References

1. McKellar A R W, Xu Y, Jäger W *J. Phys. Chem. A* **111** 7329 (2007)
2. McKellar A R W *J. Chem. Phys.* **128** 044308 (2008)
3. Xu Y, Blinov N, Jäger W, Roy P-N *J. Chem. Phys.* **124** 081101 (2006)
4. McKellar A R W *J. Chem. Phys.* **127** 044315 (2007)
5. Tang J, McKellar A R W *J. Chem. Phys.* **119** 754 (2003)
6. McKellar A R W *J. Chem. Phys.* **125** 164328 (2006)
7. Surin L A et al. *Phys. Rev. Lett.* **101** 233401 (2008)
8. Topic W et al. *J. Chem. Phys.* **125** 144310 (2006)
9. Dumesh B S, Surin L A *Usp. Fiz. Nauk* **176** 1137 (2006) [*Phys. Usp.* **49** 1113 (2006)]
10. Toennies J P, Vilesov A F *Angew. Chem. Int. Ed.* **43** 2622 (2004)
11. Makarov G N *Usp. Fiz. Nauk* **174** 225 (2004) [*Phys. Usp.* **47** 217 (2004)]
12. Grebenev S, Toennies J P, Vilesov A F *Science* **279** 2083 (1998)
13. Chuaqui C E, Le Roy R J, McKellar A R W *J. Chem. Phys.* **101** 39 (1994)
14. Surin L A et al. *J. Chem. Phys.* **112** 4064 (2000); "Errata" *J. Chem. Phys.* **112** 9190 (2000)
15. Ginzburg V L, Sobyanin A A *Pis'ma Zh. Eksp. Teor. Fiz.* **15** 343 (1972) [*JETP Lett.* **15** 242 (1972)]
16. Tejeda G et al. *Phys. Rev. Lett.* **92** 223401 (2004)
17. Moroni S et al. *J. Chem. Phys.* **122** 094314 (2005)
18. Brookes M D et al. *Spectrochim. Acta A* **60** 3235 (2004)
19. Xu Y, Jäger W *J. Chem. Phys.* **106** 7968 (1997)
20. Surin L A et al. *Rev. Sci. Instrum.* **72** 2535 (2001)
21. Rusin F S, Bogomolov G D *Pis'ma Zh. Eksp. Teor. Fiz.* **4** 236 (1966) [*JETP Lett.* **4** 160 (1966)]
22. Surin L A et al. *J. Mol. Spectrosc.* **222** 93 (2003)
23. Škrbić T, Moroni S, Baroni S *J. Phys. Chem. A* **111** 7640 (2007)
24. von Haeften K et al. *Phys. Rev. B* **73** 054502 (2006)
25. Andronikashvili E L *Zh. Eksp. Teor. Fiz.* **16** 780 (1946); **18** 424 (1948)
26. Paesani F, Kwon Y, Whaley K B *Phys. Rev. Lett.* **94** 153401 (2005)
27. Paesani F et al. *J. Chem. Phys.* **122** 181106 (2005)

PACS numbers: **61.43.–j**, **78.47.–p**, **78.55.–m**
DOI: 10.3367/UFNe.0179.200903n.0322

Single molecules as spectral nanoprobes for the diagnostics of dynamic processes in solid media

A V Naumov, Yu G Vainer

In this report, we briefly review the results of works directed at the development and use of the method of single-molecule spectroscopy (SMS) for investigating dynamic processes in disordered solid-state media.

The first optical experiments on the detection of single-quantum systems were carried out in the mid-1970s in the gas phase with rarefied atomic beams and single ions in electromagnetic traps [1]. An important step in the way of recording spectra of single molecules (SMs) in condensed media was the development of methods of selective laser spectroscopy of impurity centers, i.e., the method of the excitation of fine-structure fluorescence spectra and of burning-out stable spectral holes [2]. We note that both these scientific avenues were developed for the first time at the Institute of Spectroscopy, Russian Academy of Sciences (ISAN).

The first experimental electronic absorption spectra of single chromophore molecules introduced into a solid-state matrix were observed by Moerner and Kador in 1989 [3]. Soon after this, in 1990, Orrit and Bernard [4] showed that when recording individual spectra of SMs by the method of excitation of fluorescence spectra in the optical range, the signal-to-noise ratio radically increases in comparison with the case of absorption spectra [4]. The possibility of the direct measurement of optical spectra of SMs in solid-state matrices opened the way to setting up various experiments. Since that time, systematic studies of condensed media with the application of the SMS method (see [5] and the references therein) have begun. This area of studies is being vigorously developed now. According to the Web of Science (database of the Institute of Scientific Information, USA), the number of scientific publications devoted to this field in the journals analyzed by the Web of Science was more than 1300 in 2007 alone.

One of the most promising avenues of the SMS method in studies of the properties of condensed media is the use of isolated impurity molecules as spectral nanoprobes for obtaining information on dynamic processes in the local environment of these molecules. The main idea of this approach is that the parameters of an electron transition of an impurity chromophore molecule embedded into a solid-state matrix are very sensitive to the parameters of the local environment of the molecule. The inertialess nature of this spectral probe allows conducting studies with a maximally high time resolution (up to several femtoseconds), and the smallness of the probe dimensions allows obtaining information about the medium on a nanoscopic scale. As a consequence, the optical spectra of impurity molecules contain unique data on the local structure of the matrix and, most importantly, on dynamic processes that occur in the nearest vicinity of such molecules.

