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Detection and spectroscopy of single molecules in rare gas matrices: dibenzanthanthrene in krypton and xenon

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Abstract

Fluorescence microscopy coupled with matrix isolation techniques have allowed the observation of dibenzanthanthrene molecules in rare gas matrices. Fluorescence excitation spectra of single molecules were measured in both Kr and Xe matrices. Spectral lines with Lorentzian shapes as well as irregular noisy profiles were observed. © 1999 Elsevier Science B.V. All rights reserved.

1. Introduction

Significant advances have been made in the optical detection and spectroscopy of individual molecules. Most importantly, single-molecule techniques provide information on the properties of molecules which would be difficult or impossible to obtain from measurements simultaneously involving many molecules. While standard measurements probe large ensembles of molecules and yield the mean values of parameters of interest, single-molecule measurements completely remove the ensemble averaging and allow the construction of a histogram of distributed values for an experimental parameter. Clearly, such a distribution contains more information than a mean value alone. Details of the underlying distribution may reveal whether a property under investigation is inhomogeneous or not. Consequently, single-molecule spectroscopy and single-molecule detection have matured into powerful instruments of optical spectroscopy and have led to a wealth of scientific information which is summarized in comprehensive reviews covering this new and highly exciting field [1–9].

Detection of the signal from just one molecule is accomplished with ultrasensitive fluorescence techniques and by experimental designs which limit the excited volume. The need for a careful choice of chromophores with appropriate photophysical properties puts a serious limitation on the number of systems that can be investigated by single-molecule spectroscopy. High-resolution spectroscopic studies are generally conducted at cryogenic temperatures. After a decade of single-molecule spectroscopy in solids, ~ 20 guest/host combinations are known, as

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recently summarized in Ref. [5]. Most of the investigated guest impurities belong to the class of rigid conjugated hydrocarbons. Among the most extensively studied have been pentacene, perylene, terrylene, dibenzanthanthrene (DBATT) and their derivatives. So far, mainly organic hosts have been used: crystals such as *p*-terphenyl, anthracene, naphthalene, various *n*-alkane matrices like hexadecane, dodecane and octane, and polymers like polyethylene and polyvinylbutyral [10]. Single-molecule spectra can also be measured using fluorescence microscopy, a method which is especially suited for investigating many molecules in parallel and analyzing the positions of molecules within a sample [11–14].

We recently combined fluorescence microscopy with classical matrix isolation procedures which resulted in the observation of single terrylene molecules in vapour deposited *n*-decane and *n*-hexane solids [15]. Based on these results, we started to investigate whether single molecules could be detected in rare gas matrices. Isolation of guest molecules in such matrices is very promising for the investigation of guest–host interactions because the structure of such samples should be simpler and more predictable by theoretical models. Many investigations on the gas–solid borderline have been performed with rare gas clusters [16]. Hole burning experiments have been conducted in Ar, Kr and Xe matrices doped by phthalocyanine [17]. The use of rare gas matrices has also been recently proposed in the context of possible detection of fluorescence from single molecular ions [18].

Due to a considerably smaller solvent shift, the electronic transitions of terrylene embedded in rare gas matrices are significantly less shifted to the red than in organic crystals and fall outside the spectral range easily accessible with a standard single-mode dye laser. Therefore, we chose dibenzanthanthrene (DBATT) – intensely investigated and found to be especially well-suited for single-molecule spectroscopy due to its photophysical properties [19].

2. Experimental

Dispersed fluorescence and fluorescence excitation spectra of DBATT in rare gas matrices were measured with a fully computer-controlled total luminescence spectroscopy (TLS) system consisting of two double grating SPEX 1402 monochromators [20]. Samples were deposited onto a sapphire window attached to a cold finger in an optical cryostat and kept at 4.2 K. Xe matrices for electronic absorption spectroscopy were prepared in a Displex closed cycle refrigerator (Air Products) and analysed at 20 K with a Shimadzu UV 3100 spectrophotometer.

Details of the experimental apparatus for the single-molecule detection and spectroscopy are described elsewhere [12,15]. Mixtures of a rare gas with high-temperature vapours of DBATT were sprayed directly onto the surface of the mirror objective surface thus forming a matrix. The objective (numerical aperture (N.A.) = 1 and a magnification of 80) was mounted in an electrolytic copper holder and the whole assembly was attached by screws to the cold finger of the cryostat.

