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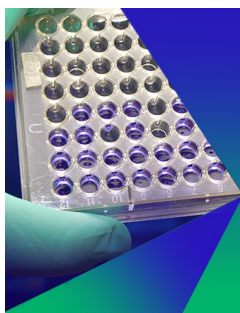
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Dissociative electron attachment to the radiosensitizing chemotherapeutic agent hydroxyurea

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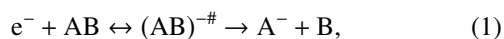
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Dissociative electron attachment to hydroxyurea was studied in the gas phase for electron energies ranging from zero to 9 eV in order to probe its radiosensitizing capabilities. The experiments were carried out using a hemispherical electron monochromator coupled with a quadrupole mass spectrometer. Diversified fragmentation of hydroxyurea was observed upon low energy electron attachment and here we highlight the major dissociation channels. Moreover, thermodynamic thresholds for various fragmentation reactions are reported to support the discussion of the experimental findings. The dominant dissociation channel, which was observed over a broad range of energies, is associated with formation of NCO^- , water, and the amidogen (NH_2) radical. The second and third most dominant dissociation channels are associated with formation of NCNH^- and NHCONH_2^- , respectively, which are both directly related to formation of the highly reactive hydroxyl radical. Other ions observed with significant abundance in the mass spectra were NH_2^-/O^- , OH^- , CN^- , HNOH^- , NCONH_2^- , and ONHCONH_2^- . Published by AIP Publishing. [<http://dx.doi.org/10.1063/1.4953579>]

I. INTRODUCTION

Understanding the mechanisms behind the interactions of incident particles and the main target for the radiation therapy—DNA molecules—plays a key role in its controlled sensitizing. Therefore, it is of great importance, in addition to investigating damage induced to DNA molecules, to explore the mechanisms by which various types of radiation, e.g., ions, electrons, or photons may interact with the environment of the cellular target as well as to understand the possible pathways through which such damage is mediated. In the case, when sensitizing molecules are introduced to the cell, their interactions with the environment need to be accounted for and resulting DNA damage assessed, accounting also for secondary species produced upon sensitizer interaction with incident radiation. Proper quantification and control of such processes may help to reduce radiation dose and thus lower the harmful effects on the healthy tissue.¹ One of the possible mechanisms is dissociation of the radiosensitizing molecules due to resonant attachment of secondary low energy electrons (LEEs) and formation of reactive species such as radicals or ions. Dissociative electron attachment (DEA) was shown to be the dominant process for fragmentation of molecules of biological interest in this context.² This mechanism follows a general scheme,



according to which a molecule AB, after capturing a low energy electron (LEE), forms a transient negative ion (TNI)

$(\text{AB})^{-\#}$ that can further dissociate into an anion A^- and a neutral fragment B.

In the present investigation, we have focused on a simple chemotherapeutic drug, hydroxyurea³ (HU, with condensed structural formula OH-NH-CO-NH_2). Its inhibitory effect on DNA metabolism was discovered in the mid-sixties of the past century^{4,5} and it was promptly tested as tumor treating therapeutic agent.^{6,7} Although it was proven that administration of HU in conjunction with radiation therapy results in a lower overall survival rate than the combination of radiation therapy with the drugs 5-fluorouracil or cisplatin for some types of cancer,⁸ concomitant HU treatment and radiation therapy yet outmatch the exclusive radiotherapy.⁹ Nowadays, HU is widely used in acute leukemia treatment for older patients that are not suitable for induction chemotherapy.¹⁰ Other studies revealed that a long term use of HU reduced the mortality¹¹ and improved the quality of life by reducing the abundance of pain episodes^{12,13} of patients treated for sickle cell anemia. For some cases of cervix and uterus cancer,^{14,15} it was found that HU was efficient in treating cancer bearing no significant effect on cellular mechanisms. It was also suggested that HU may be used as a radiosensitizer upon administration to patients suffering from cervix and head or neck cancer.¹⁶ Due to its ability to prevent proviral DNA synthesis,^{17,18} HU may be considered also for HIV replication blocking and hence, possible AIDS treatment.¹⁹

HU, which decomposes very rapidly in aqueous acidic medium, is readily absorbed after oral administration and enters cells via passive diffusion. The highest level of the drug in the blood can be noted within 2–4 h after administration and more than half of its dose is excreted with the urine