The most important advantage of the SMS method is that it allows completely removing the effect of ensemble averaging over a large number of impurity centers, which is inherent in conventional methods of selective laser spectroscopy. Moreover, in contrast to the known methods of investigating the dynamics of solid-state media (based on acoustic or thermodynamic measurements, or on the registration of Raman spectra, inelastic neutron scattering, etc.), the SMS method allows investigating dynamic processes at the level of single molecules and, thus, almost completely removing the averaging over the volume of the sample investigated. This advantage plays a fundamentally important role in the investigations of strongly disordered solid-state media (glass, polymers, polycrystals, nanostructured objects, biological media, and other complex systems), because in such media, as a result of the significant microscopic inhomogeneity of the substance, the parameters and the nature of the local dynamics can differ radically at different points of a sample.

We consider the basic ideas of the low-temperature dynamics of disordered solid-state media in more detail. Numerous studies have demonstrated that at low temperatures, the dynamic characteristics of such media differ significantly from the appropriate properties of crystals. It has also been proved that the observed anomalous properties of disordered media have a universal nature and are practically independent of the concrete structure and chemical composition of the substance. In the specialized literature, diverse disordered media frequently are therefore simply called glasses.

In 1971 [6, 7], a fruitful idea was proposed on the existence of specific elementary low-energy excitations in such disordered solid-state media, namely, of so-called tunneling two-level systems (TLSs). According to the model suggested in these works, which was called the standard model of low-temperature glasses, the jumps in the TLSs (the tunneling transitions initiated by phonons of groups of atoms or molecules between two lower levels in double-well energy potentials) make the basic contribution to the dynamics of glass at temperatures $T < 1$ K.

The model of TLSs allows describing the majority of dynamic phenomena in glasses observed at temperatures $T < 2-3$ K. The anomalous dynamic properties of glass that are manifested at higher temperatures (up to several tens of kelvins) are usually ascribed to the presence of *quasi-localized low-frequency vibrational modes* (LFMs) in disordered solid-state media (see, e.g., [8 – 12]), the modes that can be regarded as vibrations of groups of atoms or molecules in a single-well potential.

The soft-potential model (SPM) describes the TLSs and LFMs in the framework of a common approach [13, 14] and can be considered a generalization of the TLS and LFM models. The soft-potential model allows describing the dynamics of glasses in a wider temperature range than the standard model of glasses. However, this model, which is finding ever increasing acceptance among researchers, just like the TLS model, is purely phenomenological and cannot describe all dynamic phenomena observed in glass.

To describe the effect of jumps in a TLS on the electronic transition frequency (and therefore on the optical spectrum) of an impurity molecule a very simple model called the *stochastic random-jump model* is widely used [15]. For a more rigorous analysis of a TLS – chromophore-molecule interaction, some authors use the dynamic approach (see, e.g., [16]). In the stochastic model, the transitions in a TLS adjacent to the chromophore molecule lead to sharp changes in the frequency of the electron transition of this molecule. The transition frequency of a single molecule at an instant t is determined by the totality of *independent* transitions in all TLSs surrounding this molecule:

$$\omega(t) = \omega_0 + \sum_j \zeta_j(t)\, \upsilon_j\,, \tag{1}$$

where the stochastic variable $\zeta_j(t) = 0$ ($\zeta_j(t) = 1$) if the jth TLS at the instant t is in the ground (excited) state and ω_0 is the frequency of the 0 – 0 transition of the SM under the condition that all TLSs that interact with the SM are in the ground state. In the case of a dipole – dipole type of the TLS – chromophore-molecule interaction, the quantity υ_j is expressed as

$$\upsilon_j \sim \frac{\Delta\mathbf{M}_j\, \Delta\mathbf{M}_{\mathrm{OM}}}{r_j^3} = 2\pi\Lambda\, \frac{A_j\, \varepsilon_j}{E_j\, r_j^3}\,, \tag{2}$$

where $\Delta\mathbf{M}_j$ is the change in the dipole moment of the TLS caused by its jump, $\Delta\mathbf{M}_{\mathrm{OM}}$ is the change in the dipole moment of the impurity molecule caused by its transition from the ground to the excited state, r_j is the distance between the TLS and impurity molecule, Λ is the constant of the TLS – chromophore interaction, ε_j is the orientational parameter, A_j is the parameter of asymmetry of the TLS, and E_j is the energy splitting of the TLS.

In the experiment, the interaction between an SM and an active TLS is manifested (depending on the relation between the characteristic frequency of jumps and the time of measurement) as spectral jumps or a broadening of spectral lines, as well as in the form of spectrum splitting.

The observed width of a separate spectral peak is determined by the interaction of the chromophore with vibration-type excitations (LFMs and acoustic phonons) and with a large quantity of remote TLSs. A quadratic electron – phonon interaction of the chromophore with the LFM leads to a uniform broadening Γ and a frequency shift of spectral peaks in the spectrum of the SM. The broadening Γ is usually described by a formula [16] that is valid in the case of a weak LFM – chromophore coupling,

$$\Gamma = \frac{w \exp\left(-\Delta E/kT\right)}{\left[1 - \exp\left(-\Delta E/kT\right)\right]^2}\,, \tag{3}$$

where ΔE is the energy of the LFM, w is the constant of the quadratic LFM–chromophore interaction, and k is the Boltzmann constant. Thus, the spectrum shape of the impurity molecule, as well as the temperature and time dependences of its characteristics, contains information on the parameters of elementary excitations in the local environment of this molecule and on the interaction of these excitations with this molecule.

In spite of the significant effort expended in the study of the low-temperature dynamics of glass, the majority of fundamental questions in this field remain open. First of all, this concerns the microscopic nature of the dynamic processes that occur in such media (in particular, the microscopic nature of TLSs and LFMs). The problems of the applicability limits of the models suggested (the overwhelming majority of which are purely phenomenological); the problems of phenomena observed in some experiments that cannot be described in the framework of these models; and the problem of the connection between the local dynamics of glass and the concrete structure and chemical composition of the medium studied remain unsolved.

