8. Summary

Vibrational spectroscopy as well as ^{13}C NMR spectroscopy supported by multivariate chemometric techniques are valuable tools for qualitative and quantitative analysis of raw plant material and pharmaceutical preparations.

In this thesis, I presented a scheme of qualitative and quantitative analysis for selected raw plant material, starting with the preparation of samples and ending with the validation of calibration models. Models enabling quantification of the components of the analyzed systems and determination of their important parameters. I presented advantages of spectroscopic techniques over other commonly used analytical methods. Spectroscopic analytical methods are much simpler, faster and cheaper than currently used chromatographic, spectrophotometric and electrochemical ones, which require extraction of the quantified substances. Vibration spectroscopy allows for sample analysis in its native form. With heterogeneous samples, it is only necessary to grind them. Additionally, the measurement time required to collect IR and Raman spectra is several times shorter than that for a typical chromatographic measurement. It is significant that a single spectrum allows for the determination of values of many parameters for the analyzed sample.

Spectra of raw plant material are usually difficult to interpret. Extracting the desired information from them is not easy. Therefore, it is necessary to use advanced chemometric techniques. Preprocessing of spectra can help improve the signal-to-noise ratio and reduce the influence of baseline instability and variability of selected measurement parameters. I obtained many of the presented results using SNV or MSC corrections and by differentiating the spectra. Application of PCA analysis helped me detect outliers and select validation samples.

The combination of the reference analysis results with the Raman, IR and NMR spectra of the analyzed samples enabled me to determine the content of a number of substances and the values of selected physicochemical parameters for the tested samples. While I built PLS calibration models, it was important to select the appropriate spectral ranges characteristic of the determined groups of compounds. To characterize the developed models

and check their predictive abilities, I calculated the relative standard errors of prediction for calibration and validation sets.

Multivariate analysis of IR and Raman spectral data allowed me to construct PLS models for quantification of polyphenols, tannins, ellagitannins, procyanidins, agrimonin, 3-O-gallloylquinic acid and catechins in the rhizomes of Potentilla tormentilla. Models developed based on Raman and NIR spectra turned out to be slightly more accurate than those created on the basis of ATR and DRIFTS data.

Using the Raman, NIR, ATR, DRIFTS and ^{13}C NMR spectra of the ground bee pollen samples, I was able to determine the content of protein, fat, sugars and polyphenols as well as ABTS antioxidant activity and the pH of samples with $RSEP_{val}$ errors in the range of 1.2-3.8%. The presented results show that based on a single spectrum of a powdered sample of bee pollen, recorded using one of the mentioned techniques, important parameters of this complex natural product can be determined without the need for extraction.

Gas chromatography is the most important analytical technique used to control the quality of essential oils. However, its limitations in the analysis of non-volatile substances should be noted. It is also necessary to remember that the chromatographic measurement takes about 30 minutes. Vibrational spectroscopy can facilitate and speed up quantitative analysis of essential oils. I demonstrated this for peppermint oils by developing, based on Raman and ATR spectra, calibration models that enable quantification of menthol, menthone and menthyl acetate present in peppermint oils. These substances usually account for about 90% of the oil's weight.

Using the DRIFTS, Raman and NIR spectra, I was able to quantify diosmin in 8 pharmaceutical preparations, containing from 66 to 92% of an active substance, with an error of less than 2.4%. The concentration of diosmin in tablets determined based on PLS models is consistent with the results of reference analyses, with a recovery of 99.5–100.5%. The quality of determinations is comparable for the three methods used.

The developed protocols are based on a single spectrum of a powdered sample, which significantly shortens and simplifies the analysis. The presented methods are environmentally friendly, fast and reliable and can replace commonly used methods of analysis in the food and pharmaceutical industry.