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within 8 h.¹⁹ In addition, HU inhibits ribonucleotide reductase and induces a block at the G1-S phase of the cell cycle when cells are particularly sensitive to radiation. Cell death is preferential in the S phase, which is a relatively radio-resistant phase of the cell cycle.²⁰ Cell culture studies on HeLa cells revealed that HU prevents repair of sublethal radiation damage.²¹

Nevertheless, the precise mode of action of HU is still unknown. It is suspected that the biological role of this compound is related to the generation of hydroxylated nitrogen atoms²² and the mode of action can be attributed to a NO production mechanism which plays a key role in many physiological and pathophysiological functions of human bodies.^{4,7,8,20} HU has also been evaluated for its sensitizing potency upon radiotherapy via measurements of its one-electron reduction potential yielding -0.552 V.²³ The authors of this study concluded that the chemotherapeutic effect will be predominant, with very little influence arising from the radiosensitizing properties of this molecule.

In this work, we investigate DEA to HU in order to identify the most probable dissociation channels and hence, to explore fragmentation mechanisms possibly underlying the effectiveness of HU in combined chemo-radiation therapy. To our knowledge there exist no studies involving DEA to this drug yet.

II. EXPERIMENTAL

The electron attachment spectrometer used in the present study comprises a molecular beam oven, a high resolution hemispherical electron monochromator (HEM) and a quadrupole mass filter with a pulse counting system for analyzing and detecting the ionic products. The apparatus has been described previously in detail.²⁴ Briefly, as the HU sample is in solid state at room temperature and does not vaporize sufficiently, the sample was heated to about 362 K in a resistively heated oven in order to produce a molecular beam. The evaporated HU molecules were then introduced through a copper capillary with 1 mm diameter to the interaction chamber of the HEM where they interacted with an electron beam, possessing a well-defined electron energy. The anions generated by the electron attachment process were extracted by a weak electrostatic field into the quadrupole mass filter, where they were mass-analyzed and then detected by a channeltron-type secondary electron multiplier. The electron current was measured with a Faraday plate and monitored during the experiments using a picoammeter.

With the HEM, it is possible to achieve energy distributions with full width at half-maximum (FWHM) of 35 meV. To determine the energy spread and to calibrate the energy scale, the s-wave attachment to CCl_4 which leads to formation of Cl^- was used. In the present experiments the FWHM was about 100 meV with an electron current of about 5 nA. This energy resolution used represents a reasonable compromise between the product ion intensity and the energy spread to resolve resonances in the ion yields. The HEM was constantly heated to the temperature of 350 K in order to prevent surface charging. The pressure in the main vacuum

chamber of the mass spectrometer was about 10^{-6} mbar to ensure collision-free conditions.

The sample of the hydroxyurea with a purity of 98% was purchased from Sigma Aldrich, Vienna, Austria.

III. QUANTUM CHEMICAL CALCULATIONS

In order to complement and support the analysis and interpretation of experimental data on DEA to HU, the G4(MP2) extrapolation scheme²⁵ was used for the determination of thermochemical thresholds for various fragmentation reactions induced upon electron attachment. G4(MP2) is the most recent of the Gx extrapolation schemes developed by Curtiss and coworkers.²⁵ The method yields an average deviation of about 0.05 eV from experiment for the 454 energies compiled in the G3/05 test set.²⁵

In particular, the ground state (free) energies of reactants, $E(\text{R})$, and products, $E(\text{P})$, were calculated and subsequently, the thermochemical reaction threshold, $E(\text{P} \rightarrow \text{R})$, was calculated according to $E(\text{P} \rightarrow \text{R}) = E(\text{P}) - E(\text{R})$. Note that these thermochemical reaction thresholds can only serve as lower bounds (within an estimated accuracy of 0.1–0.15 eV) for experimentally determined appearance energies. Thermochemical reaction thresholds for a variety of eventual fragments formed upon DEA to hydroxyurea are summarized in Table I. Apart from zero and room temperature (298.15 K), we have also considered the used heating temperature of 362 K.