Single-molecule spectroscopy revealed unique possibilities to achieve essential progress in resolving these problems, because this method allows investigating the dynamics of the medium at the level of single molecules (up to the measurement of the individual parameters of single elementary excitations). The first measurements [17] of spectra of SMs in an amorphous polymer carried out in 1994 already afforded the first direct experimental proof of the existence of tunneling TLSs in such media.

Unfortunately, the development of the SMS as the method of studying the dynamics of complex disordered solid-state systems met with a number of essential difficulties, namely, the low value of the signal-to-noise ratio realizable in such measurements, which is determined by the smallness of the light signals coming from one molecule; the presence of the incidental radiation of the matrix; the presence of phonon wings in the individual spectra of molecules; and, as a result, quite a long time necessary for the reliable registration of the signal. Some difficulties were also related to the limited number of impurity systems in which it was possible to register the spectra of SMs. Significant problems appeared upon analysis and interpretation of the spectra. In particular, serious problems appeared in attempts at a correct statistical processing of the experimental spectra of SMs because of an insufficient elaboration of the corresponding procedures.

In the series of works performed in 2000 – 2008 that are discussed in this report, we substantially developed the procedure for registering SM spectra and a whole series of techniques for analysis of the data obtained, including the

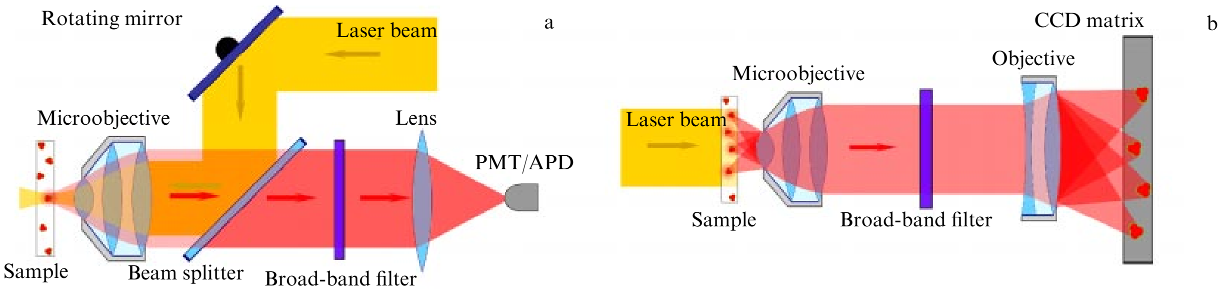

Figure 1. Schemes for the registration of the excitation spectra of SM fluorescence: (a) single-channel scheme with the use of a photomultiplier (PMT) or an avalanche photodiode (APD); and (b) multichannel scheme with a highly sensitive matrix photodetector based on a cooled CCD matrix (CCD: charge-coupled device).

procedure for the multichannel registration of the excitation spectra of the fluorescence of SMs and for the computer processing of the images of these molecules, as well as the procedure for statistical analysis of the parameters of a large number of SM spectra. This allowed eliminating many important problems of using SMS for studying the dynamics of impurity solid-state media and thus allowed achieving significant progress in the study of the fundamental laws that govern the low-temperature dynamics in disordered solid-state media (organic glasses and polymers).

Until recently, schemes of registration of signals with a single-channel photodetector [18] (photomultiplier or avalanche photodiode) were typically used for the experimental registration of the fluorescence excitation spectra of SMs in solid-state media. In recent years, experimental SMS installations for measurements at low temperatures have been constructed based on a confocal microscope. A typical setup with single-channel photodetection in a confocal regime is outlined in Fig. 1a. For the excitation of a fluorescence spectrum, a single-frequency tunable laser whose beam is focused on the sample by means of a high-aperture microobjective is typically used. The fluorescent radiation is collected with the same microobjective and is directed to the photodetection system. Broad-band interference filters and/or cutting-off light filters reject the scattered laser radiation with the nonshifted frequency and transmit the Stokes component of the fluorescence spectrum of the detected molecules. Then the radiation is directed onto the photosensitive surface of the single-channel photodetector.

Usually, *the excitation spectra of SM fluorescence* (the dependence of the integrated intensity of the SM fluorescence on the frequency of the exciting laser radiation) are recorded in experiments; these spectra can be considered the equivalent of the absorption spectrum. Retuning to provide measurements in another region of the sample in this scheme is achieved by a displacement of the sample or with the aid of a scanning mirror.

The spectra of SMs in amorphous media at low temperatures can consist of several peaks and vary randomly in time. The complex and mainly random shape of the SM spectra is caused by transitions in nearest TLSs and by any other structural rearrangements in the matrix in the local environment of an SM. A much more informative and more convenient method of measurement of such spectra is the use of a technique of a frequently repeated registration of the excitation spectra of SM fluorescence in a selected spectral range [19]. The results of such measurements are usually represented in the form of a two-dimensional pattern (2D plot), a kind of 'time topograph' of the measured spectra (Fig. 2).

An analysis of such a two-dimensional picture allows unambiguously identifying spectral peaks that belong to different SMs [20–22]. Indeed, a transition in one TLS sufficiently 'strongly coupled' to a given SM manifests itself in the form of simultaneous and identical jumps in all spectral peaks that belong to the spectrum of this SM. At the same time, this transition either is not practically manifested in the spectra of other molecules, because they are typically located sufficiently far from the TLS in question, or is manifested in the form of shifts that differ from the above-mentioned one in terms of the amount of the displacement. Thus, in the two-dimensional picture, a *spectral trail* can be distinguished, which is frequently also called a *spectral trajectory*, corresponding to the time evolution of the individual spectrum of the chosen molecule.

