Temperature dependencies of negative ions formation by capture of low-energy electrons for some typical MALDI matrices

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Abstract

Some typical matrix assisted laser desorption/ionization (MALDI) matrices (gentisic, nicotinic, succinic, sinapinic, caffeic acids, urea and 6-aza-2-thiothymine) have been investigated by electron capture negative ions mass spectrometry (ECNI-MS) with purpose to find such decay channels of target molecule that may be important for processes of desorption and "primary" ions formation in MALDI. The temperature dependencies of the process of dissociative electron attachment to molecules of MALDI matrices were obtained for six different temperatures in the range of 70–300 °C. Also the relative dissociative attachment peak cross-sections for molecules under investigation were measured and ionization and desorption abilities of the MALDI matrices were calculated. The general conclusions are (1) a capture of low energy electrons by molecules of MALDI matrices leads to an appearance of anions CN^- and $[M - H]^-$ that may take part in formation of analyte anions, (2) gas molecules in MALDI (CO_2, H_2, CH_3CN and so on) are formed by a capture of low energy electrons that may leads to desorption of the analyte in according to CO_2-MALDI model, (3) the dissociative attachment cross-section for molecules of matrix determines the ability to form both anions and gas bubbles as well as indicates how well a matrix preserves positive analyte ions from neutralization by the free low energy electrons.
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1. Introduction

Matrix assisted laser desorption/ionization mass spectrometry (MALDI-MS) [1–4] is an excellent technique for analysis of large biological molecules like peptides, proteins, oligonucleotides and oligosaccharides. In spite of undoubted success of this method [5–7], there are many unanswered questions concerning the processes in MALDI plume as well as in the solid sample. A pathway of desorption and ion formation of analyte molecules in MALDI is not clearly understood up to now. In the recent work [8] it is reviewed a lot of parameters of MALDI experiment (laser characteristics, sample morphology, preparation protocols) which influences the desorption of MALDI sample including the excitation of samples, the subsequent phase change and the dynamics of plume expansion. Some of the desorption mechanisms [9] take into consideration a formation of gas bubbles assisting the ablation of the solid sample due to decomposition of matrix molecules under the heating [10]. But such an explanation of MALDI desorption doesn't work

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properly in the case of using thermally stable matrices because of there are no gas molecules formed in this case by thermal decomposition of the matrix.

To best of our knowledge ionization mechanisms including gas-phase photoionization, thermal ionization and excited state proton transfer encounter significant difficulties [11]. "Primary" ionization mechanisms, i.e., generation of the first ions from neutral molecules in the sample are difficult to explain especially in terms of photoionization by one photon–molecule interactions. "Secondary" ionization mechanisms result from various gas-phase reactions in MALDI plume with the participation of "primary" ions and lead to the final mass spectrum observed in MALDI. So, the problem is to find appropriate mechanism for the "primary" ions formation. Recently, it was proposed several others models for the "primary" ionization processes in MALDI including the cluster ionization mechanism [12,13] as well as models of the "secondary" ionization events in MALDI plume [14,15].

On the other hand an existence of free electrons in MALDI plume is a proven fact by now [16]. Moreover the energy distributions of these electrons are measured for different types of metal substrates [17,18]. In according to this work free electrons in MALDI plume appear just after laser pulse by photoemission from metal substrate used for solid MALDI sample. Then these electrons exists for a time in the solid sample as well. As it is shown below in such a way the free electrons may to condition both the desorption of MALDI sample and the "primary" negative ions formation.

The energies of free electrons in MALDI do not exceed 2 eV [18], i.e., processes of dissociative attachment of electrons may be important in MALDI plume and solid sample because of such a reaction takes place at low electron energies, below 20 eV and, as a rule, the cross-section for dissociative attachment increases strongly as energy of incident electron tends to zero [19]. So, it is very useful for understanding MALDI processes to investigate some typical MALDI matrices by electron capture negative ions mass spectrometry (ECNI-MS), which gives a detailed description of the processes of transient parent negative ion formation and its decay followed by low-energy electron–molecule interaction. In addition, investigation of temperature dependencies of dissociative electron attachment by MALDI matrices represents another one interesting thing because of the temperature of molecules in MALDI sample is varied from ambient temperature (before applying the laser pulse) to 500–700 K (after applying the laser pulse) in MALDI plume [20,21]. Thus the matrix molecules which may capture the free electrons in MALDI are rather "hot" while the cross-section for process of electron capture depends strongly on the temperature of target molecule. Such an investigation is also a subject of this work.

It should be clearly noted that this work consider only reaction of the dissociative low-energy electron attachment to molecules of typical MALDI matrices and the possible role of such a process in conventional MALDI experiments. The negative ions of matrix molecules can be formed in the gas phase as well as in the solid state [22,23] by attachment of low-energy electrons. Anions formed then undergo rapid unimolecular decomposition which gives fragments anions and radicals (hydrogen atoms, for example) which can be very reactive in MALDI plume. The cross-section for dissociative electron attachment can be 4 to 5 orders of magnitude larger then typical photoionization and excitation cross-sections especially if a slow electron collides with an excited molecule. In respect to the process of dissociative electron capture it is unimportant where is the origin of free electrons in MALDI as it was discussed elsewhere [17], it is more than enough that energies of these electrons are low.