In addition, the dipole moment of hydroxyurea was computed at the CCSD/aug-cc-pVTZ^{26–29} level of theory based on the optimized geometry obtained at the B3LYP/6-31G(2df,p)^{30–32} level of theory, i.e., the level of theory used for optimization within the G4(MP2) scheme. All calculations were performed with the Gaussian 09 suite of programs.³³

IV. RESULTS AND DISCUSSION

A. Negative ion mass spectra

Dissociative electron attachment to HU resulted in formation of various anionic fragments, which are visible in the mass spectra shown in Fig. 1. The spectra were taken at incident electron energies of $\sim 0, 1, 3, 5, 7,$ and 9 eV and the most prominent assigned fragment anions are indicated in Fig. 1. The calculated thermodynamic reaction thresholds for these anions are included in Table I.

The HU anion was not observed at any of the incident electron energies since the lifetime of the transient negative anion towards autodetachment and dissociation was probably not sufficient to detect it on mass spectrometric timescales. We note that the calculated value of the dipole moment of HU is 3.62 D. For molecules exhibiting such a high dipole moment, a dipole-bound anion may form. Fragmentation may then be activated through this dipole-bound state which acts as a doorway state.³⁴ We observed the dehydrogenated molecular anion as the heaviest ion with m/z 75. This anion was detected in the mass spectra taken at 3 and 9 eV. No evidence of the parent ion and only the formation of its dehydrogenated

TABLE I. Reaction thresholds obtained using the G4(MP2) method at the three considered temperatures $T = 0, 298.15,$ and 362 K for various eventual fragmentation channels upon DEA to hydroxyurea. Negative values of reaction thresholds indicate exothermic reactions (see text for further explanation and discussion).

m/z	$e^- + \text{HU} \rightarrow$	Reaction thresholds (eV)		
		T = 0 K	T = 298.15 K	T = 362 K
75	$\text{ONHCONH}_2^- + \text{H}$	2.09	1.77	1.68
75	$\text{OHNCONH}_2^- + \text{H}$	1.72	1.39	1.31
75	$\text{OHNHCONH}^- + \text{H}$	1.59	1.29	1.21
60	$\text{OHNHCO}^- + \text{NH}_2$	2.67	2.18	2.06
59	$\text{OHNCO}^- + \text{NH}_3$	0.64	0.14	0.02
59	$\text{ONHCO}^- + \text{NH}_3$	1.10	0.59	0.46
59	$\text{NHCONH}_2^- + \text{OH}$	0.31	-0.13	-0.23
59	$\text{OHNHCNH}^- + \text{OH}$	3.92	3.48	3.37
59	$\text{ONHCNH}_2^- + \text{OH}$	3.94	3.51	3.40
58	$\text{NCONH}_2^- + \text{H}_2\text{O}$	-0.80	-1.28	-1.40
58	$\text{NHCONH}^- + \text{H}_2\text{O}$	-0.72	-1.21	-1.33
44	$\text{CONH}_2^- + \text{HONH}$	2.54	2.00	1.87
43	$\text{NHCNH}_2^- + \text{O}_2\text{H}$	4.16	3.63	3.51
42	$\text{NCO}^- + \text{NH}_2 + \text{H}_2\text{O}$	-0.71	-1.46	-1.67
42	$\text{ONC}^- + \text{NH}_2 + \text{H}_2\text{O}$	2.23	1.47	1.28
41	$\text{NCNH}^- + \text{H}_2\text{O} + \text{OH}$	0.75	-0.12	-0.34
40	$\text{NCN}^- + 2\text{H}_2\text{O}$	-0.64	-1.43	-1.62
33	$\text{O}_2\text{H}^- + \text{NHCNH}_2$	4.11	3.57	3.43
32	$\text{HONH}^- + \text{CONH}_2$	2.98	2.43	2.30
32	$\text{O}_2^- + \text{NH}_2\text{CNH}_2$	3.49	3.03	2.91
26	$\text{CN}^- + \text{O}_2\text{H} + \text{NH}_3$	2.54	1.61	1.38
26	$\text{CN}^- + \text{H}_2\text{O}_2 + \text{NH}_2$	3.40	2.47	2.24
26	$\text{CN}^- + \text{H}_2\text{O} + \text{NH}_2 + \text{O}$	4.79	3.54	3.23
17	$\text{OH}^- + \text{NHCONH}_2$	0.81	0.37	0.26
17	$\text{OH}^- + \text{H}_2\text{O} + \text{NCNH}$	1.51	0.64	0.42
17	$\text{OH}^- + \text{OHNHCNH}$	3.57	3.12	3.00
17	$\text{OH}^- + \text{ONHCNH}_2$	2.05	1.64	1.54
16	$\text{O}^- + \text{NH}_2\text{CONH}_2$	1.13	0.75	0.66
16	$\text{O}^- + \text{OHNHCNH}_2$	4.96	4.57	4.47
16	$\text{O}^- + \text{NH}_2 + \text{CONH}_2$	5.18	4.30	4.09
16	$\text{NH}_2^- + \text{OHNHCO}$	3.27	2.77	2.64
16	$\text{NH}_2^- + \text{O} + \text{CONH}_2$	5.78	4.91	4.69
15	$\text{NH}^- + \text{OH} + \text{CONH}_2$	5.82	4.94	4.72
1	$\text{H}^- + \text{ONHCONH}_2$	3.21	2.89	2.81
1	$\text{H}^- + \text{HONCONH}_2$	3.40	3.07	2.98
1	$\text{H}^- + \text{HONHCONH}$	4.55	4.23	4.15

