Dissociative Electron Attachment to Amino-Acids: the case of Leucine

ARTICLE *in* CHEMICAL PHYSICS LETTERS · AUGUST 2009

Impact Factor: 1.9 · DOI: 10.1016/j.cplett.2009.07.021

Chemical Physics Letters 477 (2009) 245–248

Contents lists available at ScienceDirect

Chemical Physics Letters

journal homepage: www.elsevier.com/locate/cplett

Dissociative electron attachment to amino-acids: The case of Leucine ☆

H. Abdoul-Carime [b,*], C. König-Lehmann [a], J. Kopyra [c], B. Farizon [b], M. Farizon [b], E. Illenberger [a]

[a] Institut für Chemie–Physikalische und Theoretische Chemie, Freie Universität Berlin, Takustrasse 3, D-14195 Berlin, Germany
[b] Université de Lyon, Université Lyon 1, Villeurbanne, CNRS/IN2P3, UMR5822, Institut de Physique Nucléaire de Lyon, F-69003 Lyon, France
[c] University of Podlasie, Department of Chemistry, 3 Maja 54, 08110 Siedlce, Poland

ARTICLE INFO

Article history:
Received 11 June 2009
In final form 6 July 2009
Available online 9 July 2009

ABSTRACT

In this work, mechanisms that control the dissociation of Leucine amino-acid by low-energy (<10 eV) electrons are investigated. At sub-excitation energies, Dissociative Electron Attachment is initiated by a shape resonance, whereas core-excited resonances is involved at higher energies. Under the present conditions, we observe three fragment ions $(Leu\text{-}H)^-$, $(Leu\text{-}16\ amu)^-$ and anion at 42 amu. The $(Leu\text{-}16)^-$ species observed near 1.0 eV and the (m/z) 42 anion produced near 7.8 eV are interpreted to arise from the decomposition of the amino-acid side group. The $(Leu\text{-}H)^-$ anion observed at 1.1 eV is induced by from the fragmentation of the carboxylic group.

1. Introduction

Chemistry induced by low energy electrons is now established in a large variety of research fields [1–3], among which are nano-scale lithography and radiation science. In nanolithography, demands for smaller and smaller dimensions in integrated circuit are growing [4] and standard electron beam or combined Atomic Force Microscope to Scanning Tunneling Microscope (i.e., Scanning Probe Lithography [5]) techniques have been shown to be appropriated methods to take up the challenge [6]. In radiation science, ballistic secondary electrons, with the most probable and mean energies in the vicinity of 9 eV–10 eV, are the copiously produced species by the attenuation of energetic primary ionizing quanta [7,8] and their subsequent lethal actions have been demonstrated [9,10].

While investigating biological molecular systems and their interaction with electrons is in straight line of radiation science, in the nano-scale field it arises from the use of (bio-)polymers for processing new type of material such as bio-sensors or for preparation of surfaces for subsequent usage (e.g., nanopatterning) [11,12]. (Bio-)polymers are complex structures of simpler sub-units linked together via chemical bonds. In DNA, a sub-unit consists of a sequence of phosphate–sugar–DNA base (e.g., a nucleotide) and in proteins, amino-acids are the building-block. Previous investigation have shown that damages to bio-polymers by low-energy (<15 eV) electrons arise by the alteration of their sub-units [13–15]. Thus, the investigation of the degradation of sub-units becomes a pre-requisite for a better comprehension of the alteration of bio-polymers, and more particularly their functionalities.

In the present work, we investigate gas phase Leucine (Leu) in order to reveal the intrinsic properties with respect to low energy electron interaction. The structure of Leu is exhibited in Fig. 1. Most amino-acids consist of a carboxylic (COOH), an amino (NH_2) and a side group bound to the so-called α-C. Leucine is the fourth of a series of 6 non polar aliphatic amino-acids differentiated by the length and the structure of the side group. The side chain of Glycine (Gly) consists of a simple H atom, that of Alanine (Ala), Valine (Val), and Leucine are CH_3, $(CH_3)_2$–CH, $(CH_3)_2$–CH–CH_2, respectively. The side group confers to the amino-acid its specific functionality within the bio-polymers. For Leucine, the hydrophobic side chain $(CH_3)_2$–CH–CH_2 is involved, as an example, in the peculiar so-called 'zipping' structure or bio-molecular Velcro® [16]. Previous DEA experiments undertaken on gas phase Gly, Ala, Val have reported large number of dissociation channels [17–19]. In Leu only three fragment ions are observed as discussed below.