An analysis of spectral trajectories gives important information on the processes in the matrix and changes in its parameters in the *local environment* of the observed chromophore molecule. For instance, as was shown in [20–22], the time evolution (broadening, frequency jumps, splitting of spectral lines) of the observed spectral trajectories of impurity molecules of tetra-*tert*-butylterrylene (TBT) introduced into amorphous polyisobutylene (PIB) is described well by the standard model of glasses. Namely, the splitting or the frequency jumps of the observed spectral lines of the SM between 2^N spectral positions (see Fig. 2) can be explained by the coupling of the observed molecule to N nearby TLSs (with the transitions in them leading to appropriate changes in the frequency of the spectral line). In turn, the observed broadening of spectral lines can be explained by the coupling of the impurity molecule to the vibrational excitations of the matrix and to a large number of TLSs located far from the impurity molecule (this coupling leads to only insignificant spectral jumps in comparison with the observed width).

The spectral trajectories of SMs observed in low-temperature experiments in disordered media can demonstrate a complex behavior, which may not always be explained in the framework of the standard model of glasses. For instance, in [22], cases were observed where an SM interacts with two TLSs, which, in turn, interact with each other. In some cases, spectral trails have been registered that indicate the existence of three-level systems in disordered solid-state media, as well as spectral trails that manifest themselves in a continuous frequency drift in the SM spectrum.

An unprecedented source of information about the local dynamics of the matrix can be the temperature dependence of

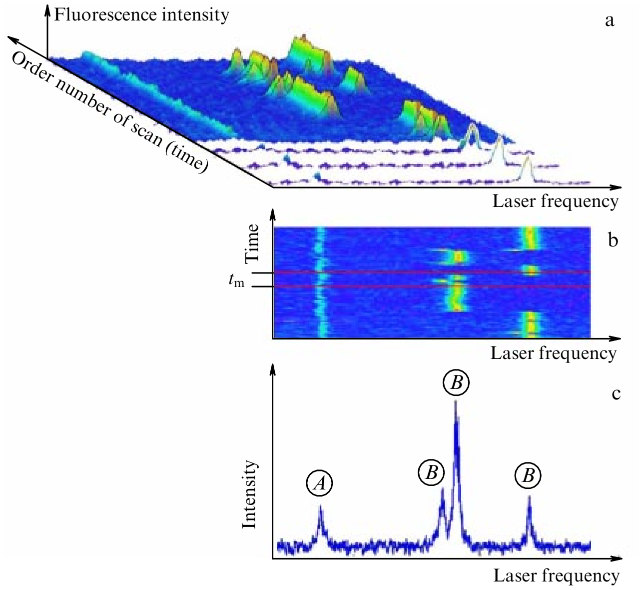

Figure 2. (a, b) Temporal evolution of single-molecule spectra of TBT embedded in PIB recorded repeatedly at $T = 2$ K. The results of measurements are presented in the form of (a) three-dimensional and (b) two-dimensional images. The color (gray) scales in (a) and (b) correspond to the intensity of fluorescence. The spectrum of molecule A does not reveal pronounced jumps during the time of observation. The spectrum of molecule B exhibits 'jumps' between four spectral positions (interaction with two nearby active TLSs). (c) The resulting integral spectrum, which is the sum of all spectra taken in the time interval $t_m = 120$ s (shown in Fig. 1b by horizontal lines). The total spectrum corresponds to two molecules: A (singlet) and B (triplet).

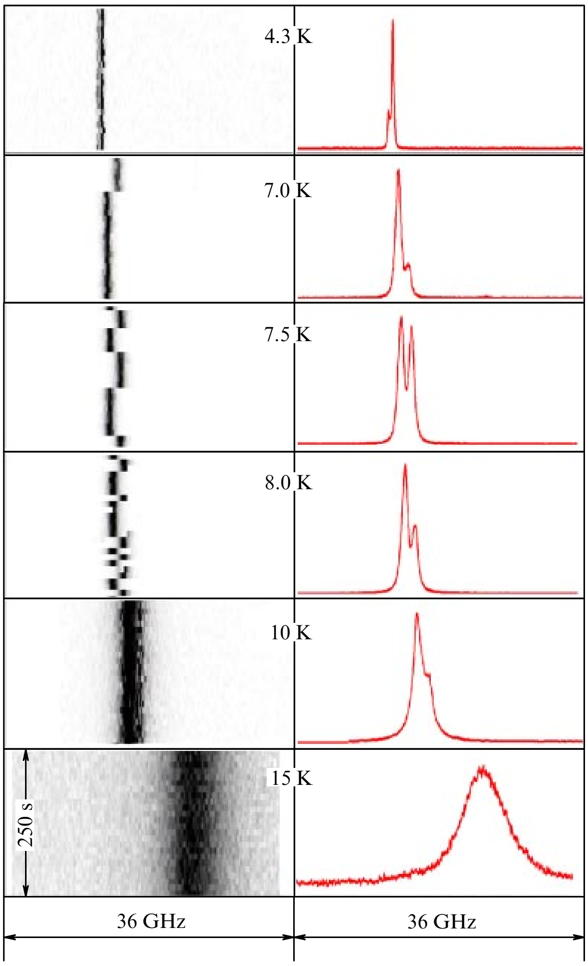

Figure 3. Spectral trails (left panel) and the corresponding total spectra (right panel) of the same single molecule of TBT embedded in PIB at various temperatures (borrowed from [25]).

the parameters of SM spectra. Figure 3 shows examples of spectral trails and the corresponding integrated spectra of the same SM measured at different temperatures. At $T = 7$ K, transitions in TLSs are activated that cause significant jumps of the spectrum between its two positions. In the range between 7 and 8 K, the characteristic rate of such jumps increases considerably. With a further increase in temperature, the jumps of spectral lines are masked with a strong broadening caused by the coupling of the SM to the LFMs. A shift of the SM spectrum with increasing the temperature is also observed.

