OC5. Pollen composition discrimination by ATR-FTIR Spectroscopy.

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Bee pollen is a mixture of different pollens grain collected in different flowers that improve a different composition (Campos *et al*, 2008) and different colors. Bee pollen is known by its rich nutritional composition, is composed of proteins, lipids, sugar, fiber, minerals, amino acids, phenolic compounds and vitamins.

FTIR-ATR spectroscopy represents a useful technique for identifying chemical structures, which is based on the analysis of absorption peaks at certain wave numbers (expressed in cm^{-1}).

The aim of this study is compare different spectral regions of pollen from different floral sources in order to predict differences in the chemical composition. 16 different bee pollen samples with different botanical origin were used.

FTIR-ATR spectra were acquired with a Bruker FT-IR spectrometer (Alpha) using a diamond single reflection attenuated total reflectance (ATR) device and a zero filling of 2. Duplicate spectra per sample were obtained with 32 scans per spectrum at a spectral resolution of 4 cm^{-1} in the wavenumber range from 4000 to 400 cm^{-1}. Principal component analyses (PCA) were performed using OPUS Quant 2 (Bruker Optics, Ettlingen, Germany).

Second-derivatives of FTIR spectra are generally used to improve resolution in the original FTIR spectra. So, in the spectral data analyze was used the second derivation and selected the zones of proteins, lipids, carbohydrates and fibers absorption to better interpret the spectra.

The pollen spectra present significant differences in several spectral zones which indicate a different chemical composition.

The pollen spectra present a band appeared between 3650 and 3000 cm^{-1} corresponding to hydrogen bonded O–H stretching vibrations and two well defined peak at 2926 cm^{-1} due to C–H stretching vibrations and 2500 cm^{-1}. Between bands at 1200–970 cm^{-1} are mainly due to C–C and C–O stretching in pyranoid ring and to C–O–C stretching of glycosidic bonds. There are also a higher variability with different peaks on region between 1700 and 1200 cm^{-1} corresponding to aromatic and sugar compounds. These spectral zones were already identified for these groups of compounds by Coimbra et al. (1998) and Gómez-Ordóñez and Rupérez (2011).

The PCA analysis of the transformed spectra shows clearly the difference between samples. It was possible to group the 16 samples in different classes concerning the peaks area and the compound groups. This allows creating a classification model that identifies an unknown pollen sample. In the future it will be possible to make the calibration with the standard chemical methods in order to have a quantification of the bee pollen compounds by FTIR-ATR. This analysis could be promising, because is need only a pollen grain as sample.

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2) Coimbra, MA; Barros, A; Barros, M; Rutledge, DN; Delgadillo, I (1998) Multivariate analysis of uronic acid and neutral sugars in whole pectic samples by FT-IR, *Carbohydrate Polymers* 37 (3) 241–248.
3) Eva Gómez-Ordóñez, E; Rupérez, P (2011) FTIR-ATR spectroscopy as a tool for polysaccharide identification in edible brown and red seaweeds, *Food Hydrocolloids* 6 (25) 1514–1520.

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