# Phenotypic plasticities of yield, phenological development and seed traits

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### ABSTRACT

Understanding, quantifying, and exploiting the interaction between genotype and environment (G x E) is at the core of plant improvement. This paper focuses on G x E from a physiological perspective. We present a theoretical framework largely based on Bradshaw's principles of phenotypic plasticity updated to account for recent developments in physiology and genetics. Against this framework we discuss (a) associations between plasticities of different traits and (b) plasticity of seed size and composition. We show that plasticity of sunflower phenological development could be positively associated with yield plasticity under conditions when this is a desirable trait, i.e. when there is no trade-off between yield in low and high yielding environments. We propose that allometric models linking rate and duration could be useful to quantify phenotypic plasticity of agronomically important seed traits.

**Key words**: genetics – genotype x environment interaction – phenotypic plasticity – physiology.

### INTRODUCTION

Grain and oil yield, and quality traits of sunflower depend on environmental (E), genetic (G) and G x E factors. Table 1 is a meta-analysis of 69 sunflower trials over 18 years in northern Argentina (n = 8,974), highlighting the challenge involved in breeding and selection for oil yield in sunflower where environmental and G x E sources of variation dominate. Understanding, quantifying, and exploiting G x E is at the core of plant improvement.

Table 1.	Meta-analysis	of 69	sunflower	trials	over	18	years	in	Argentina	(n =	8,974).	The
partitionin	ng of oil yield v	arianc	e uses a Rest	ricted	Maxin	num	ı Likeli	hoc	d approach	(REM	IL) assu	ming
all variabl	les are random.											

Random term	Variance component	s.e.
year	72692	32521
year.trial	64000	13060
year.trial.rep	4340	653
year.trial.rep.block	2623	327
genotype	2972	671
year.genotype	5704	612
residual (avg across trials)	46731	6567

Breeders are well aware of the issues involved in G x E, whereas physiologists and ecologists look at the same type of problem from the perspective of *phenotypic plasticity* or *norms of reaction* (Bradshaw, 1965; Bradshaw, 2006; De Witt et al., 1998; Pigliucci, 2001; Pigliucci et al., 1995). *Phenotypic plasticity* is "the amount by which the expressions of individual characteristics of a genotype are changed by different environments" (Bradshaw, 1965). The aim of this paper is to discuss selected aspects of phenotypic plasticity of sunflower yield and seed traits from a physiological perspective.

This article has three parts. First, we introduce some principles related to phenotypic plasticity that provide the theoretical background for the paper. Second, we explore the notion of positive associations between plasticities. Using data from sunflower trials involving a large number of hybrids and environments, we show preliminary evidence for a positive link between phenotypic plasticity of yield and phenotypic plasticity of phenological development. Third, we present a novel quantitative model to analyse seed size variation in terms of rate and duration of seed growth. For most grain species, including sunflower, we show that plasticity of seed size could be ascribed to specific allometric conditions, and that plasticity of seed size could be an important driver of yield plasticity. This allometric model could also be applied to quality related traits, e.g. oil concentration.

## DISCUSSION

This paper is informed by three established principles (1-3) and a newer, less tested proposal (4):

- 1. "The plasticity of a character is an independent property of that character and is under its own specific genetic control" (p. 119 Bradshaw, 1965). Bradshaw (1965) insightfully formulated this proposal over forty years ago, and Reymond et al. (2003) have demonstrated unequivocally that phenotypic plasticity is a trait on its own, with its own genetic control. A corollary to this principle is that plasticity evolves (Pigliucci, 2005; Zhivotovsky et al., 1996) and therefore could be considered as a breeding aim on its own. The findings of Reymond et al. (2003) open a new, more robust opportunity to use QTLs as breeding tools, and highlight the need for appropriate quantitative models that relate traits and environmental drivers, or alternatively, establish physiologically meaningful relationships between traits.
- 2. Plasticity is specific for a character and is specific in relation to particular environmental influences (Bradshaw, 1965). This adds a layer of complexity to the subject, because the plasticity of a trait (e.g. kernel oil concentration) may be high or low depending on the environmental drivers.
- 3. There is a hierarchy of plasticities, i.e. stable traits are often associated with plastic, related traits (Bradshaw, 1965). The trade-off between seed number and size is a typical, agronomically relevant case of this principle whereby high plasticity in number is associated with low plasticity in size. Sadras (2007) has provided an evolutionary interpretation that matches the notion of a hierarchy in the plasticities of seed size and number in annual plants.
- 4. There are cases of *positive* associations between plasticities of certain traits. Analysis of the association between plasticity of fruit yield and plasticity of phenology in wine grape favoured the hypothesis of a positive, rather than negative (principle 3) correlation between plasticities (Sadras, Petrie, and Robinson, unpublished).