As shown in [23–26], an analysis of the temperature dependence of the parameters of the SM spectra allows determining not only the local but also the individual parameters of single elementary excitations of a disordered medium. For instance, in [23], an analysis of the temperature dependence of the ratio of the intensities of two spectral components in the SM spectrum (which is split as a result of coupling of the selected SM to a TLS that suffered rapid jumps, in comparison with the time of measurement) allowed determining the asymmetry of this TLS (energy difference between the ground and excited states). It was also found that TLSs exist in which the populations of the energy levels do not obey the Boltzmann statistics. An analysis of the influence of an external electric field on the spectrum of an SM coupled to a TLS [27] allowed measuring the complete set of individual parameters of a single TLS, i.e., the tunneling parameter, the asymmetry, and the electric dipole moment.

An analysis of the individual temperature dependences of the widths $\Gamma(T)$ of peaks of the SM spectrum [24–26] permitted us to obtain data on the local parameters of LFMs in the nearest environment of impurity TBT molecules in amorphous PIB. We measured the $\Gamma(T)$ dependences in the temperature range from a few to several tens of kelvins, in which the predominant contribution to the processes of broadening of the SM spectra comes from the coupling to LFMs. The local parameters (the frequency of an LFM and the quadratic coupling constant of this mode to a given molecule) were determined by approximating each $\Gamma(T)$ dependence using formula (3). It was found that the frequencies of the LFM in this system have a significant dispersion, which can be regarded as a consequence of the localized nature of the LFM in this medium.

As can be seen from the above examples, the use of the SMS for measuring local parameters of solid-state media offers unique possibilities for the direct observation of dynamic processes in such media on the microlevel (up to the detection of elementary quantum mechanical events). Such measurements, undoubtedly, are of great interest from both the scientific and practical standpoints. They are especially important in the case of media characterized by strong local inhomogeneity.

To correctly relate the experimentally measured local parameters of a solid-state medium to its macroscopic (functional) characteristics, structure, and chemical composition and to correctly compare the results of the measurements of such parameters with theoretical estimations, the averaged values of these parameters must be known. Wide opportunities for determining these values are given by a statistical analysis of data on the parameters of SM spectra obtained from a large number of such spectra. This idea was first realized in a series of our works [20, 21, 28 – 31]. We developed an approach based on the use of moments and related derivative functions, (cumulants) for a quantitative description of the complex shapes of SM spectra in disordered solid-state media at low temperatures.

In [20, 21], we measured and calculated the distributions of the first four moments of SM spectra (spectral shifts, generalized width, asymmetry, and peakedness of the spectra). The distributions obtained were compared with the distributions of the moments of model SM spectra for the same system calculated in the framework of the stochastic theory of SM spectra in low-temperature glasses [32]. This allowed estimating the minimum distance between the impurity molecule and a TLS and thus to obtain information on the minimal size of a TLS in the system. Furthermore, the analysis carried out allowed determining the dispersion parameters of the TLS – SM coupling constant.

It was shown in Refs [28 – 30] that the distributions of the first two cumulants of the SM spectra in the impurity system of TBT in PIB at $T = 2$ K are satisfactorily described in the framework of the Lévy statistics (Lorenz and Smirnov functions for the respective distributions of the first and second cumulants). As was shown in theoretical work [33], this indicates the validity of the basic concepts of the standard TLS model assuming the dipole – dipole nature of the chromophore – TLS coupling and also the uniform distribution of TLSs. The original method of the analysis of distributions of the SM-spectrum moments that was developed and realized in [31] allowed us to determine (based on the example of an amorphous system of TBT in PIB, at $T = 2$ K) the characteristic distances (zone) where the coupling of a TLS to an impurity molecule mainly leads either to a splitting or to a broadening of the spectrum, and also the distance beyond which this coupling is practically unrevealed.

We note that in all studies related to the statistical analysis of the parameters of SM spectra, a problem of the statistical confidence of the results occurs [34]. This problem is especially acute when using the conventional single-channel detection scheme (Fig. 1a), in which the measurement of a statistically reliable number of spectra of different SMs requires an enormous amount of time, which is not always realizable. Furthermore, in such measurements, passing from one point of the sample to another requires a certain time, which prevents simultaneous observations of local dynamic characteristics at different points of the sample. These problems can be solved by using a multichannel measuring method, based on the scheme of a fluorescence microscope with a multichannel photodetector (Fig. 1b). In this case, the images of all SMs that fall into the field of view of the microobjective are recorded simultaneously with the aid of a matrix detector. This scheme permits implementing a parallel synchronous control over the spectral characteristics of all SMs whose electronic transition frequencies lie in the chosen frequency range of measurements.

The multichannel registration of SM spectra in solid-state media was first realized in works performed in 1998 – 2000 at the Swiss Federal Institute of Technology Zurich (ETH Zürich) [35]. However, the use of this equipment in practice was hindered for a number of reasons: (a) the matrix detectors at that time had low ultimate sensitivity and high noise; (b) the electronic and computer equipment did not allows ensuring a reasonable time resolution of experiments; (c) the registration of a significant number of SM spectra required a large disk space (several tens of gigabits); and (d) the processing of the measurement results took too long, even when using specialized electronic computing work stations. All these problems were acquiring a catastrophic scale in attempted measurements of the spectral trails of a large number of SMs in solid-state media. As a consequence, the procedure developed and the equipment designed did not receive wide acceptance.

