

Molecular evidence of temperature decrease influence on coffee flower evocation

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Rationale:

Flower evocation takes place when meristems became committed to differentiate in reproductive instead of vegetative organs under the lead of environmental signals. For coffee plants in the south-south-eastern Brazil those signals were predicted to be temperature and day-length [1]. Genes *FLC/M* and *VRN* are recognized for their roles in model plant flower evocation driven by temperature. *FLC/M* are major suppressors when flowering do not occur without vernalization. By their turn, *VRN* genes keep *FLC* chromatin methylated, to allow flowering. Aiming to verify how coffee orthologs are involved in flower evocation, semi-quantitative analyses in late and early *C. arabica* were accomplished.

Material & Methods:

Primer pairs to amplify *FLC/M* and *VRN* transcripts were planned according to *Coffea* orthologs [2]. Leaves from early and late genotypes [2] were collected during 2021 and 2022, and eventually processed for total RNA extraction (Quickzol, Ludwig). For this work, two plants of each genotype growing in different experimental blocks, from March to May-2021 were evaluated and compared, being 21 biological (different plants/blocks/months) and technical (different RNA extractions/reverse transcriptions) replicates for the early, and 17 biological/technical replicates for the late genotype. Amplicon intensities were quantified using the ImageJ software 'Gel Analysis' routine, and normalized taking the *GAPDH* gene as the internal reference [3] to calculate relative quantities (RQs) of expression. Statistical analysis were performed with SigmaPlot.

Results:

The time lapse March-May was chosen in reason of day-length (12:30 to 10:45 hours) and temperature (minima ranging from 21.9 down to 5.5°C under the first cold spells), in Londrina-PR, Brazil (23°35'S and 51°16'W). Despite not so powerful as qPCR, semi-quantitative analyses were able to access gene expression trends. *FLC* was higher in the late genotype ($P = 0.011$), peaking in April ($P = 0.017$). Interaction gene x genotype was significant ($P = 0.001$), being $FLC > VRN$ in the late ($P = 0.03$) and $FLC < VRN$ in the early genotype, regardless replica. Despite lacking statistical significance, linear models explained gene x time and gene x genotype relationships. *VRN* and *FLC* went up and down in time, respectively, for the early genotype, trending opposite for the late one.

Conclusions & Perspectives:

Results are in agreement to the prediction model regarding temperature [1]. A powerful correspondence between phenological and molecular data has potential to be proven true in the next years, upon conclusion of qPCR for temperature and day-length-related gene expression analyses and meristem morphology study.

References:

1. Camargo, AP; Camargo, MPB. *Bragantia*, 60: 65-68. 2001.
2. Angelo, PCS. *Agricultural Sciences*, 15: 754-779. 2024.
3. Cruz, F. et al. *Molecular Breeding*, 23: 607-616. 2009.