Characterisation of Faecal Matter of Small Land Animals by Fourier-Transform Infrared Spectroscopy

Kamil Stepniak Supervised by John Keary Atlantic Technological University - Galway

Aims and Objectives

- To obtain and contrast FTIR spectra of each animal.
- Identify functional groups and compare correlation values of the spectra.
- Assess the impact of sterilisation methods on the spectra.
- Extract the organic content using dichloromethane (DCM).

Introduction

Fourier-transform Infrared Spectroscopy (Figure 1) is an analytical technique that can be used for identification of unknown materials that can be present at crime scenes or can be a source of environmental contamination. It can be used to analyse samples both qualitatively and quantitatively. IR spectra of compounds are often very characteristic and contain information about the functional groups that are present within the sample. When FTIR is used in conjunction with other analytical methods it can be used to determine the structure of any unknown sample (1).

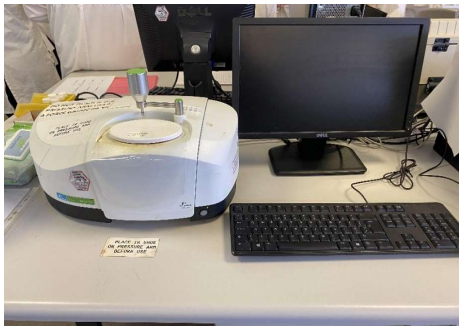

Figure 1: Perkin Elmer FTIR instrument set up

Experimental Details

- Samples were obtained from 3 different hamsters, one guinea pig and rabbits. The first step in sample preparation involved sterilising the droppings using different methods; microwave, autoclave and UV light irradiation.
- Adequate amount of each sample was placed on the FTIR crystal and spectra were recorded in % transmittance mode. Spectra were then overlayed and compared.
- The DCM extractions were carried out by placing the sample in a HPLC vial filled with 1mL DCM. The solution was stirred and left to extract for 2 hours.

Results

In the following spectrum (Figure 2) the peaks observed at 1035cm^{-1} and 1635cm^{-1} most likely indicate the presence of an amine (C-N), the two peaks visible at 2950cm^{-1} and 2850cm^{-1} correspond to -CH stretch vibrations of fatty acid methylene residues (2).

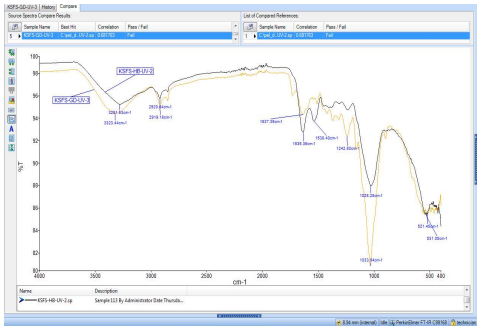

Figure 2: Overlayed spectra of sample HB and sample GD

Hamster samples sterilised using UV light (Figure 3) had the highest peak intensity and best-defined peak shape. In rabbits the autoclave yielded the best results and there was no trend observed in the guinea pig.

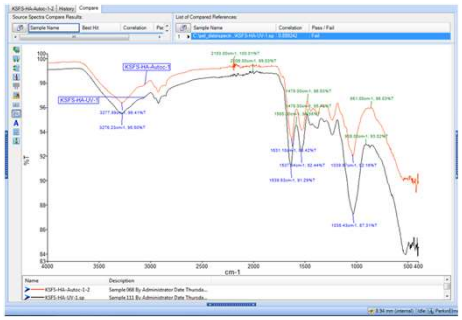

Figure 3: Overlayed spectra of sample HA

Correlation values of the overlayed spectra were obtained and presented in table 1 below. The correlation value corresponds to the degree of similarity between the two spectra. When hamsters were compared to guinea pigs, the correlation was low and ranged from 0.61 to 0.68. Correlation values of rabbit and hamster samples ranged from 0.81 to 0.89.

Table 1: Correlation values of the best selected spectra across the different samples

	HA-UV-1	HB-UV-2	HC-UV-2	GD-UV-3	RE-Autoc-1
HA-UV-1	1.00	0.95	0.96	0.61	0.81
HB-UV-2	0.95	1.00	0.95	0.68	0.84
HC-UV-2	0.96	0.95	1.00	0.62	0.84
GD-UV-3	0.61	0.68	0.62	1.00	0.89
RE-Autoc-1	0.84	0.84	0.81	0.89	1.00

The Figure 4 below shows the spectrum of rabbit sample and the DCM extractant spectrum. It can be observed that the two peaks that correspond to fatty acids methylene residues have much higher peak intensity.

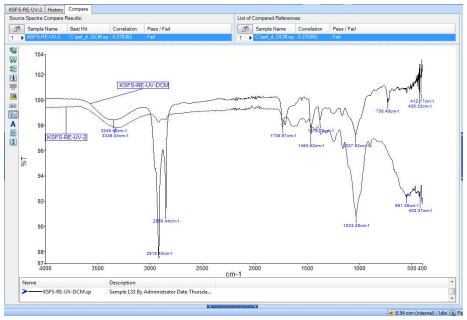

Figure 4: Overlayed spectra of sample RE and sample RE extracted with DCM

Conclusion

To conclude, there are some characteristics peaks that are shared in all samples and the spectra vary in their peak shape and peak position. The correlation values can be used to distinguish the hamsters from guinea pigs and rabbits, but not rabbits from the guinea pig. UV light irradiation has worked best on hamster samples in terms of spectra quality and autoclave worked best for rabbits.

Acknowledgements and References

1. Rees, O.J. (2010) *Fourier Transform Infrared Spectroscopy : Developments, Techniques, and Applications*. New York: Nova Science Publishers, Inc (Chemical Engineering Methods and Technology).
2. De Koninck, A.-S. *et al.* (2016) 'Detailed faecal fat analysis using Fourier transform infrared spectroscopy: Exploring the possibilities', *Clinical Biochemistry*, 49(16–17), pp. 1283–1287.

Acknowledgements: John Keary, Lab technicians, Anita Talbot and the Marine Research Centre at ATU, Dr. Damian McNeely and the whole team at Analytical services laboratory,