

Microspectroscopic infrared specular reflection studies of multi-component urinary stones at beamline D7 at the MAX IV laboratory, Lund Sweden.

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Abstract—At the beamline D7 at the MAX III ring at the MAX IV laboratory we have a setup for infrared microspectroscopy which is usable from 30 to 10000 cm^{-1} . In this project specular reflection infrared microspectroscopy was used for chemical imaging of cross-sectioned urinary stones, to determine their chemical composition and morphology simultaneously. Absorption spectral bands were recovered from reflection spectra by Kramers-Kronig transform. Use of far-infrared radiation provides high-contrast images and allows more precise constituent distribution determinations than mid-infrared because band asymmetry after the transform caused by diffuse reflection is less in the far-infrared.

I. INTRODUCTION

Urolithiasis – a condition when stones form in urinary tract. Its causes remain unclear despite many studies. The stones' composition and morphology (constituent distribution, size, shape, colour, and crystalline structure) vary. The components most frequently found are calcium oxalate (monohydrate and dihydrate), apatite, struvite, other phosphates, uric acid, and their mixtures. Knowledge of the composition is crucial to determine the underlying pathology, prescribe treatment, and prevent recurrences.

In this work, the potential of multi-spectral IR (mid-infrared and far-infrared) chemical imaging for urinary stone analysis is demonstrated. Chemical imaging is used for heterogeneous samples analysis, providing simultaneous information about their composition and morphology, i.e., distribution of the constituents. MIR and FIR chemical imaging are compared. We demonstrate that for rough samples such as cross-sectioned urinary stones FIR chemical imaging can be advantageous due to diminished diffuse reflection.

II. RESULTS

MIR and FIR imaging of a cross-sectioned urinary stone containing uric acid ($49 \pm 5\%$) and calcium oxalate ($51 \pm 5\%$) is presented. Non-normalized MIR and FIR absorption spectra are compared in Fig. 1. Baseline correction by the standard rubber band method was applied prior to analysis. Spectral bands characteristic of each component are denoted by dotted lines. As can be seen from Fig. 1, calcium oxalate absorbance is comparable in both MIR and FIR regions and is approximately 1.7. The FIR absorbance of uric acid, however, is weaker than in the MIR by 0.5 AU. The component bands are much better separated in the FIR than in the MIR (Figs. 1A, 1B). FIR microspectroscopy was used for the analysis of urinary stones for the first time. FIR microspectroscopy in specular reflection mode can be used to perform chemical and structural analysis of cross-sectioned

urinary stones, which are difficult to study by MIR microspectroscopy both in transmission mode due to their thickness and in reflection mode due to the effects of diffuse reflection. As FIR radiation has a longer wavelength than surface roughness dimensions the recorded spectra are purely specular reflection and KKT can be applied to calculate the absorbance. Different stone types can be distinguished from the FIR spectra with the same accuracy as from conventional MIR spectra. Quantitative accuracy depends on evaluation of the integrated band intensity. For well separated bands it is approximately 5%. Chemical images formed from KKT transformed FIR spectra have high contrast and represent the constituent distribution with a high level of detail. FIR chemical images have higher quality than MIR chemical images.

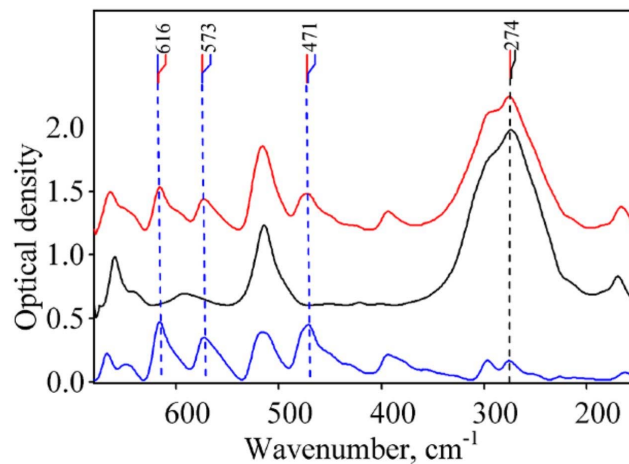
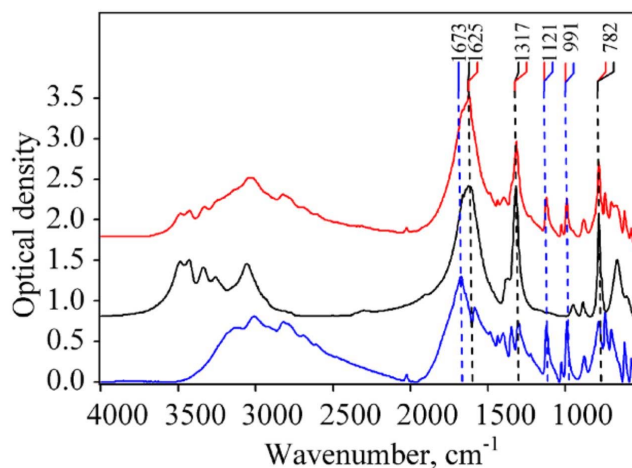


Fig 1. Non normalized absorption spectra of urinary stone containing uric acid and calcium oxalate. A. Upper frame: KBr-pellet.MIR. B. Lower frame PE pellet FIR. Top curve -urinary stone, middle curve -pure calcium oxalate, bottom curve pure uric acid.

