COEXISTENCE BETWEEN TRANSGENIC MON 810 MAIZE AND HIVES

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Introduction

- Pollen is a natural constituent of honey ($\leq$ 0.5%).
- Maize flour might be considered as an ingredient or a contaminant of honey.
- So far PCR has not been considered a helpful technique to separate pollen from contaminants.
- The aim of this study was to investigate the ability of qPCR using plasmid calibrants to provide good estimates of the transgene copy number in relation to a species specific gene copy number and look for any possible application in honey analysis.

Material

- Spiked honey samples
  - GM-F_1 pollen/honey (m/m): 0.45; 0.10; 0.04; 0.01; 0.005%
  - GM-F_1 embryos (m/m): 0.10%
  - GM-F_1 flour: transgene donor - female parent (m/m): 0.10%
  - GM-F1 flour: transgene donor - male parent (m/m): 0.10%
- Local honey (Salvaterra)
- Imported (EU and non-EU) honey

Methods

- DNA extraction: validated CTAB-based method (van den Bulcke *et al.*, 2012), slightly modified.
- Calibration curves: ERM®-AD413 (for MON810) and ERM®-AD415 (for NK603) calibrants at nominal concentration of 2×10^6 copies/μL
- Quality control for PCR efficiency and positive control: ERM®-BF413f
- Real-time PCR: according to EURL – GMFF, but with a plasmid calibrant certified for the copy number ratio instead of a reference material certified for its GM mass fraction.

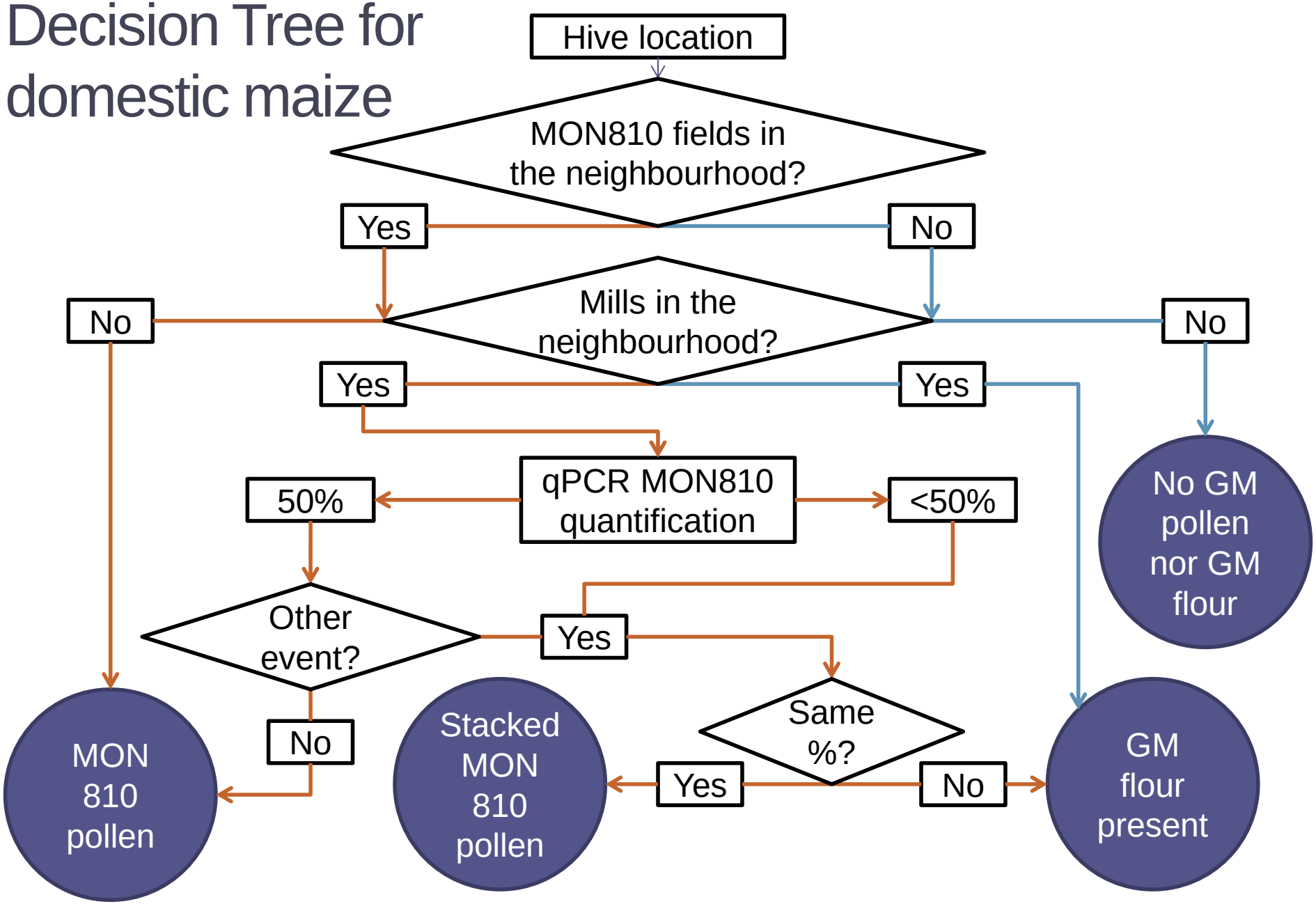
GM copy number ratios

Matrix – Spicked samples	MON810 copy number ratio (Average ± Standard deviation)
Embryos (hemizygous)*	53.73 ± 0.09
F1 seed flour (transgene donor: ♀ parent)	59.07 ± 0.02
F1 seed flour (transgene donor: ♂ parent)	38.83 ± 0.04
Pollen (from a hemizygous plant)	52.82 ± 4.25

Matrix – Commercial samples	MON810 copy number ratio (Average ± Standard deviation)
Salvaterra honey	52.63
Flowers honey**	24.02

* Model for F_2 flour
** Also positive for GM oilseed rape and NK603

Decision Tree for domestic maize



Conclusions

- The combination of qPCR and plasmid calibrants allowed to differentiate different maize tissues ploidies..
- This study confirms that PCR is not a helpful technique to separate by itself pollen from flour in honey.
- However, when dealing with single origin honey, it might be coupled with other information to determine the most likely sources of GM presence.

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