

Raman reflector kit for Tip-Enhanced Raman Scattering (upright-TERS) on opaque samples

Combining Raman spectroscopy with AFM in order to correlate the information of both techniques and enhance the optical resolution has been an important demand in modern Raman spectroscopy. Based on the JPK NanoWizard® AFM and an inverted microscope, Tip-enhanced Raman Scattering (TERS) has successfully been used in various fields of research. Analysis of proteins, lipid bilayers, viruses and human cells as well as studies of silicon nanowires or the monitoring of catalytic processes were performed by various research groups worldwide¹⁻⁶. Up to now, such TERS measurements were restricted to transparent samples. To overcome this limitation JPK Instruments designed a Raman reflector kit which enables the TERS analysis also of opaque samples on inverted microscopes.

Raman reflector kit

All AFMs of the JPK NanoWizard® product family can be equipped with the Raman reflector kit in order to enable TERS measurements on opaque samples.



Fig. 1: Picture of the upright-TERS microscope kit.

The kit contains a designated cantilever holder, a sample holder and, optional, a suitable long working distance objective (e.g. Olympus, WD 10.6 mm, NA 0.5). The kit is designed for the use with all common inverted research microscopes (Olympus, Zeiss, Nikon and Leica). The beam path for the excitation laser and the TERS signal is shown in Figure 2. The laser light (green line) is guided

around the opaque sample and reflected by the dichroic mirror above the TERS tip. The laser is focused in such a way that the focal plane matches that of the tip on the sample. The Tip-Enhanced Raman scattering signal (red line) at the TERS tip is reflected and collected in the same way.

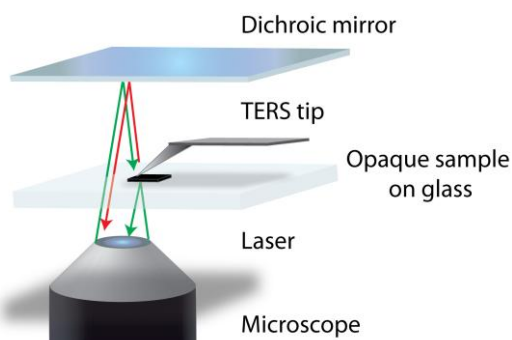


Fig. 2: Scheme of the beam path when utilizing the Raman reflector kit for TERS on an inverted microscope.

Example experiment

For an upright-TERS experiment a monolayer of p-nitrothiophenol on a smooth layer of 100 nm Au on mica was used as sample. A diode laser with an excitation wavelength of 532 nm ($P < 1$ mW) and an acquisition time of 1 s was employed. For data accumulation a 200 x 200 nm grid (10 x 10 points) was used. Homogeneous spectra were collected of the reacted compound with constant signal enhancement. Representative spectra are shown in Figure 3.

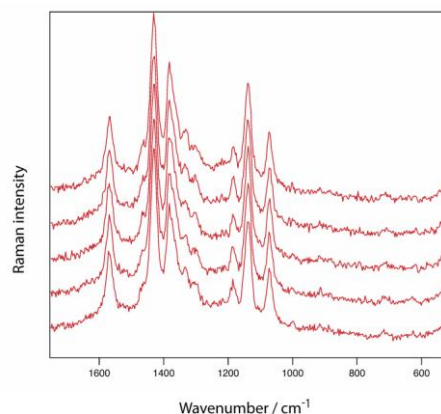


Fig. 3: TERS spectra of p-nitrothiophenol collected in upright configuration.

Additionally reference measurements were performed. The experiment was repeated in standard transmission configuration under similar conditions and with the same TERS tip. Again a Monolayer of p-nitrothiophenol was used as sample but this time on optical transparent gold nanoplates (height < 30 nm). By using the same excitation source ($\lambda = 532 \text{ nm}$, $P < 1 \text{ mW}$, $t_{\text{acq}} = 1 \text{ s}$) and a 40x oil immersion objective (NA 0.9) TERS spectra comparable to the upright-TERS spectra were collected. Figure 4 displays some of the collected spectra.

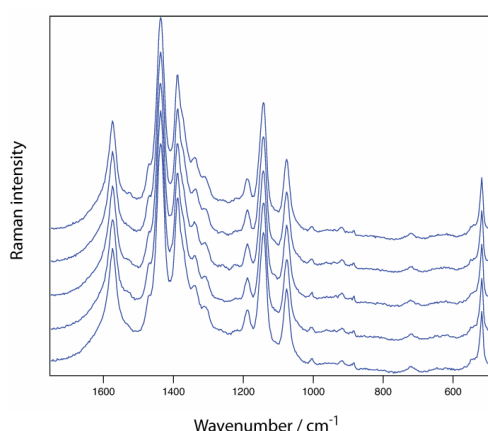


Fig. 4: Reference measurement: TERS spectra of p-nitrothiophenol measured with same TERS tip in standard transmission configuration.

Conclusion

It is clearly visible that both measurements yield constant signal enhancements and that the illumination geometry does not influence the spectra. The Raman reflector kit does not affect the band position and allows reproducible spectra. The results shown are in perfect agreement with results presented in literature (4). The Raman reflector kit can be incorporated into existing transmission setups in order to allow TERS analysis of opaque samples in an extremely straightforward and simple way.

Application fields for TERS on opaque samples

- Single molecules such as DNA or proteins
- Organic thin film composition studies
- Biomaterials surface properties
- Properties of chemical modified surfaces
- Geological materials

Specifications

- Compatible with common inverted microscope Raman spectrometer setups
- **Sample holder:**
 - Raman reflector kit sample holder
- **Stages:**
 - JPK TAO™ modules
- **Objective lenses:**
 - e.g. Olympus LMPLFLN50x 0.5NA 10.6 mm WD
 - Similar objective lenses from other suppliers are possible

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