The appearance of highly sensitive CCD cameras with internal electron multiplication and low noise, a significant increase in the efficiency of computers, the possibility of rapid writing/processing of significant arrays of data, and, most importantly, the development of original algorithms of the automated recognition of fluorescent images of SMs allowed us to realize a technique of multichannel detection of spectral trails of SMs with the aid of a fluorescence microscope at a qualitatively new level. The technique developed opened up new possibilities and allowed us to obtain a number of fundamentally new results, which are discussed below. In Refs [36 – 38], a technique of measurements of the frequency spectrum of LFMs (vibrational density of states, or VDOS) was proposed and developed in solid-state media from the spectra of SMs embedded in the medium to be studied. The technique was based on measurements and analysis of individual temperature dependences of the widths of separate spectral peaks of a large number of impurity molecules.

At each temperature, fluorescent images of the detected SMs were recorded synchronously with a retuning of the laser source frequency in the selected spectral range in the form of a sequence of video frames from a CCD camera. The digitized video frames were saved in the permanent memory of a computer in the form of a block of data, which were used for further analysis. The individual fluorescence excitation spectra of SMs observed in the field of view of the microobjective with a sufficiently high signal-to-noise ratio were extracted from the saved data using a specially developed computer program. It is significant that this procedure was automated. The program determined the positions of groups of adjacent pixels of the CCD matrix whose signals gave a spike at a certain frequency of the laser excitation, which could be interpreted as a spectral peak at this frequency (Fig. 4). Each such group of pixels was correlated with a luminescent image of a certain (ith) SM. The dependence of the intensity of fluorescence signals detected by a given group of pixels on the number of a frame (i.e., at the excitation frequency) is interpreted by the program as a fluorescence excitation spectrum of a given SM. The coordinates of the image of this SM, x_i, y_i, on the CCD matrix are determined as the coordinates of the centers of gravity of pixels in a given group (Fig. 4a). The approximation of separate spectral peaks in the measured SM spectrum by a Lorenz function allowed determining the width of the spectral line Γ_i of a given SM (Fig. 4b).

To obtain the temperature dependence of the widths of spectral lines of the observed SMs, the measurements were repeated at various temperatures in the range from 8 – 10 to 30 – 40 K, where the contribution from LFMs to the broadening of the spectral lines of SMs predominates over the contributions of other processes. In the course of measurements, a special control was performed of the constancy of the

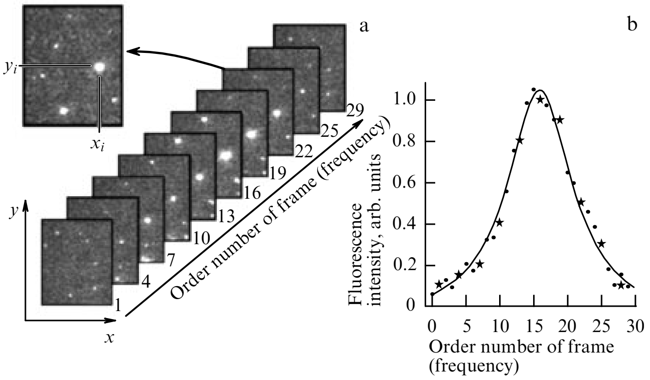

Figure 4. The procedure of extracting the SM spectrum from a sequence of video frames (schematic): (a) identification of luminescent images of the same single molecule upon scanning of the spectrum; and (b) the fluorescence excitation spectrum of a selected ith molecule with coordinates x_i, y_i (points) and its approximation by a Lorenz function (solid line). The asterisks correspond to the video frames shown in Fig. 4a (borrowed from [37]).

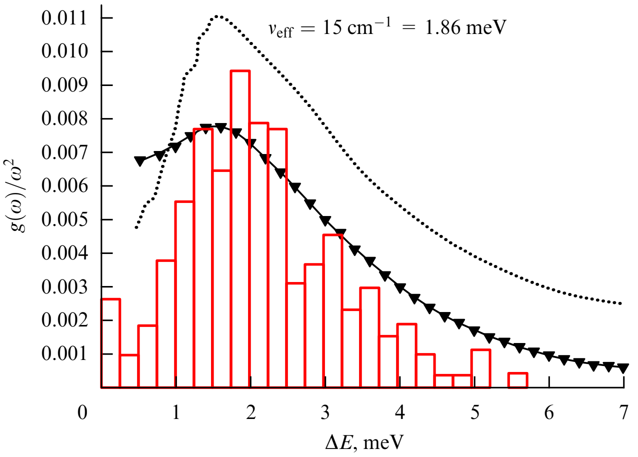

Figure 5. Energy spectrum of LFMs and the boson peak in frozen toluene measured using three experimental methods. Histograms represent the spectrum of LFMs in frozen toluene doped with TBT (measured by the SMS method). Triangles correspond to the spectrum of the density of vibrational states (VDOS, or the boson peak) measured for solid toluene doped with molecules of ferrocene (nuclear scattering method). The dotted line shows the VDOS spectrum measured in the undoped toluene at $T = 7$ K (Raman scattering). The arrow indicates the effective (averaged) energy of LFMs measured (by the photon-echo method) for solid toluene doped with molecules of zinc octaethylporphyrine.

coordinates of the molecules depending on temperature. The approximation of each measured temperature dependence using formula (3) allowed determining the local parameters of an LFM, such as the energy ΔE_i and the quadratic coupling constant w_i in the nearest nanovicinity of the ith SM.

Using the procedure developed, we measured the energy spectra of LFMs for amorphous PIB and frozen toluene doped with TBT molecules [36–38]. In [38], the measured energy spectra of LFMs were compared with the data on the VDOS (which is frequently called a boson peak in the literature) measured for pure PIB and toluene by inelastic neutron scattering, Raman scattering, and inelastic nuclear scattering. The measured LFM spectrum in [38] was also compared with the effective values of the LFM energy measured for the same matrices using the photon echo method.

