

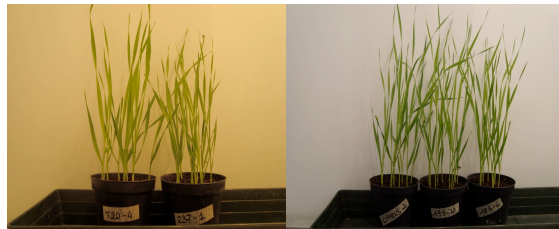
The impact of intra and inter specific nuclear cytoplasmic interaction on the regulation of central metabolism in wheat



Crosatti* C., Quansah* L., Atienza, S.G., Cattivelli, L., and Fait, A.



Background Three different alloplasmic lines TH237, T183 and T195 were used to investigate the effect of *H. chilense*, *Ae. uniaristata* and *Ae. squarrosa* cytoplasm respectively, on nuclear-cytoplasmic interaction in T20 and Chris euplasmic genetic background of *T. aestivum* (Table 1). In the present study we research the effect of foreign cytoplasm on the metabolic regulation in plant response to environmental conditions.



Euplasmic controls T20 with its corresponding alloplasmic line TH237 (left) and Chris with the corresponding alloplasmic lines T195 and T183 (right).

Table 1. Alloplasmic lines		
Alloplasmic line	Euplasmic control (genome donor)	Cytoplasm donor
T183	<i>T. aestivum</i> cv. Chris	<i>Ae. uniaristata</i>
T195	<i>T. aestivum</i> cv. Chris	<i>Ae. squarrosa</i>
TH237	<i>T. aestivum</i> acc. T20	<i>H. chilense</i>

Method Plants from all five genotypes were grown under controlled conditions at two light intensities (150 and 600 μE m⁻² s⁻¹). We employed a GC-MS based method to analyze leaves collected from all genotypes and conditions at two developmental stage (young-4th leaf and fully expanded-2nd leaf, 4L and 2L, respectively). Furthermore grains from plants grown under standard conditions were collected at early, mid and late maturation and metabolome profiled.

Results

Results represent the fold change in metabolite content of the alloplasmic lines compared to their euplasmic controls. We measured significant differences in the metabolism of the alloplasmic lines under all conditions and developmental stages. For instance, the cytoplasm of *Ae. squarrosa* (T195) lead to a decrease activity of the TCA when introduced into a *T. aestivum* nuclear background, with the exception of fumarate which accumulated in 4th leaf under 150 μE (Fig. 1). Sugar contents were differentially affected among lines (Fig.2). For instance the cytoplasm of *Ae. uniaristata* (T183) lead to a significant accumulation of fructose and raffinose – in 4L-150 μE, whilst T195 (*Ae. squarrosa* cytoplasm) displayed higher content of raffinose against a background of generally reduced sugar levels (4F-600 μE).

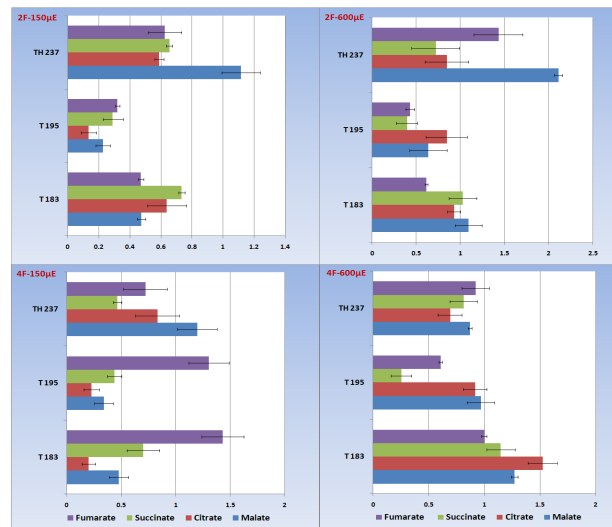


Figure 1. Changes in the level of intermediate metabolite in the TCA cycle. Values indicate either increase or decrease compared to the euplasmic control which is given the value 1. Results are representative of five biological replicate of each metabolite

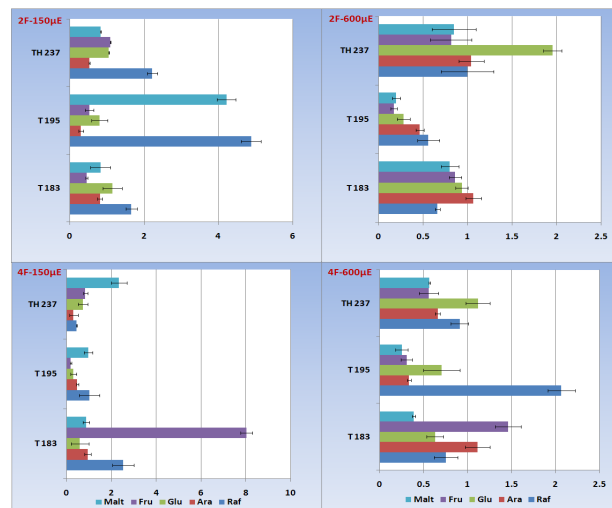


Figure 2. Changes in the level of selected sugars. Values indicate either increase or decrease content as compared to euplasmic control of the alloplasmic lines (to which is given the value of 1). Results are representative of five biological replicate of each metabolite

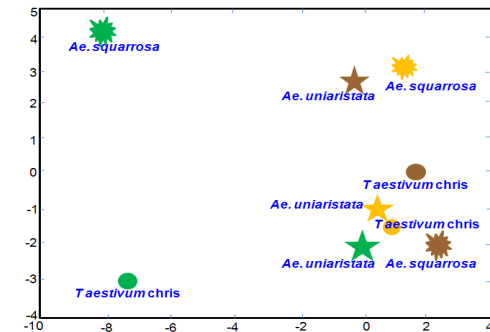


Figure 3. PCA of metabolite profile of alloplasmic lines with their genome donor (refer Table 1). Colors; green, yellow, and brown denote early, mid and late maturation of grain respectively. PCA was done together on the whole data set but plots (Figs. 3 and 4) were separated to facilitate results interpretation of the euplasmic lines with their genome donor

To further determine the impact of cytosol-nucleus interaction on cereal metabolism, we monitored the changes occurring in grains during development. Differences were found among genotypes particularly during early maturation (Fig. 3). Although this preliminary result needs further confirmation, a first analysis suggest that the cytoplasm of *Ae. squarrosa* has a greater impact on seed metabolome than the other two cytoplasm tested particularly the early stage of seed development. *H. chilense* cytoplasm for instance, did not have much impact on the seed metabolome (Fig. 4).

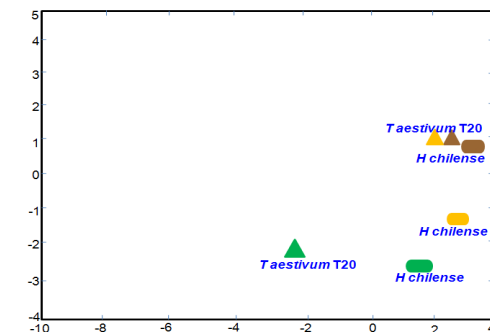


Figure 4. PCA of metabolite profile of alloplasmic line with its genome donor (refer Table 1). Colors; green, yellow and brown represent early, mid and late maturation of grain

In support of the earlier report, this study shows differences in the metabolic profile of leaves and grains of different alloplasmic lines as compared with their euplasmic controls providing an experimental system for the investigation of interactions between nuclear and cytoplasmic genomes and of their consequences on cell metabolism.