form is commonly observed upon DEA to organic molecules such as amino acids and nucleobases.^{35,36} According to our calculations, the energetically most stable anion formed this way would be through dehydrogenation of the primary amine group with the thermodynamic reaction threshold of 1.21 eV at 362 K. This is in contrast with results of Remko *et al.*,³⁷ who calculated the most stable anionic species in the gas-phase to form due to deprotonation of the secondary amine group. According to Remko *et al.*,³⁷ the latter anion is more stable by about 0.03 eV at the B3LYP/6-311+G(d,p) level of theory at 0 K than the anion formed upon deprotonation of the primary amine group, whereas in case of our G4(MP2) calculations the respective energy difference is about -0.13 eV; a discrepancy which can be explained by the use of the different employed theoretical levels of theory.

Over the whole energy range that was examined, an anion with m/z 42 was detected, being in most cases also the dominant anion in the spectrum and particularly abundant at ~ 0 eV. By its mass-to-charge ratio, this anion can correspond to either NCO^- or CNO^- , as illustrated schematically in Fig. 2. However, formation of CNO^- is an endothermic process associated with a free reaction energy of 1.28 eV at 362 K according to our calculations. In contrast, formation of NCO^- yields a free reaction energy of -1.67 eV at 362 K and hence, an exothermic process. Thus we assign the predominant dissociation channel of HU close to zero eV to formation of the cyanate anion, NCO^- , via the following reaction:



The cyanate molecule has an extraordinary high (adiabatic) electron affinity (3.62 eV according to our G4(MP2) calculations), comparable to those of halogen atoms. Formation of this anion has also been observed upon DEA to other molecules of biological interest such as pyrimidine bases.³⁸

At low energies, close to ~ 0 eV, the two most abundant anions apart from NCO^- in the mass spectrum yielded m/z 41 and 59 and were assigned to the formation of NCNH^- and NHCONH_2^- , respectively. By comparison with the thermodynamic thresholds for the dissociation channels involving anions with m/z 59 (see Table I), it can be seen that formation of NHCONH_2^- corresponds probably to abstraction of the hydroxyl group from the secondary amine group with a free reaction energy of -0.23 eV. Formation of water and anions with m/z 58 yields also highly exothermic thermodynamic reaction thresholds, which is reflected too in the mass spectrum at ~ 0 eV corresponding to the assignment of NCONH_2^- . The anion formed at ~ 0 eV incident electron energy and detected at m/z 43 is indicated as HNCNH_2^- in Fig. 1. However, the thermodynamic threshold for formation of this anion yields 3.51 eV. For this reason, this peak in the mass spectrum as well as the one for m/z 46 is ascribed rather to impurity species present in the experimental chamber. At m/z 40, we note, however in comparably low abundance, eventually formation of the CN_2^- anion, which, at least thermodynamically, is a possible fragmentation pathway according to our calculations.

At higher incident electron energies (5–9 eV), anions with $m/z = 16$ and $m/z = 17$ are present in the respective mass spectra. The hydroxide anion with $m/z = 17$ is formed due to abscission of the hydroxyl group from the parent molecule above a thermodynamic threshold of 0.26 eV at 362 K according to our calculations. The ionic fragment with m/z 16 cannot be assigned unambiguously to the formation of either O^- or NH_2^- , as their lowest formation thresholds are both well below 5 eV.