A sufficiently good agreement of the parameters of the experimental spectrum of LFMs with the data on VDOS (Fig. 5) obtained by different methods allows formulating two important conclusions:

(1) the LFMs that cause the experimentally observed broadening of the spectral lines of isolated molecules of TBT embedded into PIB and toluene have the same microscopic nature as the vibrational excitations that determine the shape of the boson peak in impurity-free PIB and toluene;

(2) the introduction of small concentrations of TBT into PIB and toluene does not lead to noticeable changes in the vibrational dynamics observed in the matrix under investigation.

The technique of rapid multichannel detection of SM spectra and the equipment designed for this method allowed solving one more important and complex problem, namely, to significantly facilitate the search for new impurity matrix systems suitable for recording the excitation spectra of SM fluorescence at low temperatures. It should be remembered that in using a single-channel registration scheme, a serious problem exists in searching for an SM in both the coordinate in the field of view of the microobjective and the frequency within a wide inhomogeneous absorption contour. The multichannel detection scheme of fluorescent images substantially facilitates this problem, because in this case the need is eliminated in the spatial search for SMs in various regions of the sample. This opens up fundamentally new possibilities in investigations of the spectral dynamics of impurity solid-state media using SM spectra.

In [34, 39], we for the first time detected SM spectra in low-molecular glass (frozen solutions of TBT molecules in conventional and deuterated toluene) and obtained an unexpected result: we found that on the microscopic level, the dynamic processes in low-molecular glasses at $T < 2$ K, in contrast to the processes in amorphous polymers, cannot be adequately described by the standard model of glasses. We found an additional contribution to the spectral dynamics, which manifested itself in the form of continuous drifts and frequency-unrepeatable random jumps of the spectra of the SMs under consideration (Fig. 6). It is probably these effects that prevented the experimental detection of SM spectra in these liquids. It may be supposed that the unsuccessful attempts to measure SM spectra in other low-molecular liquids are also related to the same effects.

An analysis of the influence of the deuteration degree of toluene molecules (isotope effect) on the distribution of the widths of the spectral lines of SMs and on the energy spectrum of LFMs showed that the dynamics of this matrix at $T = 2$ K is determined by the excitations of the tunneling type, whereas at $T > 7$ K, the observed dynamics is determined by excitations of the vibrational type.

To summarize, we can conclude that single-molecule spectroscopy is a powerful tool for investigations of the dynamics of disordered solid-state media at a local level. The suggested technique of fast multichannel detection of SM spectra and the equipment design permitting the realization of the original methods of automated computer-aided processing of fluorescent SM images, as well as the related methods of statistical analysis of individual parameters of a large number of experimental spectra of SMs, open up qualitatively new opportunities for the investigation of the principal features and microscopic nature of dynamic processes in solids.

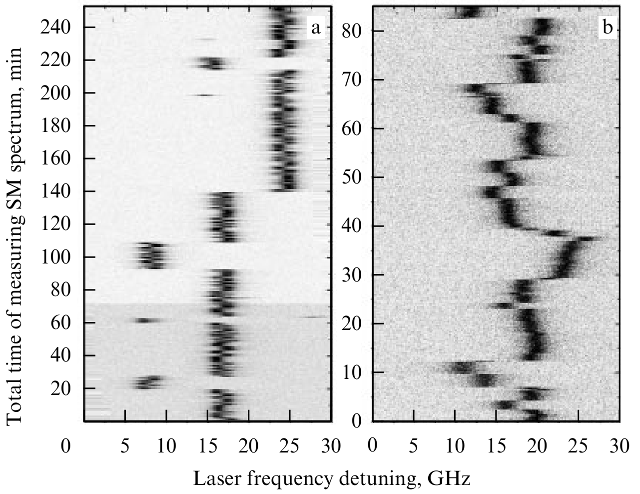

Figure 6. Spectral trails of single TBT molecules introduced into amorphous PIB (a) and a frozen solution of toluene (b), measured at $T = 7$ K (according to the data in [38]).

We are grateful to L Kador and M Bauer for their fruitful cooperation in the investigations performed in this work. This work was supported in part by the Russian Foundation for Basic Research grants Nos 07-02-00206 and 08-02-00147 and by the Deutsche Forschungsgemeinschaft, Sonderforschungsbereich. A.V.N. also appreciates the support from the Ministry of Education and Science of the Russian Federation and from the U.S. Civilian Research and Development Foundation (Basic Research and Higher Education Project), and also support from the Grant of the President of the Russian Federation (MK-239.2007.2).