The matrix deposition was preceded by pre-cooling the objective down to ~ 40 K (Kr) and ~ 60 K (Xe). These relatively elevated temperatures are recommended [21] to obtain the good optical quality of rare gas solids. Then the DBATT crystals were heated inside the nozzle and the rare gas flow (Kr or Xe) was started. When a stable nozzle temperature of ~ 510 K was attained, the objective surface was exposed to the stream of gas (flow rate ~ 1 mmol/h) for 1 min. The presence of DBATT in the sample was checked by using a tunable dye laser for fluorescence excitation and an intensified video camera for

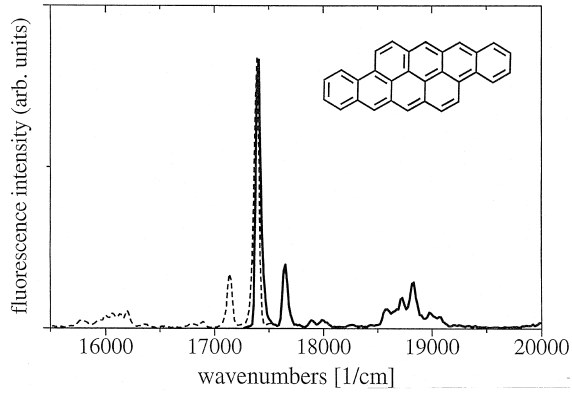

Fig. 1. Fluorescence excitation (solid line, observed at 17150 cm^{-1}) and dispersed fluorescence (dashed line, excited at 17635 cm^{-1}) spectra of DBATT in a Kr matrix at 5 K. The monochromator resolution was 30 cm^{-1}.

detection. The deposition procedure was repeated when fluorescence was not detectable. The roughly estimated upper limit for the ratio of guest to host molecules was 1:10 000. After the deposition, the cryostat was filled with liquid helium and the helium vapour was pumped off to reach a temperature of ~ 2 K.

The optical arrangement was the same as described in Ref. [15]. The beam of a cw single-mode Rh6G dye laser (spectral bandwidth 2 MHz) was focused on the matrix surface close to the objective axis. The excitation spot diameter was ~ 50 μm; an intensity of 0.2 W/cm^2 gave a satisfactory signal-to-noise ratio. The resulting images, produced by individual fluorescing molecules, were recorded with a video camera equipped with an image intensifier (Hamamatsu C2400-25). Rayleigh-scattered pump radiation was blocked by Schott RG610 glass filters placed between the cryostat window and the camera. To obtain the spectral positions and line shapes of individual molecular resonances, the laser was scanned over 4 GHz in 4 MHz steps with an integra-