2. Experimental/materials and methods

Electron capture reactions by single molecules of MALDI matrices were studied in the gas-phase by passing an electron beam through ionization chamber of the mass spectrometer where an equilibrium flow of molecules under investigation were created. The negative ions formed were recorded mass spectrometrically

as a function of incident electron energy. Such a dependence, namely yield of defined negative ion versus electron energy, so-called curve of effective yield, was recorded for each peak in the mass spectrum which is determined by various fragment negative ions corresponding to different decay channels of parent molecular anion. If the lifetime of transient parent molecular anion is as long as 10^{-6} s and more, so-called long-lived molecular anion [19], then it is observable mass spectrometrically and it is seen on the mass spectrum as well as the fragment anions. The general concepts of the electron attachment spectroscopy and, specifically ECNI-MS, are described in details elsewhere [23,24].
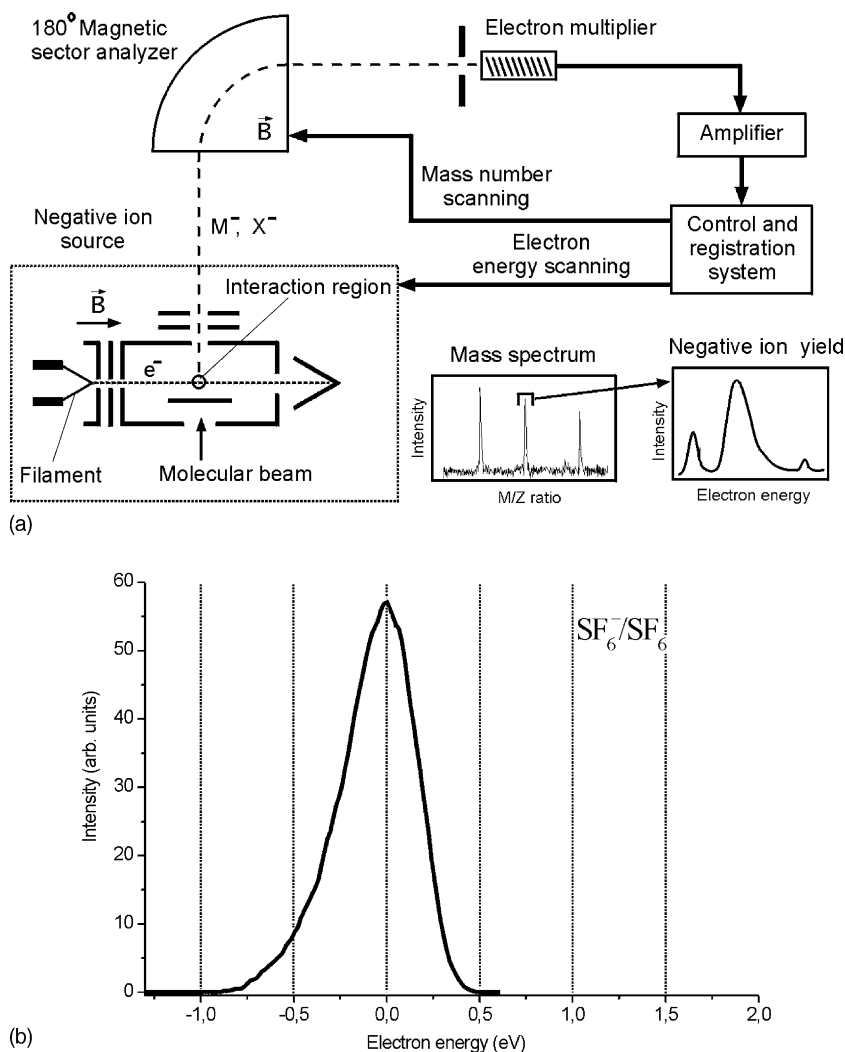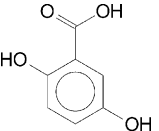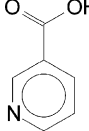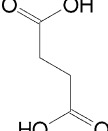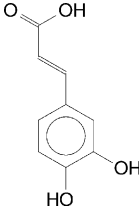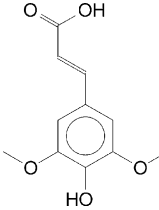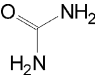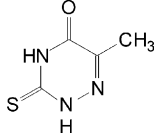

Fig. 1. (a) Schematic representation of the negative ion mass spectrometer. Drift time for the SF_6^- ion (m/z 146) from the moment of formation in the ion source, through the mass separator system to the moment of detection by the secondary electron multiplier, is about 25 μs. (b) Yield of SF_6^- ion from molecule SF_6 vs. the electron energy measured at minimal accessible temperature (75 °C); such a dependence represent the electron energy distribution in the ion source of the mass spectrometer or in other word the energy resolution in our experiments.