References

1. Balykin V I et al. *Pis'ma Zh. Eksp. Teor. Fiz.* **26** 492 (1977) [*JETP Lett.* **26** 357 (1977)]
2. Personov R I, Al'shits E I, Bykovskaya L A *Pis'ma Zh. Eksp. Teor. Fiz.* **15** 609 (1972) [*JETP Lett.* **15** 431 (1972)]; Gorokhovskii A A, Kaarli R K, Rebane L A *Pis'ma Zh. Eksp. Teor. Fiz.* **20** 474 (1974) [*JETP Lett.* **20** 216 (1974)]; Kharlamov V M, Personov R I, Bykovskaya L A *Opt. Commun.* **12** 191 (1974)
3. Moerner W E, Kador L *Phys. Rev. Lett.* **62** 2535 (1989)
4. Orrit M, Bernard J *Phys. Rev. Lett.* **65** 2716 (1990)
5. Kulzer F, Orrit M *Annu. Rev. Phys. Chem.* **55** 585 (2004)
6. Anderson P W, Halperin B I, Varma C M *Philos. Mag.* **25** 1 (1972)
7. Philips W A *J. Low Temp. Phys.* **7** 351 (1972)
8. Elliott S R *Europhys. Lett.* **19** 201 (1992)
9. Laird B B, Schober H R *Phys. Rev. Lett.* **66** 636 (1991)
10. Schirmacher W, Diezemann G, Ganter C *Phys. Rev. Lett.* **81** 136 (1998)
11. Taraskin S N et al. *Phys. Rev. Lett.* **86** 1255 (2001)
12. Gurevich V L et al. *Pis'ma Zh. Eksp. Teor. Fiz.* **76** 650 (2002) [*JETP Lett.* **76** 553 (2002)]
13. Karpov V G, Klinger M I, Ignat'ev F N *Zh. Eksp. Teor. Fiz.* **84** 760 (1983) [*Sov. Phys. JETP* **57** 439 (1983)]
14. Parshin D A *Fiz. Tverd. Tela* **36** 1809 (1994) [*Phys. Solid State* **36** 1811 (1994)]
15. Klauder J R, Anderson P W *Phys. Rev.* **125** 912 (1962); Reinecke T L *Solid. State Commun.* **32** 1103 (1979)
16. Osad'ko I S *Selektivnaya Spektroskopiya Odinochnykh Molekul* (Selective Spectroscopy of Single Molecules) (Moscow: Fizmatlit, 2000) [Translated into English (Berlin: Springer, 2003)]
17. Kettner R et al. *J. Phys. Chem.* **98** 6671 (1994)
18. Tamarat Ph et al. *J. Phys. Chem. A* **104** 1 (2000)
19. Ambrose W P, Moerner W E *Nature* **349** 225 (1991)
20. Naumov A V et al. *Phys. Rev. B* **63** 212302 (2001)
21. Naumov A V, Vainer Yu G, Bauer M, Kador L *J. Chem. Phys.* **116** 8132 (2002)
22. Vainer Yu G, Naumov A V, Bauer M, Kador L *Opt. Spektrosk.* **94** 936 (2003) [*Opt. Spectrosc.* **94** 873 (2003)]
23. Bauer M, Kador L, Naumov A V, Vainer Yu G *J. Chem. Phys.* **119** 3836 (2003)
24. Naumov A V, Vainer Yu G, Bauer M, Kador L *Phys. Status Solidi B* **241** 3487 (2004)
25. Vainer Yu G, Naumov A V, Bauer M, Kador L *J. Chem. Phys.* **122** 244705 (2005)
26. Naumov A V, Vainer Yu G *Izv. Ross. Akad. Nauk, Ser. Fiz.* **70** 470 (2006) [*Bull. Russ. Acad. Sci.: Phys.* **70** 532 (2006)]
27. Bauer M, Kador L *J. Chem. Phys.* **118** 9069 (2003)
28. Barkai E et al. *Phys. Rev. Lett.* **91** 075502 (2003)
29. Barkai E et al. *J. Lumin.* **107** 21 (2004)
30. Vainer Yu G et al. *Opt. Spektrosk.* **98** 806 (2005) [*Opt. Spectrosc.* **98** 740 (2005)]
31. Vainer Yu G, Naumov A V *Opt. Spektrosk.* **98** 814 (2005) [*Opt. Spectrosc.* **98** 747 (2005)]
32. Geva E, Skinner J L *J. Phys. Chem. B* **101** 8920 (1997)
33. Barkai E, Silbey R, Zumofen G *Phys. Rev. Lett.* **84** 5339 (2000)
34. Vainer Yu G, Naumov A V, Bauer M, Kador L *J. Lumin.* **127** 213 (2007)
35. Bach H et al. *Opt. Mater.* **9** 376 (1999); Donley E A et al. *J. Phys. Chem. A* **103** 2282 (1999)
36. Vainer Yu G, Naumov A V, Bauer M, Kador L *Phys. Rev. Lett.* **97** 185501 (2006)
37. Naumov A V, Vainer Yu G *Izv. Ross. Akad. Nauk, Ser. Fiz.* **72** 748 (2008) [*Bull. Russ. Acad. Sci.: Phys.* **72** 708 (2008)]
38. Vainer Yu G, Naumov A V, Kador L *Phys. Rev. B* **77** 224202 (2008)
39. Naumov A V, Vainer Yu G, Kador L *Phys. Rev. Lett.* **98** 145501 (2007)

PACS numbers: **07.07.–a**, **87.64.–t**, 87.85.fk
DOI: 10.3367/UFNe.0179.200903o.0329

Optical biosensors of genotoxicants based on DNA nanoconstructions and portable dichrometers

O N Kompanets, Yu M Yevdokimov

1. Introduction

In this report, we briefly review results obtained in the last five years in the field of the development and practical use of biosensor methods and devices for the rapid determination of genotoxicants in liquids. Special attention is given to the nanobiotechnological approach developed in the Engelhardt Institute of Molecular Biology (IMB), Russian Academy of Sciences, with the instrumental support of the Institute of Spectroscopy, Russian Academy of Sciences (ISAN), which suggests the use, as integral biosensing units, of nanoconstructions that consist of double-stranded DNA molecules immobilized in optically transparent isotropic hydrogels having the property of abnormal optical activity, and of portable dichrometers as the recorders and transducers of circular dichroism.

Progressively increasing anthropogenic environmental pollution dictates the need for developing methods and devices for medical and ecological diagnostics, and also methods for controlling the quality of food products and medicinal preparations to ensure highly sensitive and rapid determination of the presence in them of toxicants that are hazardous to health and whose 'target' is the genetic material of cells. Such toxicants include some antibiotics and other