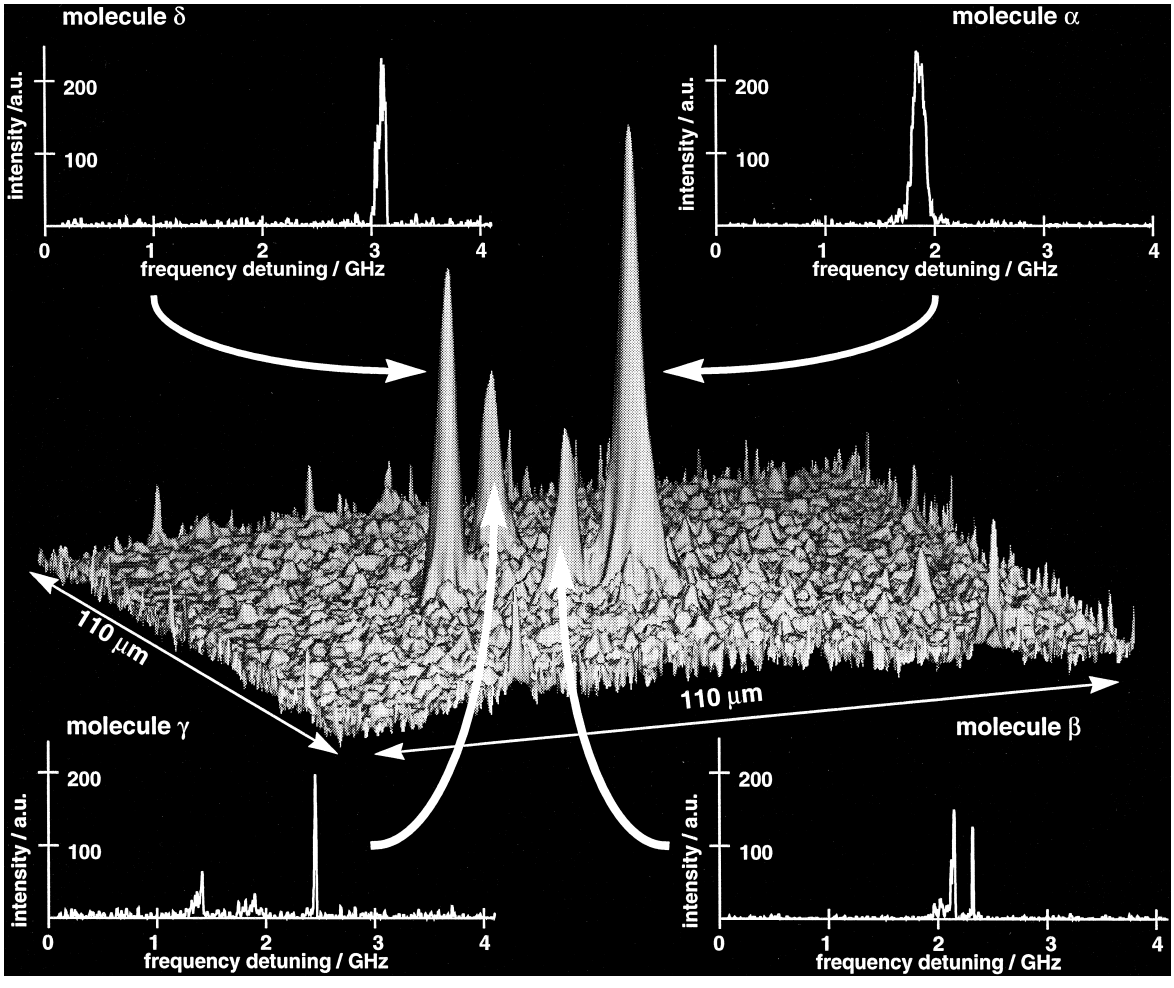

Fig. 2. The three-dimensional representation of an integrated fluorescence microscopy image (summation of all video-frames acquired during a frequency scan) around 17 368 cm^{-1} for a DBATT/Kr matrix. Insets show the fluorescence excitation profiles of 4 single DBATT molecules. Molecule α produces a regular Lorentzian line with fwhm ~ 100 MHz, molecules β, γ and δ provide the examples of 'spectral jumps' (see text).

tion time of 0.32 s per video-frame at each frequency point.

3. Results and discussion

Our first experiments with bulk samples of DBATT molecules isolated in various rare gas matrices demonstrated that the spectral position of the 0–0 electronic transition changed significantly with the host, being around 18 180 cm^{-1} (550 nm) in Ne, 17 605 cm^{-1} (568 nm) in Ar, 17 390 cm^{-1} (575 nm) in Kr and 17 065 cm^{-1} (586 nm) in Xe. Such a systematic red-shift of 0–0 molecular electronic bands with the increasing mass of host atoms is commonly observed and is ascribed to the increasing polarizability of the host [17]. For single-molecule studies, Kr and Xe hosts were most favourable since our excitation source, a linear single-mode dye laser, operated in the 568–600 nm range.

Fig. 1 presents the dispersed fluorescence and fluorescence excitation spectra of DBATT, as measured by the TLS apparatus in krypton. The 0–0 transition around 17 390 cm^{-1} is identified by the overlap of the main bands of the dispersed fluorescence and fluorescence excitation spectra. The bandwidth of the inhomogeneous 0–0 transition, ~ 60 cm^{-1}, is 4 times broader than that reported for the hexadecane matrix [19], the vibronic structure, however, is similar.

After the preparation of a Kr matrix doped with DBATT directly on the objective surface (see Section 2), blinking spots were identified with the video camera, when excited within the spectral range of 0–0 electronic transitions (17 420–17 360 cm^{-1}). The continuous irradiation of the sample led to the disappearance of some spots in tens of seconds. Small manual laser detuning of several tens of MHz caused dramatic changes in the detected pattern. Disappearing spots were substituted by other ones. These observations allowed the identification of individual point-like images recorded by the camera as being due to selectively excited single DBATT molecules.

