

Modelling the GE interaction for leaf and silk growth rates in maize: genetic analyses of time courses and responses to water deficit

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Plants subjected to water deficit undergo rapidly fluctuating conditions. Those are day-to-day fluctuations for soil water status, with soil drying or rapid re-watering, and hourly fluctuations for evaporative demand, with changes in light or in vapour pressure deficit during the day. Even in water deficit, plants experience a relatively favourable water status at the end of the night, and loose water during the day so their water status in the afternoon is unfavourable. Consequently, the plant growth rate and photosynthesis rate have rapid fluctuations, with different time course for each genotype (Sadok et al. 2007). Because of this complexity, it is tempting to define tolerance to water deficit at the temporal scale of the whole cycle, by comparing overall performance of genotypes in network of field experiments which buffer the short term fluctuations. This view has allowed considerable progress in drought tolerance in the last 30 years (Campos et al. 2004), but makes it difficult to refine the adaptation of genotypes to changing climatic conditions, or to allow local adaptations to target populations of environments (Chapman et al. 2002).

We have used a different strategy, in which the phenotype is not defined by a plant trait at a given time, but as the sensitivity of the considered genotype to soil water status or to evaporative demand, established over a range of environmental conditions (Reymond et al. 2003; Sadok et al. 2007). It consists, first, of expressing all rates per unit thermal time, thereby obtaining temperature-independent rates (Sadok et al. 2007). Second, the major environmental conditions involved in leaf development were identified for short time intervals (minutes to hours). Temperature, evaporative demand and soil water deficit had an overriding effect on leaf growth rate, whereas light and plant carbon balance had minor effects (Ben Haj Salah and Tardieu 1997; Sadok et al. 2007). Third, response curves of leaf elongation rate to temperature, evaporative demand and soil water status were established and the parameters of these responses were analysed genetically (Reymond et al. 2003; Welcker et al. 2007). This allowed simulation of leaf growth in novel inbred lines as defined by their alleles at QTL (Reymond et al. 2003; Sadok et al. 2007). This also allowed comparison of the genetic determinisms of several organs of the plant, namely leaves and silks (Welcker et al. 2007).

1. Modelling the time courses of the elongation rates of leaves and silks.

The leaf elongation rate of a given maize leaf is constant over 5 to 7 days in the absence of environmental changes. This period of time is that necessary for the next two leaves to appear above the whorl (e.g. for leaf 6, until the appearance of leaf 8). This has been observed under stable temperature in a growth chamber (Ben Haj Salah and Tardieu 1995), and is observed under naturally fluctuating conditions provided that rates are expressed per unit thermal time (Sadok et al. 2007). In the latter case, leaf elongation rate is constant during the night, and consistent over 5-7 nights and over different experiments for a given genotype. This baseline leaf elongation rate is, therefore, a genotypic characteristic, which can be analysed genetically in mapping populations.

Night-time leaf elongation rate decreases between nights when the soil water status declines with soil water depletion. The relationship between soil water potential and night time leaf elongation rate is common to several experiments for a given genotype. It is, therefore, a second stable genotypic characteristic. In the same way, elongation rate decreases in the morning with increasing evaporative demand, and a stable relationship is observed for a given genotype between the leaf elongation rate and the vapour pressure difference between meristem and air (VPD_{ma}). Both effects are approximately additive so we can predict leaf elongation rate in any combination of temperature, evaporative demand and soil water status.

Silk elongation essentially follows the same pattern, but with a continuously declining trend. Silk elongation rate is maximum during the night and a gap is observed during the day, which is proportional to VPD_{ma} . Night time silk elongation rate declines linearly with soil water potential, in the same way as leaves. This suggests that common processes may drive the elongations of silks and leaves, with a likely predominance of hydraulic mechanisms. In both organs, the fluctuations of elongation rate with evaporative demand are very rapid, suggesting a physical mechanism rather than mechanisms involving cell cycle or cell wall properties. In both cases, the amount of sugars in the growing zone is equal or higher in plants subjected to water deficit than in well watered plants. Combined with the fact that elongation rate is faster during the night than during the day, this makes unlikely a mechanism based on the availability of carbon to growing cells.

2. Genetic analysis of the sensitivity of leaf growth to water deficit or evaporative demand

The responses of leaf elongation rate to water deficit and evaporative demand have been studied in 100 to 220 recombinant inbred lines of three mapping populations, namely two crosses of temperate parents (F-2 x Io and F-

2 x F252) provided by A. Charcosset (INRA), and one cross between two tropical parents identified by JM Ribaut (CIMMYT) as having contrasting behaviours under water deficit. Each population was studied in 5-8 different experiments, and response curves were common to all experiments for each recombinant inbred line (Reymond et al. 2003, Sadok et al. 2007, Welcker et al. 2007). Heritabilities ranged from 0.5 for the lowest cases to 0.8 for the highest. Surprisingly, the ranges of sensitivities did not appreciably differ between mapping populations, although tropical lines originated from a dry region and were selected for drought tolerance.