# Does phenological plasticity contribute to yield plasticity?

Finlay and Wilkinson (1963) developed a method to quantify trait plasticity, that has been widely applied to the analysis of grain yield in annual crops. Calò et al. (1975) used this approach to quantify phenological plasticity of grapevine. Fig. 1 illustrates the rationale of this method applied to the analysis of plasticity of flowering time of sunflower hybrids grown in diverse environments of northern Argentina. The coefficient of phenotypic plasticity is the dimensionless slope of the linear regression between date of flowering of an individual variety in a particular environment, and the mean value of the trait across varieties in that particular environment. A variety with slope = 1 has average stability over all environments, a variety with slope > 1 has above-average plasticity, and a variety with slope < 1 has below-average plasticity.



Fig. 1. Quantification of phenotypic plasticity of flowering in sunflower using the method of Finlay and Wilkinson (1963).

In the case study of this paper, environments resulted from the combination of locations and seasons, and the data set comprised 32 hybrids grown in at least 15 environments. For this data set, plasticity for yield ranged from 0.72 to 1.29 (Fig. 2). All hybrids performed similarly in the more stressful environments (i.e. the slope of the regression between minimum yield of each hybrid and its yield plasticity was not significantly different from zero, P = 0.34). Higher plasticity was associated with the ability to capture the benefits of better environments, with a rate of increase in maximum yield of 1939 kg/ha per unit increase in plasticity (P < 0.0001). A similar conclusion was reached from analysis of oil yield: oil yield plasticity ranged from 0.72 to 1.30, was correlated with plasticity of grain yield (r = 0.90, P < 0.0001) and was related to maximum (rate = 1024 kg oil/ha per unit increase in plasticity, P < 0.0001) but not with minimum oil yield (P > 0.25). High yield plasticity in this particular combination of hybrids and environments is therefore a desirable trait, as it does not involve tradeoffs between stress tolerance and yield potential.



Fig. 2. Phenotypic plasticity of grain yield in sunflower hybrids was related to their ability to capture the benefits of the best environments (slope of maximum yield vs plasticity significant at P < 0.0001) and independent of their performance in the more stressful environments (slope of minimum yield vs plasticity not different from zero; P = 0.34).

In a broad sense, phenological development is recognised as the more important attribute of crop adaptation (Passioura, 1996; Passioura, 2007; Richards, 2006; Sadras and Trápani, 1999). This relates to a series of tradeoffs. Firstly, there is a trade-off between late flowering that allows for canopy and root development (Giménez and Fereres, 1986) and the decline in potential grain set generally associated with low radiation-to-temperature ratios of late flowering crops (Cantagallo et al., 1997). Secondly, in some environments, flowering date may also involve trade-offs between the risk of frost and the risk of heat stress, terminal drought, rainfall at harvest or diseases. For the combination of hybrids and environments in this analysis, we found yield plasticity was higher in late-flowering hybrids, with mean flowering date accounting for 47% of the variation in yield plasticity (Fig. 3) and 40% of the variation in oil yield plasticity (not shown). Flowering plasticity was unrelated to mean flowering date, and accounted for 20% of the variation in yield plasticity (Fig. 3) and 16% of the variation in oil yield was associated with both mean flowering date (r = 0.51, P = 0.003) and flowering plasticity (r = 0.40, P = 0.02) whereas minimum yield was weakly related to mean flowering date (r = 0.34, P = 0.06) and unrelated to flowering plasticity (P = 0.34).



Fig. 3. Plasticity of grain yield in a collection of 32 sunflower hybrids in northern Argentina was associated with both late flowering, and flowering plasticity. Inset shows flowering plasticity was not associated with mean flowering date.