One of the possible mechanisms underlying cell damage mediated by HU is through its hydrolysis to hydroxylamine (HA) and subsequent production of nitric oxide.²² Also formation of the CONH_2^- ion with m/z 44 upon DEA to HU may lead to formation of (in this case neutral) dehydrogenated hydroxylamine. Unfortunately, the significant formation of this anion was observed only for incident electrons of energy of 1 eV (see Fig. 1), whereas the thermochemical

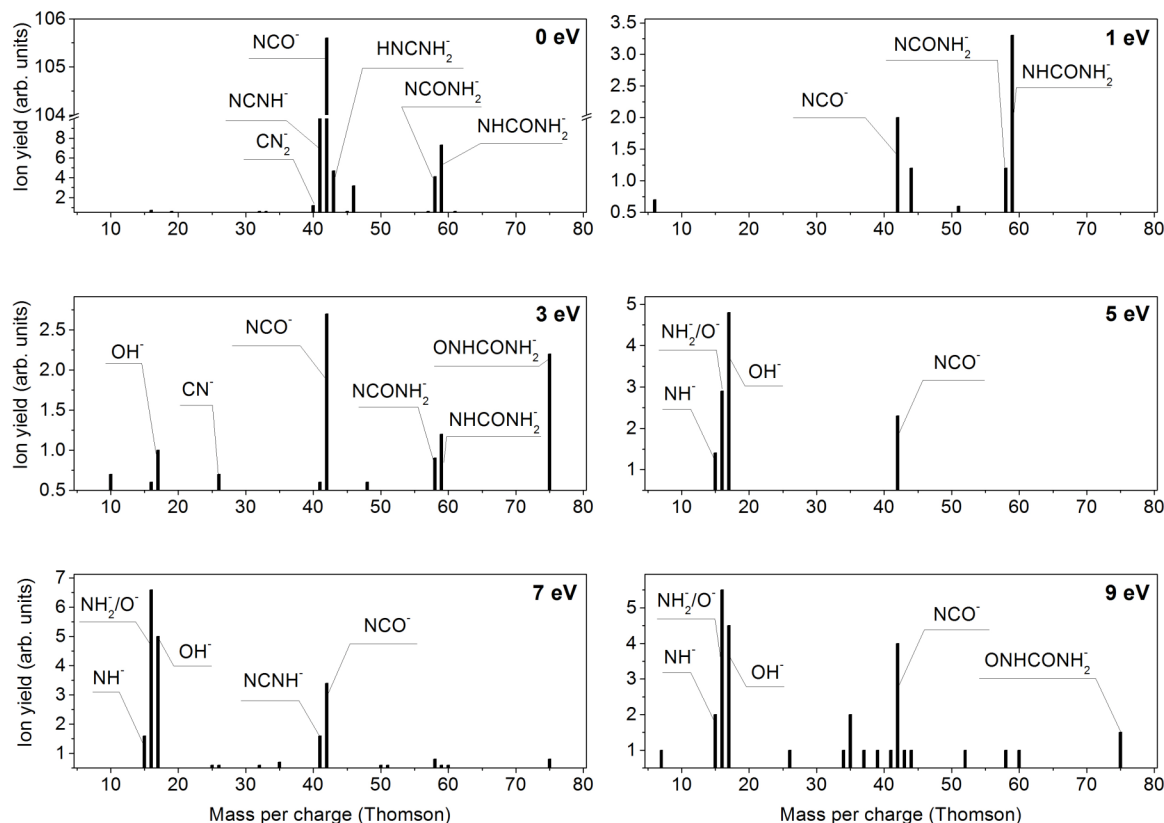


FIG. 1. Negative mass spectra obtained at the electron energies of ~ 0 , 1, 3, 5, 7, and 9 eV. The ion peaks are labeled according to their assigned chemical composition. See text for further details.

calculations predict its appearance for incident energies at least above 1.87 eV (see Table I), thus it was attributed also to impurities.

B. Ion efficiency curves

For the most prominent anions, ion efficiency curves are shown in Figs. 3 and 4. Their possible chemical composition and positions of their resonances are listed in Table II. For comparison, resonances obtained by Naff and co-workers³⁹ for the urea molecule, H_2NONH_2 , are also included in Table II.