2. Experimental

The experimental apparatus has been described elsewhere [20]. Briefly, an electron beam of defined energy (~10 nA, FWHM ~0.25 eV) generated by an trochoidial electron monochromator crosses at right angles an effusive beam of Leu. This latter emanates from a vessel containing approximately 20 mg of 99% purity powder (Aldrich Ltd.) heated by two *in vaccuo* halogen bulbs. These lamps also prevent the powder from condensation on the surfaces (e.g., plates), which otherwise may lead to undesirable change in contact potentials during measurements. The operating

☆ During the submission process of the manuscript, we have been informed that Dr. P. Papp and co-workers from the Department of Physics at the Comenius University have also performed DEA to Leucine and Isoleucine measurements. In the case of Leucine, their observation corroborates with our findings.

* Corresponding author.
E-mail address: hcarime@ipnl.in2p3.fr (H. Abdoul-Carime).

0009-2614/$ - see front matter © 2009 Elsevier B.V. All rights reserved.
doi:10.1016/j.cplett.2009.07.021

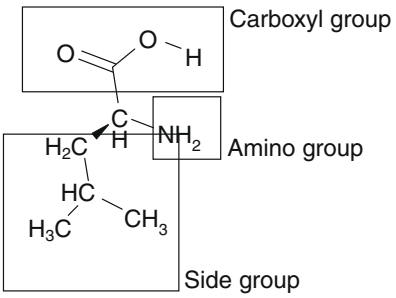

Fig. 1. Structure of Leucine amino-acid. The dashed-square defines the different groups within the molecule: the carboxyl group, the amino group and the side group. Each amino-acid differs from another by the side group that confers to the molecule its specific functionality.

temperature of approximately 420 K measured by a platinum resistance directly at the oven is well below the molecular decomposition temperature (480 K [21]). Therefore, the original structure of Leu produced in gas phase is likely to remain intact. Thermal alteration of amino-acids may lead, for instance, to the formation of CO_2 from the carboxylic COOH group [22]. Thus, the O^- anion would have been detected, which is not seen presently. The negative ions are extracted from the reaction volume by a small electric field (< 0.5 $V{\cdot}cm^{-1}$) towards a quadrupole mass analyzer, and are detected by single pulse counting techniques. Finally, calibration of the energy scale is performed by means of SF_6, in a sequential procedure, avoiding the presence of SF_6 while recording spectra from Leu. Indeed, SF_6^- generated in the collision region can induce dissociative electron transfer reactions in the target molecules, leading to dehydrogenated anions [23,24].

3. Results and discussion

Within our experimental conditions, we detect three negatively charged fragments ascribed to the dehydrogenated Leucine $(Leu\text{-}H)^-$, the loss of a neutral 16 amu species to form the $(Leu\text{-}16)^-$ anion and the (m/z) 42 anion. Fig. 2 displays their yields as a function of the incident electron energy. The curves exhibit resonant structures located at (1.1 eV and 4.6 eV), (1.0 and 2.0 eV) and (∼5.2 and ∼8.0 eV) for the respective ion fragments. Such structures are typical signatures of DEA [25].

At energies below the first electronic state of such molecules (∼4.5 eV [26]), DEA is initiated by a shape resonance, for which the excess electron occupies a usually un-filled π^* molecular orbital (MO) [27]. This temporary negative ion then undergoes dissociation into a negatively charged fragment and in general one neutral counterpart. The loss of hydrogen for the $(Leu\text{-}H)^-$ formation observed at 1.1 eV is likely to arise from the decomposition of the COOH carboxyl group. This assignment is supported by theoretical [28,29] and experimental [17–19,27] investigations on formic acid HCOOH and similar aliphatic amino-acids. However, mechanisms of hydrogen loss from the carboxylic –COOH group are still a matter of current discussions. Indeed calculations suggest that the excess electron is initially resonantly trapped in a π^* state. Such a state is non dissociative in the case of the planar geometry of HCOOH. Therefore some out-of-plane distortions are required for the π^*-MO to be coupled to a dissociative σ^*-MO, that further leads to the cleavage of the HCOO–H bond [28]. On the other hand in a more recent R-matrix study, it has been shown that DEA involves the capture of the excess electron into a strongly anti-bonding σ^*(O–H) MO [29]. The unresolved broad structure following the sharp peak can be attributed to a competition between DEA and excitation of vibrational modes as seen in Electron Energy Loss Spectra (EELS) and DEA to Glycine and Alanine experiments [30]. The resonant structure is observed here in the lower energy (1.1 eV) than that reported previously for various dehydrogenated amino-acid ion fragments, i.e., ∼1.2 eV [17–19,24]. From the values of electron affinity and bond dissociation of similar amino-acids, the dissociation threshold is estimated to be near 1 eV [18]. It should be noted that the values are calculated for molecular systems at ground state while, experiments are undertaken at 420 K. DEA to vibrationally excited molecules experiments have demonstrated appearance/enhancement of structure located at lower energies [31].