To obtain the information on spatial positions of all spots appearing during the narrow band laser scan, the collective image was produced in each experiment as a sum of all detected fluorescence microscopy images. The example, for a 4 GHz scan, is presented in Fig. 2. The overall size of the investigated sample area was 110 $\mu m \times 110$ μm. Four prominent, well-resolved peaks can be noticed. These are very sparse and the probability of finding randomly distributed molecules closer to each other than the optical resolution (~ 3 μm) is negligibly low. We conclude that each of them belongs to separate molecules denoted α, β, γ and δ. Analysis of the fluorescence signal from these molecules as a function of the laser frequency leads to the four plots included in Fig. 2. The spectral profile of α exhibits a Lorentzian shape. The three other molecules, however, show an irregular dependence on the excitation frequency. Out of 11 molecules observed in 4 independent frequency scans, only two had regular Lorentzian shapes with fwhm (full width at half maximum) ~ 100 MHz (the one in Fig. 2) and ~ 70 MHz. It should be noted that the laser beam intensity of ~ 0.2 W/cm^2 causes a significant saturation broadening of line widths. As demonstrated for DBATT in a hexadecane Shpol'skii matrix at 1.8 K [19], an intensity of 0.1 W/cm^2 led to line widths of the order of 50 MHz, i.e. 3 times broader than under unsaturated conditions. Considering that some broadening is also due to the relatively high matrix temperature (~ 4 K [15]), the ~ 100 MHz line widths are not surprising. About 80% of the analysed molecules had irregular profiles (like β, γ and δ in

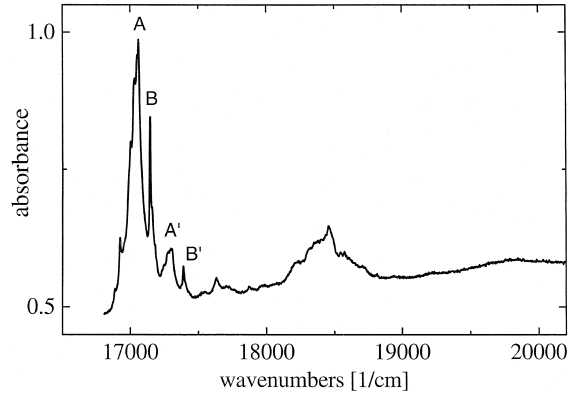

Fig. 3. Electronic absorption spectrum of DBATT in a Xe matrix at 20 K. The substantial background comes from the scattering properties of the sample. A and B denote the two most prominent site bands (out of several discernible in the spectrum), A′ and B′ are their vibrationally excited counter-parts.

Fig. 2) with characteristic sudden disappearances and reappearances of emission. A fluctuation of the fluorescence intensity may arise from a 'spectral jump', i.e. the 0–0 frequency change of a single molecule which would abruptly take it out of resonance. For the molecules β and γ one can even observe more than one such jump. In these cases the resonance frequency changed, by chance, within the spectral range covered by the scanning laser, so the molecule could be observed to fluoresce again. Depending on the guest/host combinations, both spontaneous and light-induced spectral diffusion have been reported [6]. Further studies are needed to understand the exact nature of these jumps in our experimental system.

Another guest/host combination preliminarily tested in this work was the DBATT/Xe matrix. Fig. 3 shows the electronic absorption spectrum of this solid deposited on a sapphire window and kept at 20 K. In a rather congested origin around 17 000 cm^{-1}