QTLs were identified for the sensitivities to soil water deficit and to evaporative demand. In each population, 5 to 7 QTLs were identified for each sensitivity. Most QTLs of sensitivity to evaporative demand tended to co-localise with those of the sensitivity to soil water status, in spite of the fact that they were determined in different experiments, *i.e.* well watered and day-time measurements for the sensitivity to evaporative demand, and water deficit and night - time measurements for the sensitivity to soil water potential.

In total, 7 "hot zones" were determined over the three mapping populations. The effect of each of those was studied in 3 populations of insertion lines, (i) an advanced backcross of a tropical line (CML 444) in the temperate line F252, (ii) BC5-S3 insertion lines of the early flint line Gaspé in B73, provided by R. Tuberosa (U. Bologna), (iii) two sets of reciprocal insertion lines derived from a dent by flint F1 population, provided by Biogemma. Each of the 7 targeted zones was confirmed by the magnitude of their effects in insertion lines, in spite of the fact that those lines had different genetic backgrounds than the mapping populations. Interestingly, the lines which had been chosen for QTLs of sensitivity to evaporative demand also contrasted for the sensitivity to soil water status, and *vice versa*.

This shows that an appreciable genetic variability exists for the sensitivities to water deficit and evaporative demand, which can be synthesised in the effect of 7 zones of the genome.

3 Have sensitivities of leaf and silk growths to water deficit a common genetic determinism ?

Leaf growth and Anthesis-Silking Interval (ASI) are the main determinants of source and sink strengths of maize *via* their relations with light interception and kernel number, respectively. They depend on the abilities of leaves and silks to expand under fluctuating environmental conditions, so we tested the possibility that they may have a partly common genetic determinism. This was first tested in a subset of lines of the tropical mapping population presented above, in which both silk and leaf elongation rates were measured as a function of soil water status or of evaporative demand. The rank of genotypes was common for the responses of both organs (O. Turc, unpublished).

The whole population was then studied, but the silk elongation rate was indirectly evaluated *via* ASI which was measured in 3 and 5 fields under well-watered conditions and water deficit, respectively, by JM Ribaut at CIMMYT. Half of the QTLs for ASI under well-watered conditions co-localised with QTLs for leaf elongation rate measured in the absence of water deficit, and half of the QTLs for ASI under stressed conditions co-localised with QTLs for the response of leaf elongation rate to soil water deficit (Welcker et al. 2007). The allele for leaf growth maintenance was in all cases that for shorter ASI (maintained silk elongation rate) under water deficit. Hence, source and sink strengths may have a partly common genetic determinism.

4. Toward the modelling of virtual genotypes

The results presented here suggest that (i) it is possible to genetically analyse the sensitivities of growth to evaporative demand and to soil water deficit, even in naturally fluctuating conditions. (ii) Because the genetic analysis was carried out on parameters of a model, it is possible to simulate the growth of the considered organ for any genotype of a mapping population under any environmental condition. This was tested for new lines, known by their alleles only (Reymond et al 2003). (iii) Different organs of a plant share part of the genetic determinism of the sensitivity to soil water status and of evaporative demand. This was the case for leaves and silks (Welcker et al. 2007), but also between different leaves of a plant (unpublished). It is, therefore, possible to "pyramid" alleles which determine the reactions of different organs. For this reason, a whole plant model has been designed, which combines the model of leaf growth presented here and a model coordinating the developments of all leaves of a plant (see Chenu et al, same volume). We believe that this approach may have important consequences in the research of better adaptations of genotypes to target populations of environments.

References

- Ben Haj Salah H, Tardieu F (1995) *Plant Physiol*, 109, 861-870.
- Ben Haj Salah, H, Tardieu F (1997) *Plant Physiol* 114, 893-900.
- Campos H, Cooper M, Habben, JE, Edmeades GO, Schussler JR (2004).. *Field Crops Research* 90, 19-34.
- Chapman SC, Cooper M, Podlich D, Hammer GL (2003) *Agronomy Journal* 95, 99-113.
- Reymond M., Muller B., Leonardi A., Charcosset A., Tardieu F (2003). *Plant Physiol*. 131, 664-675
- Sadok W, Naudin Ph, Boussuge B, Muller B, Welcker C, Tardieu F (2007) *PCE* 30, 135-146
- Welcker C, Boussuge B, Benciveni C, Ribaut JM, Tardieu F. 2007. *J Exp Bot*, 58, 339 - 349