Table 1
Molecular structure, weight, melting point[a] and temperature of the rod used for evaporation of MALDI matrices under investigation into reaction chamber of the mass spectrometer

	Compound						
	Gentisic acid	Nicotinic acid	Succinic acid	Caffeic acid	Sinapinic acid	Urea	ATT
Molecular structure							
Molecular weight	154	123	118	180	224	60	143
Melting point (°C)	205 (decomposition)	236–239	187–189	194–198 (decomposition)	203–205 (decomposition)	133–135	218–221
T_{rod} (°C)	105	80	100	145	135	100	60

[a] From Catalog Handbook of Fine Chemicals by Aldrich Chemical Company Inc.

Processes of dissociative electron attachment by molecules of MALDI matrices were investigated by means of ECNI-MS using the mass spectrometer MI-1201 under the following conditions: FWHM of electron energy distribution 0.4 eV, electron trap current 1 μA, electron energy range 0–12 eV, accelerating voltage 4 kV. The pressure in the reaction chamber was kept as low as 10^{-5} Torr in order to ensure single-collision conditions. Briefly the mass spectrometer used consists of the anion source, magnetic analyzer and electron multiplier. Deploying of electron energy and mass number were carried out by IBM-comparable computer using CAMAC interface. Schematic representation of the mass spectrometer is shown in Fig. 1a. The electron energy scale was calibrated by the yield of SF_6^- anion from molecule of SF_6 which gives the well-known resonance peak at 0 eV. The curve of effective yield for that resonance may be fitted by δ-function, i.e., it has a small value of FWHM [19]. Thus experimentally measured yield of SF_6^- ion represents the mirroring of electron energy distribution in the ion source. In Fig. 1b the curve of effective yield for SF_6^- anion is shown. In that way the "negative" electron energies correspond to the tail of Maxwell's distribution of electrons emitted by the hot tungsten filament. As it seen in Fig. 2b the FWHM of this resonance peak is about 0.4 eV that is the electron energy resolution in our experiments.

MALDI matrices under investigation are listed in Table 1. All of the substances investigated, namely organic acids gentisic, nicotinic, succinic, sinapinic, caffeic as well as urea and 6-aza-2-thiothymine (ATT), are solid under standard conditions. They were introduced into ionization chamber of the mass spectrometer using a rod heated in the temperature range 80–130 °C with purpose of evaporation of the sub-

stances under investigation. The rod was made from stainless-steel and was introduced in the ion source of the mass spectrometer through the sluice chamber. The temperature of the rod was chosen so to obtain satisfactory intensity of the main dissociative channel for each substance. The ion source of the spectrometer was equipped with an oven for heating of molecules under investigation up to 300 °C with purpose to obtain a temperature dependence of electron capture processes. The molecules under investigation reach the temperature required by collisions with the walls of reaction chamber of the mass spectrometer. The temperatures of the rod used in this study for evaporation of the molecules into reaction area of the mass spectrometer and melting points of the matrices taken from Catalog Handbook of Fine Chemicals by Aldrich Chemical Company Inc. are shown in Table 1. All of the substances under investigation were purchased from Aldrich (Buchs, Switzerland) and were used without further treatment and purification.

3. Results

The negative ion electron capture mass spectra for MALDI matrices under investigation at the ambient temperature were reported in details previously [25]. The relative dissociative attachment peak cross-sections for molecules of MALDI matrices are shown in Table 2. A value of the dissociative attachment peak cross-section corresponds to the maximum value of the total current of negative ions plotted versus the energy of incident electrons, i.e., they characterize how effectively the process of electron capture occurs. A method for estimating the cross-section was described elsewhere [26]. Briefly determination

Table 2
The relative dissociative attachment peak cross-section and existence of long-lived molecular anion for MALDI matrices under investigation

	Compound						
	Gentisic acid	Nicotinic acid	Succinic acid	Caffeic acid	Sinapinic acid	Urea	ATT
σ_{DA} ($\times 10^{-16}$ cm^2)	0.135	0.214	0.118	0.096	0.256	0.0012	0.083
Existence of M$^-$	Yes	?	No	Yes	Yes	No	Yes

of the relative dissociative attachment cross-section was carried out by comparison of the negative ion currents for substance investigated and SF_6 which has a well-defined cross-section for the negative ion formation [19]. The relative vapor pressure of the different molecules in the ion source was determined by mean of switching the mass spectrometer in positive ion mode.

One can see from Table 2 that sinapinic acid has the largest relative dissociative attachment peak cross-section. So, if we take this value as 100% then matrices under investigation may be arranged in according to their abilities to capture free electrons as sinapinic acid (100%), nicotinic acid (84%), gentisic acid (53%), succinic acid (46%), caffeic acid (38%), ATT (32%), urea (0.5%). These values may characterize at least ability of any matrix to protect positive ions in MALDI plume from recombination with free electrons.

Whether the long-lived molecular anion exists for molecules under investigation is also shown in Table 2. The existence of M^- means that corresponding molecule have a positive electron affinity. In the case of nicotinic acid the curve of effective yield for molecular anion is hidden by much more intense isotopic one for $[M-H]^-$ decay channel of the target molecule. This is why one can't say with confidence that there is long-lived M^- for nicotinic acid.