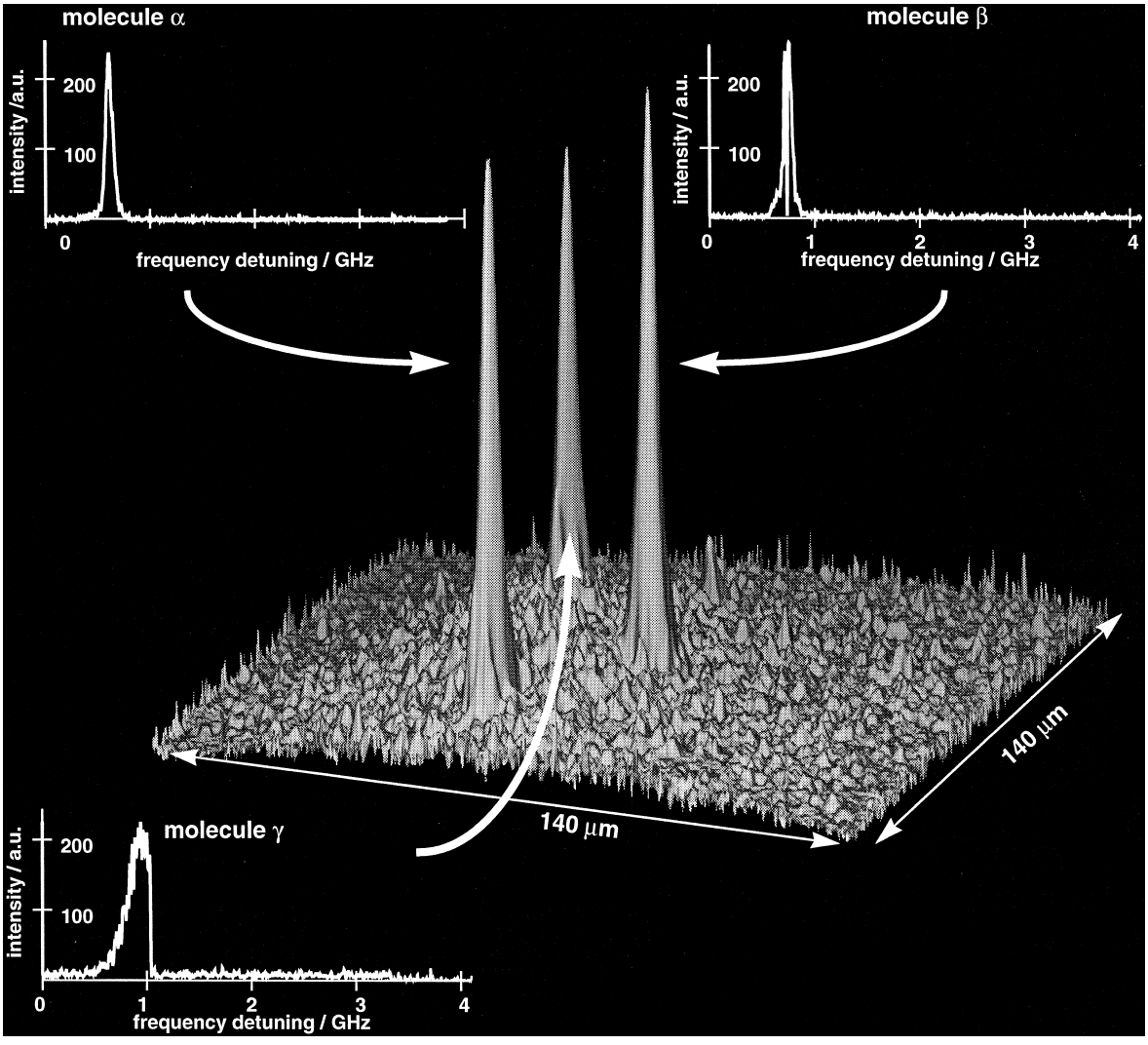

Fig. 4. The three-dimensional representation of an integrated fluorescence microscopy image similar to Fig. 2, frequency scan around 17 055 cm^{-1} for a DBATT/Xe matrix. Insets show the fluorescence excitation profiles of 3 single DBATT molecules. Molecule α produces a Lorentzian line with fwhm ~ 80 MHz, molecules β and γ exhibit 'spectral jumps'.

the two prominent bands, marked A and B, are easily discernible. These features, together with other sharp bands in the vicinity of A and B, can be assigned to the 0–0 transitions for DBATT molecules occupying different matrix sites. From the fluorescence excitation spectrum of DBATT in krypton (which does not exhibit the specific site structure), we learned that the 0–0 electronic transition is followed by a relatively strong vibronic band spaced at 245 cm^{-1} (see Fig. 1). Indeed, for the DBATT/Xe absorption the A′ and B′ bands, separated by 245 cm^{-1} with respect to A and B, strongly support our assignment.

Several bright spots were detected in the focal plane, when the DBATT/Xe matrix was deposited directly on the objective surface and excited around the maximum of absorption band A. This is illustrated by the collective image for one of 4 GHZ scans (Fig. 4). An analysis of the fluorescence signal from the α, β and γ molecules leads to the three plots included in Fig. 4. Molecule α has a Lorentzian profile with a fwhm of $\sim$80 MHz. The line width for β is similar to that for α, though a sudden drop followed by a fast recovery of the fluorescence intensity was recorded. For molecule γ, the disappearance of emission took place after reaching the maximum so one could still measure the line width which was $\sim$4 times broader than that for α.

