westgate, 2006). In this initial phase of the grain-filling process, most of the water in the achene is found in the pericarp. Thus, pericarp dominance of achene water content dynamics underpins the contrast between the

grain dry weight/water concentration relationships of sunflower and that of other grains like maize (Borrás and Westgate, 2006) and true seeds such as soybean (Egli, 1998).

**Table 1.** Achene water and dry weight contents at the time of achene maximum water content and proportions (%) of these water and dry matter contents present in the pericarp (P) and embryo (E). Values are means of three replicates.

Genotype	At the time of maximum achene water content					
	Achene	water c	ontent	Achene dry weight		
	(mg)	Proportion in		(mg)	Proportion in	
		Р	Е		Р	Е
P30	140	65	35	30	58	42
M734	133	74	26	31	70	30
CF19	86	72	28	22	67	33
P20	83	75	25	18	71	29
VDH	80	74	26	19	67	33
Aguara	72	73	27	18	67	33
HA89	59	62	38	17	66	34
M734i	60	78	22	14	79	21

Associations between final achene and embryo dry weights and achene and pericarp maximum water contents

Achene final dry weight showed strong associations (r=0.95) with achene and pericarp maximum water contents (Fig. 2). Given the dominant contribution of the pericarp to achene maximum water content (Table 1), the strong association between final achene dry weight and pericarp maximum water content (Fig. 2), and the fact that maximum pericarp dry and fresh weights are achieved before those of the embryo contained within the pericarp (Fig. 1), we also tested the association between embryo final dry weight and pericarp maximum water content. We found a strong relationship (r=0.92) between these two variables (Fig. 2). Relationships between achene and pericarp final weights and their respective maximum fresh weights were also strong (data not shown).



**Fig. 2.** Relationship between final achene dry weight and achene maximum water content (A), pericarp maximum water content (B), and relationship between final embryo dry weight and maximum pericarp water content (C) for 10 sunflower genotypes.

#### Dynamics of achene volume and achene dimensions

In the genotypes for which achene water content, volume and dimensions were followed simultaneously, achene water content reached its maximum value early during the development, some time before achievement of maximum achene volume (as illustrated for DK4050 in Fig. 3A). Achene volume peaked at 18 daa (Fig. 3B) and then decreased slightly. Maximum achene length was achieved before the maxima for other dimensions of the achene, whereas maximum achene breadth was determined later during the development, and matched to the timing of maximum achene volume (Fig. 3A and 3B).



Fig. 3. (A) Dynamics of achene, pericarp and embryo water content (WC) and achene volume; and (B) dynamics of achene dimensions, in sunflower genotype DK 4050.

Plots of the variables shown in Fig. 3 in relative (to the respective maxima) terms against time from anthesis (Fig. 4) showed a strong parallelism for the trajectories of achene relative water content, relative volume and pericarp relative water content. At the time of peak achene and pericarp water contents, achene volume had reached 88-90% of its peak value (Fig. 4A). Achene dimensions showed different trajectories, with achene breadth being the last to achieve its peak value (Fig. 4B), coinciding with peak achene volume.



Fig. 4. (A) Relative (to the maximum value for each variable) dynamics of achene, pericarp and embryo water content (symbols as in Fig. 3 A); and (B) achene dimensions. Data for the hybrid DK 4050.

# Achene volume and achene water content relationship

During the anthesis (maximum achene water content phase), volume and water content were positively associated (Fig. 5), but the relationship was not 1:1, indicating that achene volume is not all occupied by water.



Fig. 5. Achene water content/volume relationship (for the anthesis/maximum WC phase) in two sunflower genotypes.

We conclude that, in sunflower, maximum achene water content is reached early in the development and that pericarp water content dynamics play a dominating role in achene water content dynamics (Fig. 1, Table 1). Differences in grain water content dynamics between sunflower and other grain crops (cereals, soybean) are probably due to a lesser importance of the pericarp in the structure of these latter grains. Because maximum achene water content and volume are determined fairly early during grain filling (e.g., over 60% of peak volume is achieved 10 days after anthesis, Fig. 4), achene capacity to compensate in size for early exposure to adverse environmental conditions later during grain filling may be limited, in contrast to that of other species such as soybean (Egli, 1998). Peak values for both achene and pericarp water content are good predictors of final achene and embryo sizes across genotypes (Fig. 2), However, the association between achene water content and volume (Fig. 5) only holds for the interval during which both variables increase in parallel (Fig. 4), and is clearly not causal, suggesting that not all the fruit volume is filled with water during the first phase of grain-filling. We speculate that the effectiveness of peak achene and pericarp water contents as predictors of final achene dry weight (Fig. 2) arises from the fact that peak values for the former two variables are achieved when achene volume has reached about 90% of its peak value (Fig. 4) and because achene volume is strongly associated with achene water content (Fig. 5). Complete resolution of the determinants of final achene size probably requires a study of the dynamics of pericarp epidermis cell division and expansion and of the contribution which cell destruction on the inner face of the pericarp during grain filling makes to the volume available for embryo growth.

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