The temperature range accessible in our experiment is 75–300 °C. The temperature dependencies of the mass spectra have been measured for six different points within this temperature range but here we present results only for minimal (about 75 °C) and maximal (about 300 °C) temperatures which were slightly (± 5 °C) different for each matrix investigated that is a feature of our experimental setup. The negative ions mass spectra of MALDI matrices under investigation are shown in Fig. 2 for the minimal (left-hand side) and maximal (right-hand side) temperatures of the molecules. Corresponding temperatures are also shown on the figure. Intensity in Fig. 2 represents the normalized on maximal value integral intensity of corresponding decay channels within the accessible electron energy range 0–12 eV, i.e., the

value of

$$I_{max}^{-1} \int_0^{\varepsilon_{max}} I(\varepsilon)\, d\varepsilon$$

where $\varepsilon_{max} = 12$ eV is the upper limit of our experimental electron energy range, $I = I(\varepsilon)$ is the yield of corresponding negative ion versus the electron energy (see Fig. 3), I_{max} is the maximal value of the negative ion yield. In Fig. 3 curves of effective yield corresponded to some of the most intense channels in the mass spectra of matrices investigated are shown for the minimal and maximal temperatures of the molecules. On the other words Fig. 3 presents dependence of relative cross-section for formation of various fragment anions versus the energy of incident electron. Such a dependence has resonance character, i.e., it looks like a set of more or less narrow peaks. The complete set of the negative ion electron capture mass spectra of MALDI matrices investigated was published in preceding paper [25].

In general for all molecules investigated there are $[M-H]^-$ anion peak in mass spectrum which absolute intensity decreases as the temperature of the molecule rising but this decay channel doesn't disappear even in spectrum corresponding to the maximal temperature of molecule. Also there is CN^- channel for nitrogen-containing matrices and absolute yield of this channel increases strongly (up to 1 order of magnitude in the case of urea, for example) as the temperature of the molecule rising especially at low electron energies. We propose that such fragment anions namely $[M-H]^-$ and CN^- if they appear in MALDI plume as a result of dissociative attachment of free electrons may lead to formation of analyte anions by charge transfer process. In according to the general rules of the negative ions formation [24] we propose that structure of $CHCOOH^-$ fragment anion in the spectra of succinic acid corresponds to the break of parent anion in half with transition of hydrogen atom on neutral fragment which is CH_3COOH in this case.

Decay channel $[M-46]^-$ in the spectra of organic acids has been interpret by loss of CO_2 and H_2 molecules rather then molecule of formic acid because of the full energy of carbon dioxide and hydrogen

Fig. 2. The electron capture negative ion mass spectra of MALDI matrices under investigation (gentisic acid, nicotinic acid, succinic acid, sinapinic acid, caffeic acid, urea and ATT) at two different temperatures of the target molecule: minimal accessible temperature (near 75 °C, left-hand side) and maximal accessible temperature (near 300 °C, right-hand side); corresponding temperatures are also shown on the figure; intensities here are normalized on maximal values for corresponding decay channels.
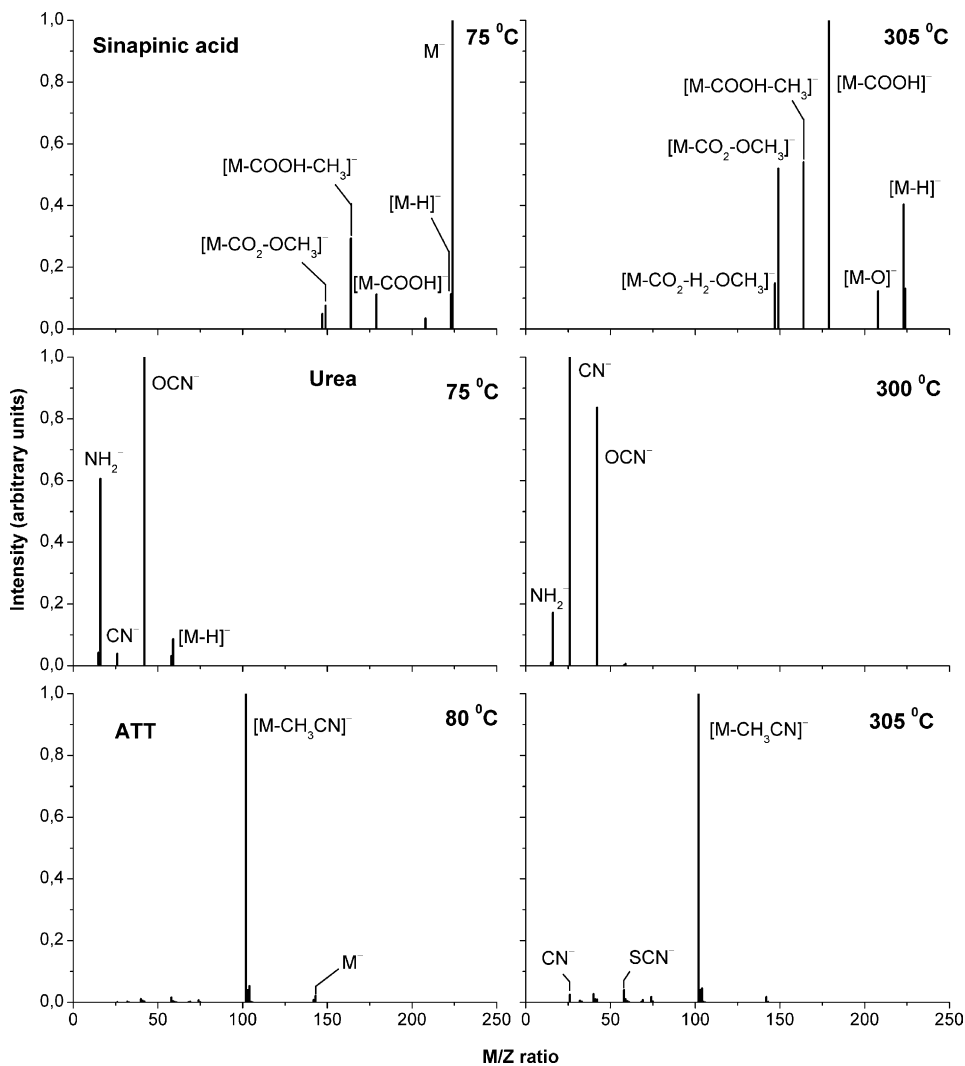
Fig. 2. (*Continued*).