The relationships between plasticity in yield and plasticity in phenology deserve further attention. Biologically, this relationship adds a new dimension to the understanding of crop adaptation. From a breeding perspective, it would be of interest to establish the genetic basis of phenological plasticity (Principle 1), and eventually exploit this trait where plasticity in yield is a desirable trait, i.e. when performance in stressful environments does not compromise performance in better environments.

#### Plasticity of seed size: allometric conditions and relationship with yield plasticity

Here we explore the allometric conditions for seed size plasticity using a multi-species comparative approach, and investigate the links between seed size plasticity and yield plasticity using a limited data set of sunflower hybrids grown in contrasting environments.

#### Allometric conditions for plasticity of seed size and quality traits

There are many growth processes that can be approximated to sigmoidal patterns with characteristic rates and durations, including leaf expansion, seed growth, accumulation of oil in seed and accumulation of sugar and pigments in fruits. For any such process, we can express the maximum value of the trait (A) as the product of rate and duration:

$$A = rate x duration$$
(1)

Sadras et al. (2007) proposed an allometric formulation of this model (Fig. 4):

 $\log duration = \log A - \alpha \log rate$  (2)

The advantage of this model is that the scaling exponent  $\alpha$  indicates three types of responses: the trait is stable as a result of full compensation between rate and duration ( $\alpha = -1$ ), the trait is variable as a result of rate ( $\alpha > -1$ ) or duration-dominated growth ( $\alpha < -1$ ). Sadras et al. (2007) used this approach to demonstrate that accumulation of anthocyanins in berries of grapevine Cabernet Sauvignon in a warm environment is highly plastic ( $\alpha = -0.75 \pm 0.041$ ), in contras to sugar accumulation which is very stable ( $\alpha > -1.11 \pm 0.050$ ). Fig. 5 illustrates the application of this concept to the analysis of seed size in grain crops. These particular experiments showed relatively stable seed size in soybean, with a scaling exponent correspondingly close to -1, and large variation in seed size of sunflower, with a corresponding scaling exponent significantly greater than -1 (P < 0.05), i.e. a flat line reflecting rate-dominated seed growth. These results cannot be considered general for these species, but particular for the combination of cultivars and environments (Principle 2).



**Fig. 4 (a).** Many plant traits, including seed size, seed oil content and leaf area, conform to an approximate sigmoidal pattern with characteristic rates and durations. (b) The allometric relationship between duration and rate allows for a quantitative characterisation of trait plasticity. Adapted from Sadras et al. (2007).

A broader test of the concept included 45 data sets involving nine crop species, and sources of variation including genotype, environment, and their interaction (Fig 6). Relative variation in seed size ranged from 5 to 274%, and the scaling exponent was strongly concentrated in the range from 0 (large, rate-driven seed size range) to -1 (narrow seed size range due to mutually cancelled effects of rate and duration). The range of seed size declined when the scaling exponent declined from approximately 0 to -

1. An  $\alpha \approx -1$  (rate and duration effects cancel each other) is necessary and sufficient for small variation in seed size, whereas  $\alpha \approx 0$  is necessary but not sufficient for large seed size variation. The magnitude of seed size variation is dependent on the variation in the rate of seed growth when  $\alpha \approx 0$ . This double condition for seed size variability is summarised in a multiple regression model with  $\alpha$ , and range of rate of grain filling as independent variables, which accounted for 73% of the variation in range of seed size.

Soybean



log rate of seed growth

Fig. 5. Examples of intra-specific scaling relationships between rate and duration of seed growth in sunflower and soybean. Multiple symbols for a cultivar indicate different experiments or seasons. The solid line is the least squares regression, and dashed lines are isolines of seed size with  $\alpha = -1$ . Standard errors (SE) are common to the scaling exponents calculated with model I ( $\alpha_{LS}$ ) or model II ( $\alpha_{RMA}$ ) regression. Data sources: sunflower, López Pereira et al. (1999a); soybean (control treatment), Egli (1999). For soybean, rate is in mg seed<sup>-1</sup> d<sup>-1</sup> and duration in d, and for sunflower rate is in mg seed<sup>-1</sup> °Cd<sup>-1</sup> and duration in °Cd. Variate units do not affect the magnitude of the scaling exponent. Adapted from Sadras and Egli (2008).