Xenon matrices, when compared to similarly excited krypton matrices, gave almost the same intensity of the single-molecule fluorescence. This point deserves some attention, since the well-known heavy-atom effect usually leads to the fluorescence quenching [22] by an increase of ISC rates. For DBATT molecules however, as observed in a hexadecane matrix, the relaxation rate of the T_1 triplet level to the S_0 singlet ground state is 4500 s^{-1}, i.e. three times higher than the $S_1 \rightarrow T_1$ rate (according to Boiron et al. [19]), which effectively diminishes the importance of the dark 'triplet trap'. The negligible role of the heavy-atom effect in our system seems consistent with these Shpol'skii matrix experiments.

In conclusion, our experiments have demonstrated the possibility of single-molecule detection in rare gas matrices using the optical fluorescence microscopy. There are many interesting issues which can be studied for such new systems, such as the temperature dependence of line width distributions, the nature of spectral jumps (the role of both spontaneous and light-induced spectral diffusion), the relation of the single-molecule orientation to its saturation intensity, etc. One can expect that further investigations will result in a better understanding of guest–host interactions and will provide new insight to the basics of the rare gas matrix isolation technique.

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References

[1] W.E. Moerner, T. Basché, Angew. Chem., Int. Ed. Engl. 32 (1993) 457.
[2] W.E. Moerner, Science 265 (1994) 46.
[3] W.E. Moerner, Acc. Chem. Res. 29 (1996) 563.
[4] M. Orrit, J. Bernard, R. Brown, B. Lounis, in: E. Wolf (Ed.), Progress in Optics, vol. 35, Elsevier, Amsterdam, 1996, pp. 61–144.
[5] T. Plakhotnik, E.A. Donley, U.P. Wild, Annu. Rev. Phys. Chem. 48 (1997) 181.
[6] T. Basché, W.E. Moerner, M. Orrit, U.P. Wild (Eds.), Single Molecule Optical Detection, Imaging, and Spectroscopy, Verlag-Chemie, Weinheim, 1997.
[7] S. Nie, R.N. Zare, Annu. Rev. Biophys. Biomol. Struct. 26 (1997) 567.
[8] X.S. Xie, J.K. Trautman, Annu. Rev. Phys. Chem. 49 (1998) 441.
[9] W.E. Moerner, M. Orrit, Science 283 (1999) 1670.
[10] B. Kozankiewicz, J. Bernard, M. Orrit, J. Chem. Phys. 101 (1994) 9377.
[11] F. Güttler, T. Irngartinger, T. Plakhotnik, A. Renn, U.P. Wild, Chem. Phys. Lett. 217 (1994) 393.
[12] J. Jasny, J. Sepiol, T. Irngartinger, M. Traber, A. Renn, U.P. Wild, Rev. Sci. Instrum. 67 (1996) 1425.
[13] H. Bach, A. Renn, U.P. Wild, Chem. Phys. Lett. 266 (1997) 317.
[14] T. Irngartinger, A. Renn, G. Zumofen, U.P. Wild, J. Lumin. 76,77 (1998) 279.

[15] J. Sepiol, A. Starukhin, T. Yu. Latychevskaia, J. Jasny, A. Renn, U.P. Wild, Special Issue of Chemical Physics, Single Molecule Spectroscopy in Physics, Chemistry and Life Sciences (in press).
[16] S. Leutwyler, J. Bösiger, Chem. Rev. 90 (1990) 489.
[17] P. Geissinger, D. Harrer, Chem. Phys. Lett. 197 (1992) 175.
[18] N. Caspary, V. Palm, K.K. Rebane, V.E. Bondybey, Chem. Phys. Lett. 283 (1998) 345.
[19] A.-M. Boiron, B. Lounis, M. Orrit, J. Chem. Phys. 105 (1996) 3969.
[20] F.A. Burkhalter, E.C. Meister, U.P. Wild, J. Phys. Chem. 91 (1987) 3228.
[21] L. Andrews, M. Moskovits (Eds.), Chemistry and Physics of Matrix-Isolated Species, Elsevier, Amsterdam, 1989.
[22] J.B. Birks (Ed.), Organic Molecular Photophysics, vol. 2, Wiley, London, 1975.