molecules in total are on approximately 0.4 eV smaller then that for HCOOH molecule as it was shown by the ab initio quantum chemical calculations in medium basis set (6-31G*) using HyperChem[TM] 7.01 molecular modeling system by Hypercube Inc. But unequivocal interpretation of that decay channel in negative ion mass spectra of molecules under investigation is a subject of individual analysis. For example the loss of fragment with m/z 46 in the case of gentisic acid, i.e., the decay reaction $M^- \rightarrow [M - 46]^-$, is a metastable

slow process. It takes parent negative ion of gentisic acid about $10\,\mu s$ (time-of-flight to the magnetic sector analyzer) to loss these fragment. In such a way we can see theses metastable ions (which are formed in magnet analyzer) on the mass spectra with fractional values of mass number. But metastable decay $M^- \rightarrow [M - 46]^-$ has been observed in the case of gentisic acid only (results are not presented here).

For all MALDI matrices which are organic acids there is a decay channel $[M - COOH]^-$ whose
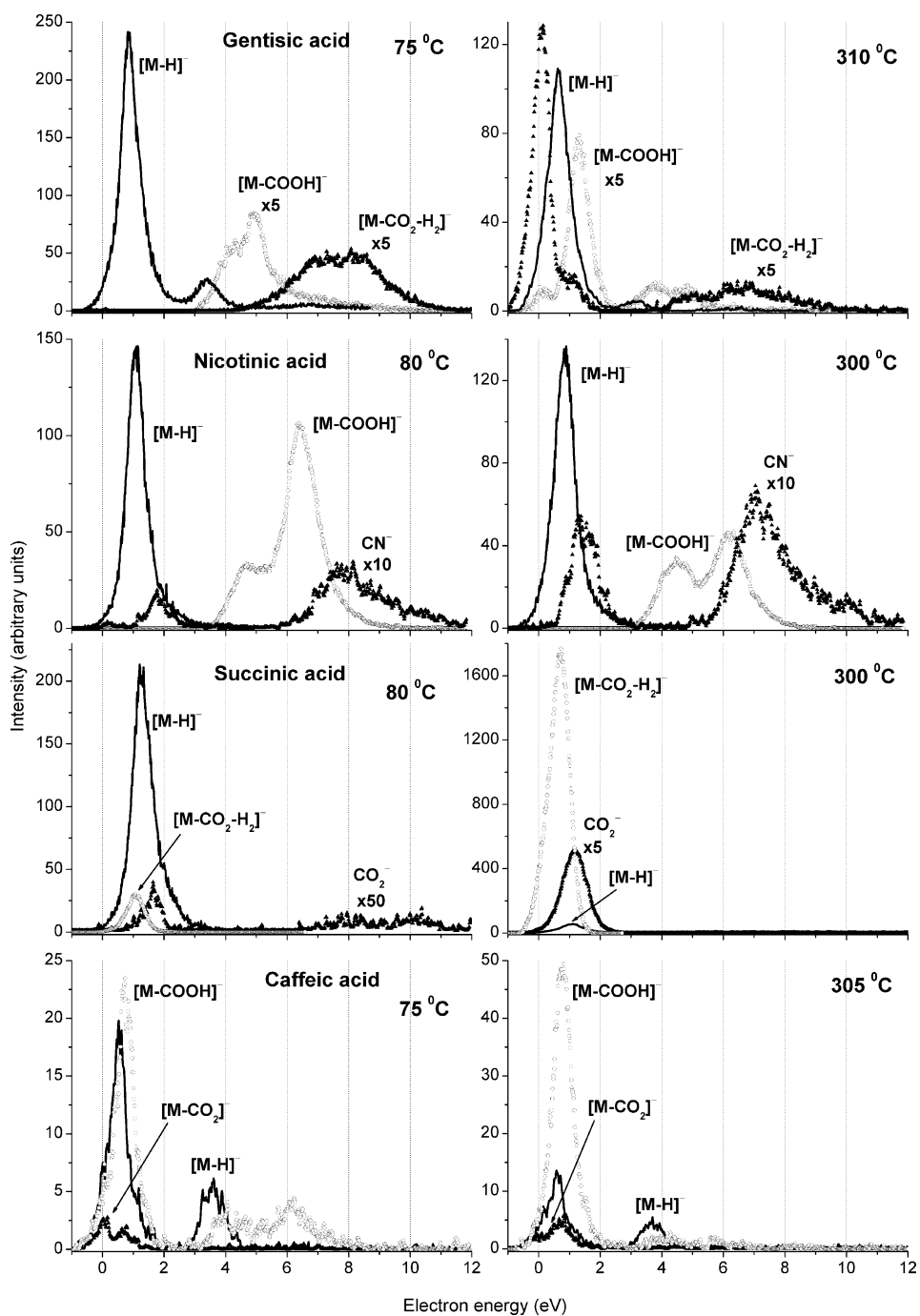