The loss of a neutral 16 amu fragment from Leucine molecule $(Leu\text{-}16)^-$ anion production is more ambiguous. From the stoichiometry, the 16 amu species may be attributed to O, NH_2 or CH_4 neutral products arising from the decomposition at

(1) the carboxylic group by
  (1a) a double C=O bond breakage, resulting in the loss of O;
  (1b) more complex pathways consisting of C–O and O–H bonds dissociation concomitantly to a C–H bond reformation producing also the $(Leu\text{-}16)^-$;
(2) the amino group via a single C–N bond rupture producing a neutral NH_2;
(3) the end of the side chain involving atoms scrambling that leads to the release of a CH_4 methane molecule.

With available thermodynamic values [32,33], the loss of O atom via (1a) and (1b) would require 7.6 and 5.5 eV, respectively, and hence unlikely at 1.0 and 2.0 eV. The cleavage of a N–H bond via reaction (2) would require 3.4 eV. It may nevertheless arise if the electron affinity of neutral (Leu-16 amu) is sufficiently high (i.e., ∼2 eV) to drive the dissociation mechanism. On the other hand, none of the previous works on DEA to various aliphatic amino-acids have reported the loss of 16 amu species and particularly at energies below 2.0 eV [17–19]. Thus, we suggest that, the loss of 16 amu species in the present work is likely to arise from the dissociation of the side group via reaction (3). In this reaction, the rupture of both H_3C–C and C–H bonds concomitantly to the reformation of a H–CH_3 bond requires 3.0–3.5 eV, suggesting that the electron affinity of the generated bi-radical must be at least equally high to support the dissociative channel. Reaction (3) can occur when the lifetime of the negative parent ion is sufficiently long to sustain deformation of molecular orbitals and substantial rearrangement of the nuclei. Molecular fragmentation induced by electron impact via concerted reactions with hydrogen atom displacement has already been reported below 10 eV for ethylene carbonate [34].

In Fig. 2, the $(Leu\text{-}H)^-$ and the (m/z) 42 amu ion yield exhibit peaks located at ∼5 eV and (∼5.2 and ∼7.8 eV), respectively. At these energies, DEA is likely to proceed via core-excited (i.e., two particle-one hole) resonances. The incoming electron transfers energy to electronically excite the target molecule and concomitantly becomes trapped by the field of the positive core [35]. The resonant feature observed in the present work may be compared to the structure reported at 5.3 eV by EELS measurements [30] and at 5.9 eV by theoretical calculations on similar amino-acids (e.g., Glycine, Alanine) [36]. It has been attributed to a $n_O\pi^*_{CO}$ MOs transition. Although involving electrons of the carboxylic group, dehydrogenation of Leucine at ∼5 eV is likely to arise from any of the C–H bond of the amino-acid. This suggestion is supported by study of DEA to partly deuterated Glycine (d-Gly), for which hydrogen atoms at the carbon sites are substituted by D atoms. It has been observed that the resonant signature near 7 eV arises from a loss of a D atom from the cleavage of a C-D bond, producing $(Gly\text{-}D)^-$ anion [37]. Therefore, we suggest that the $n_O\pi^*_{CO}$ MOs transition leads to the loss of a hydrogen atom from any of the C–H sites.