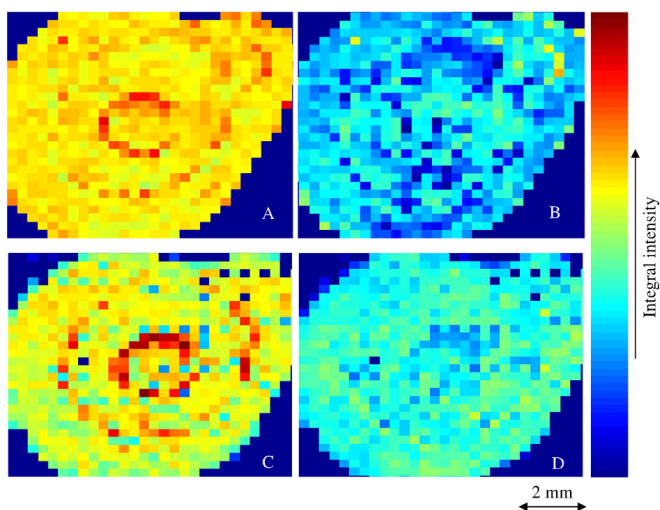


Fig. 2. Chemical images of the cross-sectioned urinary stone constructed prior to performing the *KKT*: A – MIR image of calcium oxalate (integrated band - *nas*(C-O) at 1318 cm^{-1}); B – MIR image of uric acid (integrated band - *ns*(C-C) at 1121 cm^{-1}); C – FIR image of calcium oxalate (integrated band - γ (O=C-C=O) at 274 cm^{-1}); D – FIR image of uric acid (integrated band - γ (N-H) at 472 cm^{-1}). Red represents the highest and blue the smallest concentration.

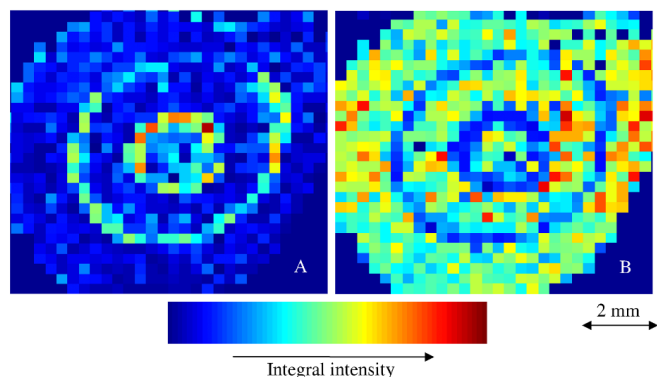


Fig. 3. Chemical images of the cross-sectioned urinary stone formed from *KKT* corrected FIR reflection spectra: A – FIR image of calcium oxalate (integrated band - γ (O=C-C=O) at 274 cm^{-1}); B – FIR image of uric acid (integrated band - γ (N-H) at 472 cm^{-1}). Red represents the highest and blue the smallest concentration.

The chemical images obtained from the raw MIR and FIR spectra are compared in Fig. 2. The cross-section area was approximately 9.3×8.3 mm and the mapping interval was 300×300 μm . These images represent the distribution of uric acid (Figs. 2A, 2C) and calcium oxalate (Figs. 2B, 2D)). When raw reflection spectra are used the information is similar for MIR and FIR images: the component boundaries are not clear and the contrast is low. The component structure seen in the optical view is hardly distinguishable in the corresponding chemical images. To obtain more informative images and better determine the stone structure the reflection spectra should be *Kramers-Kronig* transformed.

Fig. 3 shows images formed from the FIR reflection spectra after applying the *KKT*. The image contrast is better than in Fig. 4 and more structural details can be distinguished. In

addition, the images correspond well with the optical image in Fig. 4: the ring structure seen in the optical image is confirmed to be layers of calcium oxalate and uric acid.

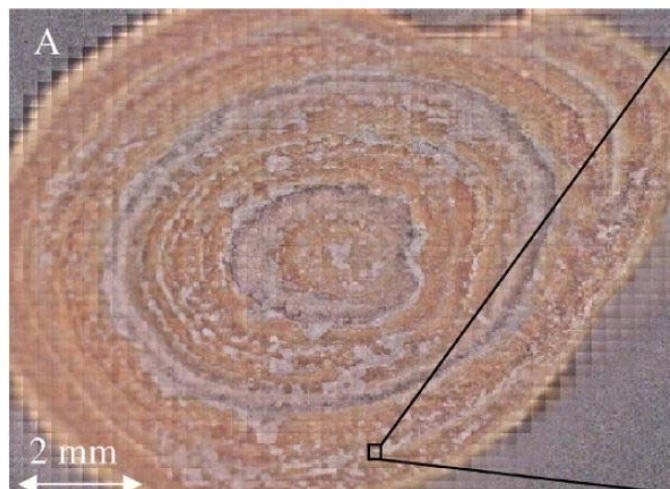


Fig. 4. Cross-sectioned uric acid - calcium oxalate urinary stone surface : A – optical image obtained by mapping.

III. SUMMARY

Specular reflection FIR imaging is superior to MIR imaging for rough ($2 - 25$ μm for MIR radiation) surfaces. Therefore, before multi-spectral IR analysis of urinary stones one should evaluate the roughness and choose the appropriate method.

FIR microspectroscopy was used for the analysis of urinary stones for the first time. FIR microspectroscopy in specular reflection mode can be used to perform chemical and structural analysis of cross-sectioned urinary stones, which are difficult to study by MIR microspectroscopy both in transmission mode due to their thickness and in reflection mode due to the effects of diffuse reflection. As FIR radiation has a longer wavelength than surface roughness dimensions the recorded spectra are purely specular reflection and *KKT* can be applied to calculate the absorbance. Different stone types can be distinguished from the FIR spectra with the same accuracy as from conventional MIR.

REFERENCES

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