Fig. 3. The curves of effective yield for the most intense decay channels of MALDI matrices under investigation (gentisic acid, nicotinic acid, succinic acid, sinapinic acid, caffeic acid, urea and ATT) at two different temperatures of the target molecule: minimal accessible temperature (near 75 °C, left-hand side) and maximal accessible temperature (near 300 °C, right-hand side); corresponding temperatures are also shown on the figure.
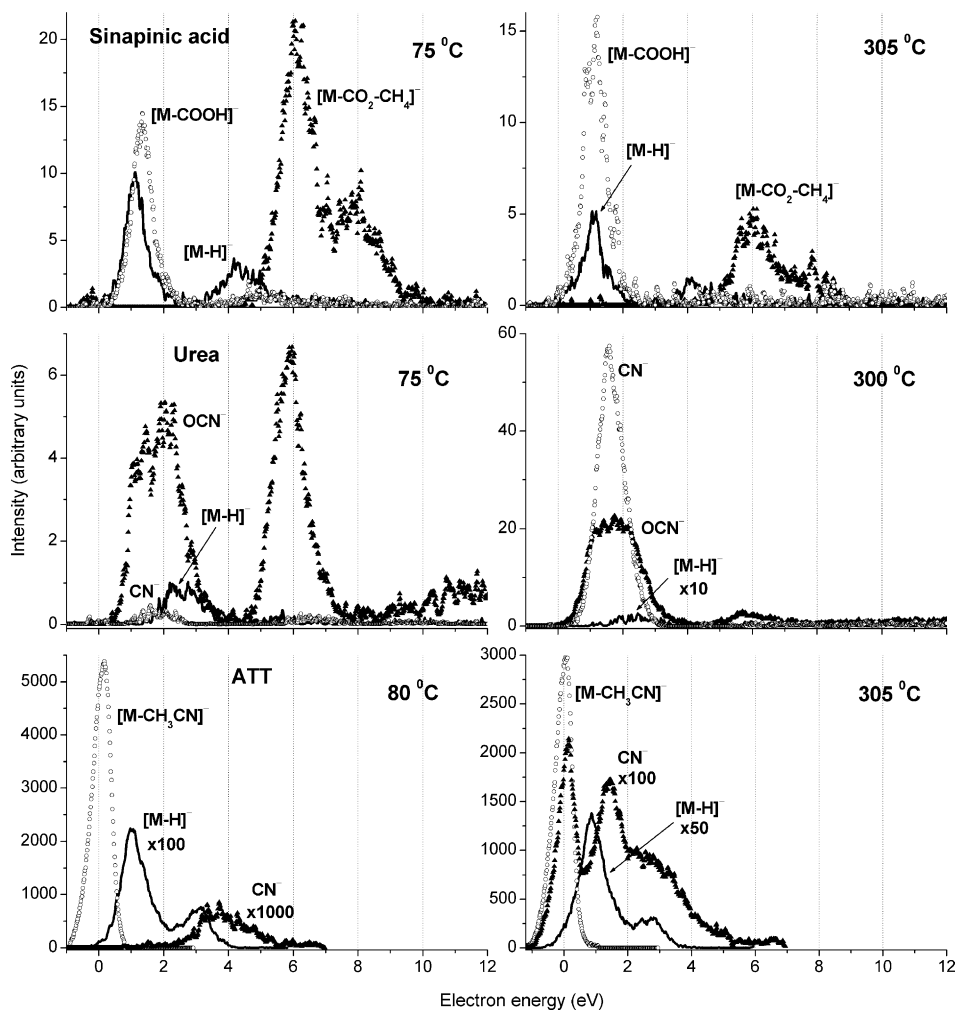
Fig. 3. (*Continued*).

absolute intensity increases dramatically (up to 2 orders of magnitude in the case of gentisic acid, for example) as the temperature of target molecule increases. Such a temperature behavior is also characteristic for $[M - CO_2]^-$ and $[M - CO_2 - H_2]^-$ decay channels in the mass spectra of organic acids under investigation. We emphasize such a fragment negative ions because of this is a source of gas molecules (hydrogen, carbon dioxide) which may assist the formation of gas bubbles and following desorption of MALDI sample in according to CO_2-MALDI model [10]. In the case of ATT the corresponding decay channel is

$[M - CH_3CN]^-$ and this channel is a much more intense then others both in at the minimal and maximal temperatures in the negative ion spectrum of ATT.

