

## **AGR1504: Using Synchrotron Methods to Detect Heat Resistant Pea – Pollen and leaf wax structure**

Peas are a pulse crop that is sensitive to temperature stress, particularly heat. Temperatures reaching 32°C in the field cause abortion of flower buds, flowers, and pods, reducing yield. In this grower-funded project, the University of Saskatchewan looked at lipids (fats, oils, and waxes) in the waxy surface or cuticle layer of pea leaves and pea pollen grains. Previously we found that pollen grains are sensitive to heat and that pea varieties may vary in pollen robustness, due to the types of lipids in their pollen coats.

The overall goal is to improve pollen in new varieties to withstand warmer field temperatures, so pea crops abort fewer flowers and maintain or improve their yield. The type and amount of lipid in pollen plays a role in enhancing pollination (likely through better pollen viability, attraction of pollen grains to the stigma, formation of the hydration lens, and pollen tube germination), and setting seeds and pods. In addition, the types of pollen lipids may be related to lipids in pea leaf wax, and because leaves are easier to sample, we want to develop a method to find more heat resistant pollen using the lipids in leaves. To do this we used a spectral technique at the Canadian Light Source which scans samples in the mid-infrared (Mid-IR) wavelength range (the red end of the rainbow to infrared). We looked at leaf wax and pollen grains from 11 pea varieties exposed to five days of high temperature (35/18 °C day/night) and compared them to samples from plants grown at normal temperatures (24/18 °C day/night). The method uses Mid-IR attenuated total reflectance (ATR) - Fourier transform infrared (FTIR) spectroscopy with uni- and multivariate spectral analyses to investigate chemical-structural features of samples.

Pollen work is technically difficult and slow compared to work on pea leaf. Growth chamber and field grown peas have similar pollen grains, but heat affects pollen lipids in subtle ways. Varieties were grouped into three groups based on their pollen lipids. Pea varieties also differ in leaf cuticle lipids. CDC Vienna exhibited the most lipids and CDC Sage had the least. Heat stress caused chemical modifications of leaf cuticles in 03H107P04HO2026, CDC Golden, CDC Sage, Naparnyk and TMP15213. A few days at higher temperature affects the esters and breaks down some of the long chain fatty acids in leaf cuticle wax. The varietal difference in lipid performance in heat means that we have potential for heat tolerance. The varietal range in pollen coat lipids is less than in leaf lipids.

We also looked at 2-dimensional and 3-dimensional X-ray imaging, like those used in medical imaging, but we made minor progress in detecting pollen tubes growing in young flowers and pods during the process of pollination. We then successfully measured tiny pods, their interior ovules (developing seeds), and the size of the funiculi (placenta) to ovules, processes soon after pollination that are difficult to measure by conventional techniques. Heat stress damage reduced ovule sizes and slowed pod growth.

The next steps are to analyze the lipids present in pollen coats and pea leaves using mass spectroscopy in a new project, and validate the Mid-IR measurements, then screen large numbers of pea varieties for heat stress resistance using Mid-IR.