DEA to HU, resulting in the formation of ONHCONH_2^- , NHCONH_2^- , NCONH_2^- , NCO^- , and CN^- is characterized by the high electron affinities of these fragments (e.g., 3.862 eV for CN or 3.609 eV for NCO^-)⁴⁰ and appears as a broad resonance feature centered around 2 eV (Fig. 3). This peak can be related to a similar one found for DEA to the urea molecule³⁹ and is attributed to a shape resonance.

In addition, we observe another broad feature, centered around 6 eV, detected for NH_2/O^- , OH^- , CN^- , HNOH^- , CNO^- , and NCONH_2^- (Fig. 3). Such a feature was also

observed for the urea molecule,³⁹ and we assign it to a core excited resonance, involving a $n \rightarrow \sigma^*$ transition.

As mentioned above, at all incident electron energies examined, the anion with m/z 42 was detected and attributed to $\text{NCO}^-/\text{CNO}^-$ formation (Fig. 3). The position of its resonance around 2.5 eV is similar to the maximum production yield for this ion formed upon DEA to urea (yielding 2.3 eV).³⁹ For this ion also a resonance structure centered at ~ 0 eV was detected (Fig. 4). In the near-zero electron energy region, three distinguished resonances, at ~ 0.0 , 0.1, and 0.4 eV can be seen. According to earlier theoretical investigations of gas-phase HU,⁴¹ there exist three stable isomers which are expected to contribute to the vibrational spectrum of this molecule. These rotational isomers differ mainly in rotations around the C–N and N–O bonds and the relative energies of their ground states are within 0.1 eV from each other. Similar results were also obtained for the urea molecule.⁴² Therefore, we assign the three resonances in the near-zero-eV region to the individual contributions of those isomers, with the one with the highest ion yield assigned to the most abundant keto-isomer of HU.

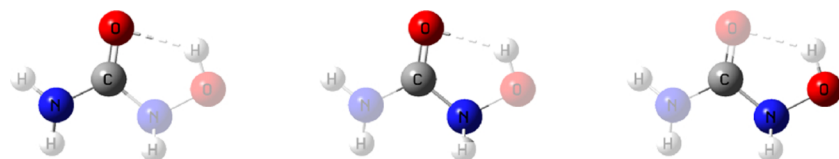


FIG. 2. Schematics indicating possible ways to obtain a fragment with m/z 42 from HU upon dissociative electron attachment: NCO^- (left and middle) and CNO^- (right) anions.