As it is mentioned above the electron capture is a resonant process, i.e., it occurs if the energy of incoming electron fits that of the temporary negative ion. That is why the yield of negative ion plotted versus the energy of incident electron looks like a set of more or less narrow peaks (see Fig. 3). In according to work [18] in MALDI we have electrons with energies about 1 eV. But some of the curves in Fig. 3 have resonant states at relatively high electron energies. Thus

any decay channel in the negative ion mass spectra in Fig. 2 is important for MALDI if corresponding curve of effective yield in Fig. 3 covers well the energy distribution of free electrons in MALDI which has a maximum at approximately 1 eV (that depends on the metal substrate) and FWHM about 0.5 eV as it was shown in the work [18].

As it is seen in Fig. 3 for $[M - COOH]^-$, $[M - CO_2 - H_2]^-$ and CN^- decay channels for organic acids and nitrogen-containing matrices, respectively, the low-energy (about 1 eV) resonances appear as the temperature of target molecule rises. For all molecules under investigation the intensity of the $[M - H]^-$ fragment anion decreases with the temperature. The maximum of this decay channel lies at approximately 1 eV for all matrices except urea, thus such a decay channel may influence MALDI processes.

4. Discussion

As it was shown elsewhere [18] free electrons in MALDI appear after the laser pulse has reached the surface of metal substrate by photoemission. As a rule the matrix absorbs the laser irradiation well. So, we believe that molecules which may capture free electrons (in a solid sample immediately after beginning of the photoemission process or a plume after its formation) are rather "hot". Indeed the temperature of MALDI sample is estimated to approximately 500–900 K for UV MALDI [20,21]. We have the negative ions spectra and anion yields for minimal and maximal temperatures of molecules under investigation. Thus we can see a tendency of the dissociative attachment with increasing of the temperature.

In according to Table 1 molecules of gentisic acid, caffeic acid and sinapinic acid undergo a thermal decomposition in solid state at the temperature of about 200 °C. In our case of gas-phase heating some of these molecules remain intact even at the 300 °C that is confirmed by the existence of $[M - H]^-$ anion in the spectra of these compounds at maximal temperature. However, some of these molecules undergo the thermal decomposition in the ion source of

the mass spectrometer with escape of CO_2 molecule. That process may be controlled by specific form and intensity of yield of O^- anion from molecule of CO_2 which gives a well-known resonance state at approximately 4 eV. Indeed at the maximal temperatures (about 300 °C) the yield for O^- anion from molecule of CO_2 have been measured in the spectra of gentisic acid (results are not presented here) but absolute value of the ion yield was too small. For reliable interpretation it is necessary to estimate the cross-section for such a process that is subject of the next publication.

Thus a decreasing (not a disappearing for all molecules under investigation) of $[M - H]^-$ anion yield for unstable matrices as the temperature increases is partly attributed to the thermal decomposition. However, for other molecules investigated such a decreasing is a temperature effect. According to Fig. 3, a decreasing of $[M - H]^-$ ion with the temperature for nitrogen-containing matrices is followed by increasing of CN^- ion. We don't consider a molecular anion M^- that exist for some molecules (see Table 2) as a possible agent for the process of charge transfer in MALDI plume because there is a rapid drop of the M^- yield at high temperature that is a general effect of autodetachment [19]. To apply our results to the possible explanation of MALDI processes we have to take into account incident electrons with energy below 2.5 eV because this is a high-energy tail for the electron energy distribution in MALDI as it was measured by Zenobi and co-workers [18]. Moreover, there are no reasons to believe that free electrons with much more higher energies exists in MALDI.

To make an estimate according to the real energy distribution of free electrons in MALDI we may fit the energy distribution by Gauss curve with maximum at 1 eV and FWHM 0.5 eV, i.e., by the expression

$$G(\varepsilon) = e^{-(\varepsilon - \varepsilon_0)^2 / 2w^2}$$

where ε_0 and w are maximum and FWHM of the Gauss distribution, respectively. Further let us introduce an *ionization ability* of any matrix in MALDI (stipulated by the capture of free electrons in MALDI

Table 3
The ionization ability of MALDI matrices under investigation by two different ionizing agents at minimal and maximal accessible temperatures

		Compound						
		Gentisic acid	Nicotinic acid	Succinic acid	Caffeic acid	Sinapinic acid	Urea	ATT
Ionization ability (%)								
By $[M - H]^-$	$\sim$75 °C	100	100	62.6	2.7	7.9	7.3×10^{-5}	6.5
	$\sim$300 °C	38.7	100	24.5	2.3	4.2	10^{-4}	8.3
By CN^-	$\sim$75 °C		100				0.17	1.5
	$\sim$300 °C		64.7				2.4	100

sample after laser shot) as

$$\sigma_{DA} \int_0^{\varepsilon_{max}} I(\varepsilon)G(\varepsilon)\,d\varepsilon$$

where σ_{DA} is the relative dissociative attachment cross-section for matrix, $\varepsilon_{max} = 12\,eV$ is the upper limit of our experimental electron energy range, $I = I(\varepsilon)$ is the yield of the possible ionizing agent in MALDI versus electron energy ($[M - H]^-$ or CN^- as we propose), i.e., corresponding curve of effective yield. Under such definition the value of ionization ability takes into account efficiency of electron capture by matrix, resonant nature of electron capture process and the energy distribution of free electrons in MALDI as well. Values of the ionization ability normalized on maximal value for matrices under investigation and two possible ionizing agent, $[M - H]^-$ and CN^-, calculated by this method are shown in Table 3.