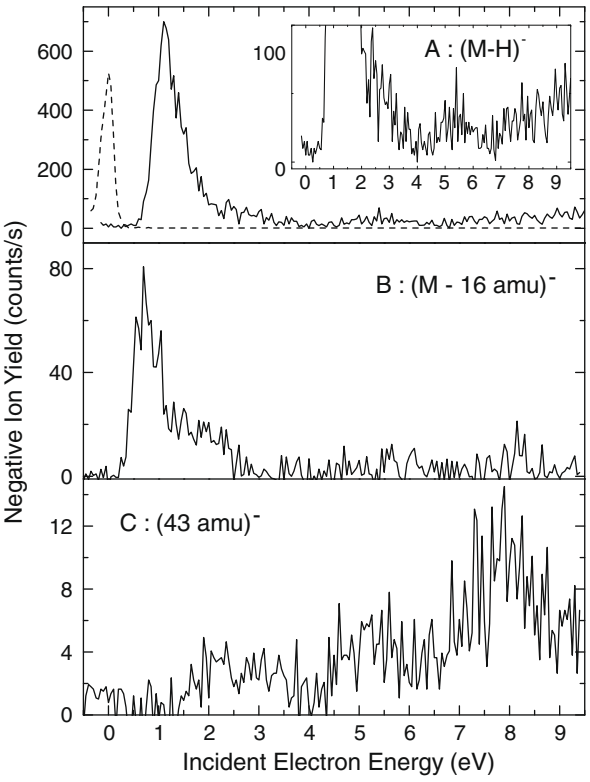

Fig. 2. Yield of ion fragments (a) $(Leu\text{-}H)^-$, (b) $(Leu\text{-}16)^-$ and (c) (m/e) 42 anion as function of incident electron energy. The 3–8 eV energy range is magnified in the inset of (a) to show the presence of a weak resonant structure. The dashed curve represents the yield function of SF_6^- calibration gas, however attenuated by a factor of 10.

The yield function of the (m/z) 42 anion fragment exhibits two clear resonances located at 5.2 and 7.8 eV, and additionally a very weak resonant feature near 2 eV. A previous study of DEA to Alanine has also reported this anion fragment with a similar signature as shown in Fig. 2, with the two lowest energy resonances coinciding with our observation [19]. In their work, Ptasinska et al., have attributed this fragment to either the $C2H4N^-$ or NCO^- arising from the fragmentation and atom re-arrangement of the amino and carboxylic group. Therefore, it is likely that the (m/z) 42 anion observed in the present work correspond to $C2H4N^-/NCO^-$ species. The high electron affinity of the iso-cyanato radical NCO (3.61 eV [38]) may drive the molecular decomposition which requires quite a complex bond dissociation and rearrangements as indicated by the structure of the amino-acid in Fig. 1. However, the ion yield observed at 7.8 eV in the present work is stronger that that reported at 9.0 eV by these authors [19]. From the structure of Leucine, the fragmentation of the side group may also produce the $(C3H6)^-$ anion species with a mass-to-charge ratio of 42. Thus, at this energy, the (m/z) 42 anion fragment can be ascribed to the $(C3H6)^-$ anion species. It is interesting to note that in electron impact to similar aliphatic amino-acids experiments (Gly and Val), this (m/z) 42 anion species has not been observed [17,18].

4. Conclusion

Under the present conditions, Leucine exhibits only three detectable fragments $(Leu\text{-}H)^-$, $(Leu\text{-}16)^-$ and (m/z) 42 anions, in contrary to previously investigated protein building-blocks [17–19,24] for which many more ion fragments have been observed.

In the gas phase, the dominant ion fragment channel by far consists of the dehydrogenation of the amino-acid at the carboxylic group. However within a polymer of amino-acids (e.g., peptides or proteins), amino-acids are joined covalently through an amide linkage formed by removal of OH from the carboxylic CO–OH group and H from the amino H–NH group of the adjacent amino-acid [39]. The loss of hydrogen from the COOH site observed in the isolated compound is not present in the bio-polymer. The alteration of the Leucine sub-unit is likely to arise mainly at the side chain, producing neutral CH_4 and ionic $(C_3H_6)^-$ and their respective counterparts, $(Leu\text{-}16)^-$ and (Leu-42). Such an alteration at the molecular level may subsequently affect the functionality of proteins. As an example, the Leucine side chain is responsible of the structure of proteins, known as Leucine zippers [40], found naturally in living organism [41] or artificially built [42,43].

Acknowledgments

This work is supported by the European Union, the Fonds der Chemischen Industrie, the Deutsche *Forschungsgemeinschaft* (DFG) and the Freie Universität Berlin. HAC and JK acknowledge support for a visit to Berlin from the European Union via the COST action CM0601 (Electron Controlled Chemical Lithography, ECCL).