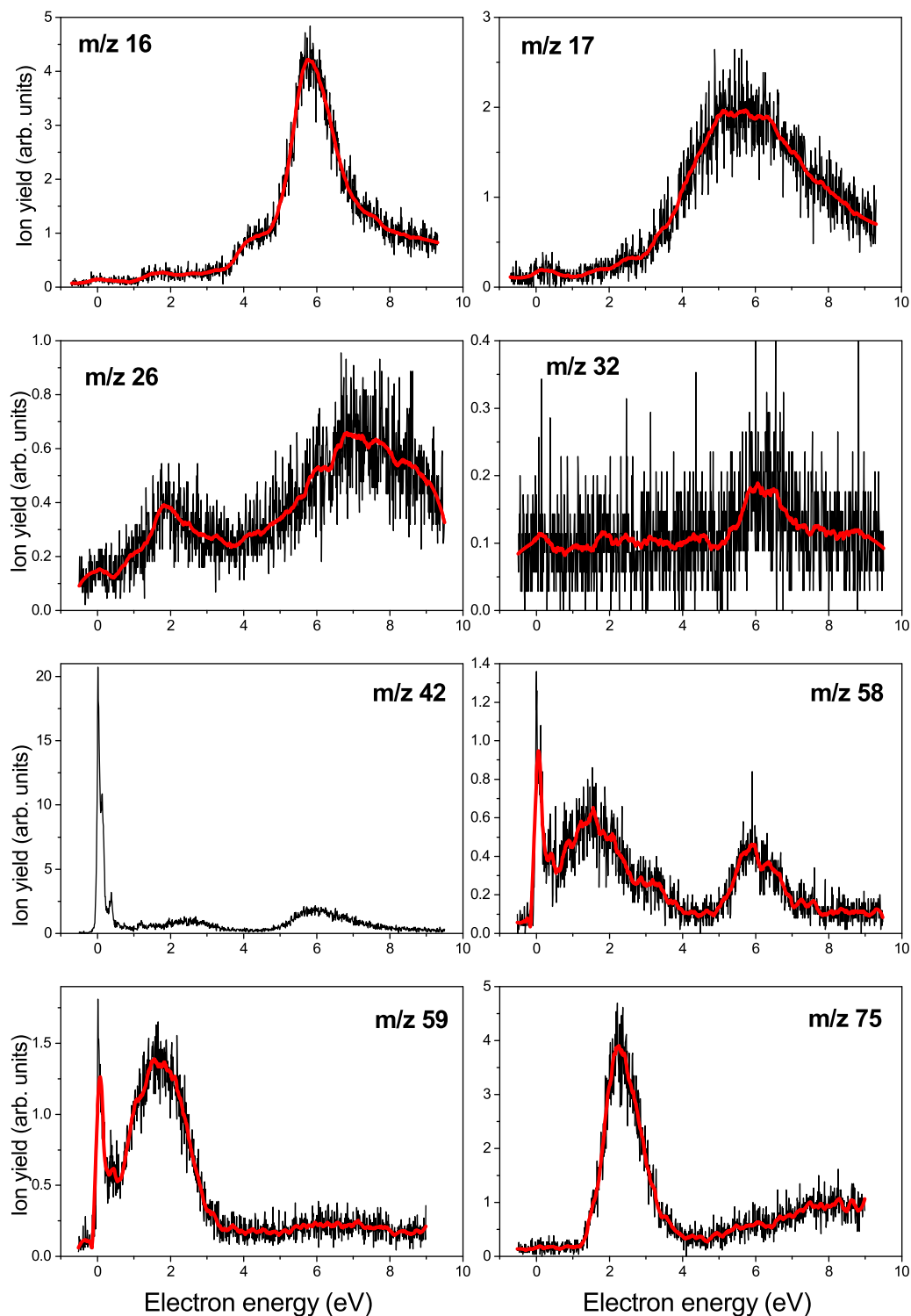


FIG. 3. Anion efficiency curves as a function of the incident electron energy for the most prominent fragments formed upon DEA to HU; the red lines are drawn to guide the eye.

In case of the ion with m/z 58, the sharp feature, positioned at about 0 eV, can be ascribed to NCONH_2^- formed via the formation of a water molecule (corresponding to the calculated free reaction energy of -1.40 eV at 362 K, see Table I), possibly via coupling with a vibrational bending mode of the parent ion. Vibrational analysis (at the B3LYP/6-31G(2df,p) level of theory) yields a vibrational mode which is associated with considerable change in geometry that could enhance

such a coupling. Moreover, we note a significantly smaller vibrational frequency of 1297 cm^{-1} associated with this mode in case of the anion compared to the respective mode of the neutral parent molecule yielding 1435 cm^{-1} . This is an indication of a weakening of the respective N–O and N–H bonds upon electron attachment, which possibly underlies fragmentation resulting in the formation of NCONH_2^- via coupling with this vibrational mode. A similar sharp feature is

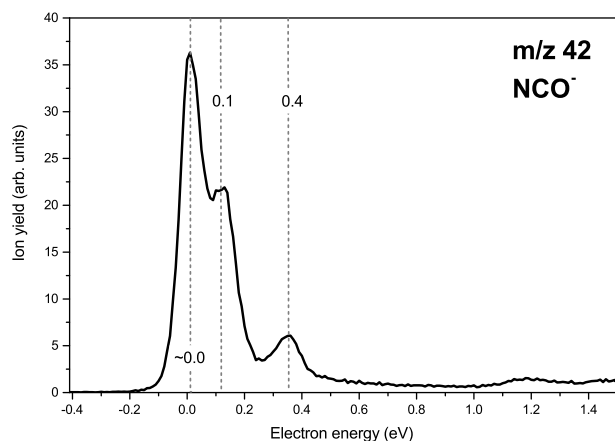


FIG. 4. Ion efficiency curve of the m/z 42 ion, NCO^- , formed upon DEA to HU, taken at the near-zero electron energy range; the dashed lines indicate the positions of the resonance peaks.

observed also in the m/z 59 spectrum ascribed to NHCONH_2^- formation (with a calculated free reaction energy of -0.23 eV at 362 K).