As a temperature of the target molecule increases there is a strong (up to two orders of magnitude in the case of gentisic acid) increasing of anion yield for $[M - CO_2 - H_2]^-$ and $[M - COOH]^-$ decay channels for molecules of organic acids as it is shown in Fig. 3. In the case of matrices without of COOH-group there

are another decay channels like $[M - gas\,molecule]^-$ which may assist the formation of gas bubbles in MALDI. For example the main dissociative channel for ATT is $[M - CH_3CN]^-$ in the spectra at minimal and maximal temperatures as well.

Further let us introduce a *desorption ability* of any matrix in MALDI (stipulated by capture of free electrons in MALDI sample after laser shot) by the same formulae but $I = I(\varepsilon)$ is the yield of the most intense decay channel like $[M - gas\,molecule]^-$ which can assists the gas bubbles formation in MALDI. Values of the desorption ability normalized on maximal value for matrices under investigation calculated by this method are shown in Table 4. Corresponding decay channels are $[M - CO_2 - H_2]^-$ for gentisic and succinic acids, $[M - COOH]^-$ for nicotinic, caffeic and sinapinic acids, $[M - H_2 - NH_2]^-$ for urea and $[M - CH_3CN]^-$ for ATT.

In this work we consider the processes of free electron capture in MALDI as possible pathways for ionization (by charge transfer processes) and desorption (in according to CO_2-MALDI model) events in conventional MALDI experiments. In such a way matrices under investigation may be arranged in

Table 4
The desorption ability of MALDI matrices under investigation at minimal and maximal accessible temperatures

	Compound						
	Gentisic acid	Nicotinic acid	Succinic acid	Caffeic acid	Sinapinic acid	Urea	ATT
Decomposition ability (%)							
$\sim$75 °C	0.03	0	38.4	18	28	0.05	100
$\sim$300 °C	0.32	0	100	2.7	2.4	0.01	2.2

according to values of the ionization abilities as follows (see Table 3): nicotinic acid (100%), gentisic acid (38.7%), succinic acid (24.5%), ATT (8.3%), sinapinic acid (4.2%), caffeic acid (2.3%), urea (10^{-4}%) and in according to the values of desorption abilities as follows (see Table 4): succinic acid (100%), caffeic acid (2.7%), sinapinic acid (2.4%), ATT (2.2%), gentisic acid (0.32%), urea (0.01%), nicotinic acid (0%). In spite of the fact that $[M - COOH]^-$ channel is presented on the spectra of nicotinic acid and it has sufficient intensity the desorption ability for such a matrix is zero because of corresponding curve of effective yield has a maximum at 6.5 eV (see Fig. 3), but in MALDI there are no electrons with such energies. Such a fact is a striking example of the resonant nature of the dissociative electron attachment process.

Let us consider only possible role of the electron capture process in MALDI. One can see that in such a way the best matrices are succinic and gentisic acids. In addition the desorption ability of gentisic acid may be increased significantly by using gold substrate for MALDI sample because in that case maximum of the energy distribution of free electrons is about 0.6 eV [18] and the distribution covers curve of effective yield of $[M - CO_2 - H_2]^-$ decay channels in the spectra of gentisic acid much more better (see Fig. 3). There are approximately equal values of efficiency for ATT, caffeic and sinapinic acids. Urea is a poor matrix in according to our experimental results.

5. Conclusions

The negative ions electron capture mass spectra and anion yield for seven typical MALDI matrices have been measured by means of ECNI-MS. The temperature dependence of dissociative low energy electron attachment by these molecules was also obtained. The general conclusions are as follows.

1. Capture of low energy electrons by molecules of MALDI matrices leads to appearance of anions CN^- and $[M - H]^-$ that may take part in charge transfer processes and formation of analyte anions in conventional MALDI experiments. In such a way the intensity of corresponding decay channel in the negative ions mass spectrum is an indication of the ionization ability of matrix caused by the electron capture only.

2. Gas molecules in MALDI sample (CO_2, H_2, CH_3CN and so on) are formed by a capture of low energy electrons as well as by thermal decomposition of matrix molecules. The latter process takes place only for unstable molecules. Thus, if that process is effective in MALDI then it leads to desorption in according to CO_2-MALDI model. In such a way the intensity of corresponding decay channel in the negative ions mass spectrum is an indication of the desorption ability of matrix caused by the electron capture only.

3. Taking into account only process of dissociative electron attachment in MALDI we propose that succinic and gentisic acids are the more effective in MALDI, urea and nicotinic acid are the least effective and others matrices investigated are at an intermediate position. However, these estimates must be very sensitive to conditions of individual MALDI experiment, for example, temperature of a plume, substrate material, thickness of solid sample and so on.

Acknowledgements

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