References

[1] S. Lacombe et al., Phys. Rev. Lett. 79 (1997) 1146.
[2] M.T. Sieger, W.C. Simpson, T.M. Orlando, Nature 394 (1998) 554.
[3] L. Sanche, IEEE Trans. DEI 4 (1998) 507.
[4] National Technology Roadmap for Semiconductors, Semiconductor Industry Association, San Jose, CA, 1997.
[5] M. Ishibashi, S. Heike, H. Kajiyama, Y. Wada, T. Hchizuma, Appl. Phys. Lett. 72 (1998) 1581.
[6] K. Wilder, C.F. Quate, B. Singh, D.F. Kyser, J. Vac. Sci. Technol. B 16 (1998) 3864.
[7] 5×10^4 electrons produced per MeV energy deposited, from the ICRU Report Vol. 31, International Commission on Radiation Units and Measurements, Washington, DC, 1979.
[8] S.M. Pimblott, J.A. Laverne, Radiat. Phys. Chem. 76 (2007) 1244.
[9] G.P. Collins, Sci. Am. 26 (August) (2003).
[10] B. Bouda, P. Cloutier, D. Hunting, M.A. Huels, L. Sanche, Science 287 (2000) 1658.
[11] C.K. Harnett, K.M. Satyalakshmi, H.G. Craighead, Appl. Phys. Lett. 76 (2000) 2466.
[12] K.L. Christman, E. Schopf, R.M. Broyer, R.C. Li, Y. Chen, H.D. Maynard, J. Am. Chem. Soc. 131 (2009) 521.
[13] H. Abdoul-Carime, L. Sanche, Int. J. Radiat. Biol. 78 (2002) 89.
[14] S.G. Ray, S.S. Daube, R. Naaman, PNAS 102 (2005) 115.
[15] H. Abdoul-Carime, S. Gohlke, E. Illenberger, J. Am. Chem. Soc. 126 (2004) 12158.
[16] E.K. O'Shea, K.J. Lump, P.S. Kim, Curr. Biol. 3 (1993) 658.
[17] S. Ptasinska, S. Denifl, A. Abedi, P. Scheier, T.D. Märk, Anal. Bioanal. Chem. 377 (2003) 1115.
[18] P. Papp, J. Urban, S. Matejcik, M. Stano, O. Ingolfsson, J. Chem. Phys. 125 (2006) 204301.
[19] S. Ptasinska et al., Chem. Phys. Lett. 403 (2005) 107.
[20] I. Hahndorf, E. Illenberger, Int. J. Mass Spect. Ion Proc. 167/168 (1997) 87.
[21] F. Rodante, G. Marrosu, G. Catalani, Thermochem. Acta 194 (1992) 197.
[22] P.H. Cannington, S. Ham, J. Electron. Spectrosc. Relat. Phenom. 15 (1979) 79.
[23] S. Gohlke, H. Abdoul-Carime, E. Illenberger, Chem. Phys. Lett. 380 (2003) 595.
[24] H. Abdoul-Carime, S. Gohlke, E. Illenberger, Chem. Phys. Lett. 402 (2005) 497.
[25] E. Illenberger, in: H. Baumgärtel, E.U. Frank, W. Grünbein (Eds.), Gaseous Molecular Ions, Topics in Physical Chemistry, vol. 2, Steinkopff, Darmstadt, Springer, New York, 1992.
[26] R. Maul, M. Preuss, F. Ortmann, K. Hannewald, F. Bechstedt, J. Phys. Chem. A 111 (2007) 4370.
[27] A.M. Scheer, P. Mozejko, G.A. Gallup, P.D. Burrow, J. Chem. Phys. 126 (2007) 174301.
[28] T.N. Rescigno, C.S. Trevisan, A.E. Orel, Phys. Rev. Lett. 96 (2006) 213201.
[29] G.A. Gallup, P.D. Burrow, I.I. Fabrikant, Phys. Rev. A 79 (2009) 042701.
[30] R. Abouaf, Chem. Phys. Lett. 451 (2008) 25.
[31] A. Rosa, F. Brüning, S.V.K. Kumar, E. Illenberger, Chem. Phys. Lett. 391 (2004) 361.
[32] S.J. Blanksby, G.B. Ellison, Acc. Chem. Res. 36 (2003) 255.
[33] T.H. Lowry, K.H. Richardson, in: Mechanism and Theory in Organic Chemistry, third Edn., Harper Collins Publishing, 1987.
[34] M. Stepanovic, Y. Pariat, M. Allan, J. Chem. Phys. 110 (1999) 11376.
[35] G.J. Schulz, Rev. Mod. Phys. 45 (1973) 423.

[36] M. Tashiro, J. Chem. Phys. 129 (2008) 164308.
[37] S. Gohlke, Dissertation, Fachbereich Chemie und Biochemie, FU-Berlin, 2006
[38] J.C. Rienstra-Kiracofe, G.S. Tschumper, H.F. Schaefer III, Chem. Rev. 102 (2002) 231.
[39] D.L. Nelson, M.M. Cox, in: Lehninger, Principles of Biochemistry, fifth Edn., W.H. Freeman and Company, 2008.
[40] P.R.E. Mittl et al., PNAS 97 (2000) 2562.
[41] M.T. Henzl, M.E. Davis, A. Tan, Biochemistry 47 (2008) 13635.
[42] W. Shen et al., Marcomolecules 38 (2009) 3909.
[43] M.G. Ryadnov, Biochem. Soc. Transac. 35 (2007) 487.