For the sake of an argument also an anion efficiency curve was recorded for the fragment with m/z 32 (Fig. 3). According to our calculations, this anion can be attributed to dehydrogenated hydroxylamine formation, HONH^- , with a reaction threshold of 2.30 eV. Due to high levels of noise, we can report only a broad structure centered around 6 eV. The inability of O_2^- formation in the gas-phase allows us to anticipate that the feature corresponds to HONH^- , i.e., the dehydrogenated hydroxylamine anion. The intensity of this anion is very low, thus the probability that upon electron attachment a reaction path involving deprotonated hydroxylamine plays a key role for radiosensitization of DNA by HU is rather negligible.

C. Formation of radicals

Our experimental setup does not allow to assign neutral fragments to specific reaction channels. Comparison with the quantum chemical data on the thermodynamic reaction thresholds, however, allows us to conclude that the three

TABLE II. Mass-to-charge ratio m/z , assigned anionic species and peak positions for some of the fragment ions formed upon DEA to HU; for comparison, resonant peak positions for DEA to urea³⁹ are also shown.

m/z (Thomson)	Anion	Resonant peak position (eV)	
		Hydroxyurea (HU)	Urea (U) ³⁹
16	NH_2/O^-	4.1, 5.9	5.8
17	OH^-	5.3	6.0
26	CN^-	1.9, 7.4	2.1
32	HNOH^-	6.2	...
42	NCO^-	$\sim 0/0.1/0.4, 2.5, 5.9$	2.3
58	NCONH_2^-	$\sim 0, 0.4, 1.5, 3.1, 6.0$...
59	NHCONH_2^-	$\sim 0, 0.4, 1.6$...
75	ONHCONH_2^-	5.8	5.8 (U-H) ⁻

most dominant fragmentation pathways associated with the formation of NCO^- (m/z 42), NCNH^- (m/z 41), and NHCONH_2^- (m/z 59) are linked to the formation of the amidogen radical and the highly reactive hydroxyl radical. Reactive oxygen radicals are relatively well-known to lead to cell damage and to be related to human cancers as well as to cancer treatment.⁴³ Especially the hydroxyl radical is highly reactive and can attack most biological molecules which leads to the propagation of free-radical chain reactions.⁴⁴ The investigation of the mode of action of the well-studied radiosensitizing chemotherapeutic drug cisplatin revealed that the production of hydroxyl radicals is partially responsible for the enhanced yield of DNA single- and double-strand breaks upon irradiation.⁴⁵ In contrast, less is known about the interaction of the amidogen radical or its hydrated form with the biomolecular environment. The radical has been linked to both the production and elimination of atmospheric nitrogen oxides.⁴⁶ The latter molecules have received consideration for their ability to induce cytotoxic and mutagenic effects when excessively present in the biomolecular environment of a cell.⁴⁷ However, further elucidation is required to clarify the effect the amidogen radical or the water-amidogen complex formed upon hydration may eventually have in biomolecular contexts. For the time being, we simply note that upon interaction between low energy electrons and HU reactive radical species are produced in high abundance, which may at least partially form the basis for HU's radiosensitizing properties.

V. CONCLUSION

In this work, fragmentation upon electron attachment over the energy range from 0 to 9 eV to the widely used chemotherapeutic drug hydroxyurea was investigated. Most of the fragments are formed not only at 0 eV but also at higher electron energies. The predominant energies, at which TNIs are formed, were found to be ~ 0 eV, ~ 2 eV, and ~ 6 eV. The comparison of the experimental data with quantum chemical calculations of thermodynamical reaction thresholds allowed us both to assign specific anion species to detected ionic fragments and to predict neutral fragments associated with some of these reaction channels. By far the most dominant dissociation channel for DEA to hydroxyurea is associated with the formation of the NCO^- and yields an abundance about 10 times higher than the second most abundant detected ion. Formation of NCO^- is associated with the formation of the amidogen radical, which might underlie the radiosensitizing properties of hydroxyurea to some extent, e.g., by subsequent production of nitric oxide or the highly reactive hydroxyl radical in the biomolecular environment. The two second most abundant ionic fragments were assigned to NCNH^- and NHCONH_2^- , which are both directly associated with the production of the hydroxyl radical, which is well known for its relation to cell damage. No evidence was found that hydroxylamine or its dehydrogenated form may be formed upon low energy electron dissociative attachment. Thus, formation of hydroxylamine upon irradiation is not expected

to contribute to an increased activity of hydroxyurea in living cells.

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