**Fig. 6.** Relationship between seed size range and α, the scaling exponent relating duration and rate of seed growth. Adapted from Sadras and Egli (2008).

Allometric analysis allowed for an integrated perspective on the interplay between rate and duration of seed filling, which in turn accounts for the genetic and environmental factors modulating seed size in grain crops. This allometric approach could be useful for evolutionary, agronomic and physiological analysis of seed size, and may also be used for other processes such as leaf growth or accumulation of oil or tocopherols in sunflower seed, where a framework of rates and durations is applicable. It would be of interest to consider the genetic substrate of parameter  $\alpha$  for traits of agronomic interest (Principle 1).

#### Seed size plasticity and yield plasticity

The allometric relationship for sunflower in Fig. 5 was derived from crops grown under favourable conditions, i.e. hybrid grain yield  $\ge 4$  t/ha, oil concentration  $\ge 50\%$  (López Pereira et al., 1999a). Under these conditions, the duration of grain filling is typically around 30-35 days or about 650 °Cd (base = 4°C), and differences in seed size are related to differences in rate of grain filling (de la Vega and Hall, 2002; López Pereira et al., 1999b). Relationships between rate and duration of grain filling could be different, however, in environments where excess or deficit of water supply during grain filling accelerate leaf senescence (Grassini et al., 2007; Hall et al., 1985).

Here we explore the relationships between seed size plasticity, quantified with parameter  $\alpha$  and yield plasticity quantified with the method of Finlay and Wilkinson (1963) for a set of four sunflower hybrids grown under six environmental conditions in Argentina (for details see de la Vega and Hall 2002). The size of the data set is restricted due to the need to conciliate the time consuming sampling necessary to derive seed growth curves and  $\alpha$ , and the relatively large number of cultivars and environments required to calculate yield plasticity. Growing conditions include a timely October sowing and a late December sowing conducive to lower yields. One of the seasons (1997/98) was "El Niño", with excessive rainfall and cloudy days detrimental to sunflower yield even for timely sown crops (Magrin et al., 1998). Yield plasticity ranked Aguará < Morgan 734 < Contiflor 15 (Fig. 7). Yield plasticity among hybrids are partially related to their patterns of seed growth (Fig. 8). In response to late sowing, Contiflor 15 and Morgan 734 reduced both rate and duration of grain filling and Aguará slightly increased the rate of seed filing at the expense of shorter duration. Even for a set of few hybrids and growing conditions, Fig. 8 illustrates the

complex interplay of rate and duration of seed filling, and relationships between seed filing pattern and yield plasticity are not straightforward. Allometric relationships between rate and duration were loose, with large standard errors (not shown). Despite of this, the scaling coefficient  $\alpha$  summarised the contrasting rate-duration relationships of these hybrids, and captured a substantial part of the variation in yield plasticity (Fig. 9). This reinforces the interest in the previous proposition of exploring the genetic basis of  $\alpha$ .



**Fig. 7.** Yield plasticity (slope of regressions) of three sunflower hybrids grown under six environmental conditions in Argentina. S1 is a timely October sowing, and S2 is a December sowing conducive to low yields.



Fig. 8. Dynamics of seed growth of three sunflower hybrids sown in October (S1) or December (S2) 1996 at Venado Tuerto, Argentina. Adapted from de la Vega and Hall (2002).



Fig. 9. Relationship between yield plasticity and  $\alpha$ , the scaling exponent relating duration and rate of seed growth, for four sunflower hybrids grown in contrasting environmental conditions.

#### **CONCLUDING REMARKS**

The insightful vision of Bradshaw (1965), providing the contemporary definition of phenotypic plasticity and the notion that plasticity is a trait of its own, with its own genetic control acquires a new dimension when Reymond et al. (2003) demonstrate that the plasticity of certain traits could be traced back to specific QTLs. Against this conceptual framework, this paper showed that a physiological viewpoint of phenotypic plasticity can contribute to the understanding of G x E of sunflower yield. For the first time, here we showed that phenotypic plasticity of phenological development could be positively associated with yield plasticity under conditions when yield plasticity is a desirable trait, i.e. where there is no tradeoff between performances in low and high yielding environments. Allometric models linking rate and duration could be useful to quantify phenotypic plasticity of agronomically important seed traits.

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