Available online at www.sciencedirect.com
SCIENCE DIRECT
International Journal of Mass Spectrometry 253 (2006) 141–145
International Journal of Mass Spectrometry
www.elsevier.com/locate/ijms

Short communication

ESI-MS fragmentation pathways of *N*-methylpyrrole polyamide/peptide conjugates

Yong Ye [a,*], Li-Feng Cao [a], Ming-Yu Niu [a,b], Xin-Cheng Liao [a], Yu-Fen Zhao [a,b,*]

[a] *Department of Chemistry, Key Laboratory of Chemical Biology and Organic Chemistry, Zhengzhou University, Zhengzhou 450052, PR China*
[b] *The Key Laboratory for Bioorganic Phosphorus Chemistry and Chemical Biology, Ministry of Education, Department of Chemistry, Tsinghua University, Beijing 100084, PR China*

Received 12 February 2006; accepted 12 February 2006
Available online 23 March 2006

Abstract

MS/MS fragmentation pathways of polyamide/peptide conjugates, which are synthetic analogues of natural products with DNA affinity are discussed. The main fragmentation pathways involve the cleavage of the C–CO between rings and carbonyl groups (*a* cleavage), CO–NH amide bonds (*b* cleavage).

Keywords: ESI/MS/MS; Poliamidic oligomer; DNA

1. Introduction

The natural product distamycin and netropsin are polyamide containing three *N*-methylpyrrole (Py) moieties. They have ability to binding to the minor groove of A/T rich sequences of double helical DNA [1–14]. A number of such compounds have been synthesized and their biological activity explored. Some of the compounds can bind DNA with great specificity [15–19]. We are synthesizing various polyamides and their peptide conjugates, then studying their interaction with DNA. In this paper, the fragmentation of a series of synthetic polyamide and their peptide conjugates was investigated using electrospray ionization mass spectrometry (ESI-MS) combined with tandem mass spectrometry (ESI-MS/MS).

2. Experimental

The polyamide and its peptide conjugates were prepared according to published procedures [20]. Mass spectra were acquired in positive ion mode using a Bruker ESQUIRE-LCTM ion trap spectrometer equipped with a gas nebulizer probe, capable of analyzing ions up to *m/z* 6000. Nitrogen was used as drying gas at a flow rate of 4 L/min. The nebulizer pressure was 7 psi. The capillary was typically held at 4 kV and the source temperature was maintained at 300 °C. The instrument was operated at unit-mass resolution; calibration of *m/z* was performed using a standard ES-tuning-mix. The samples were continuously infused into the ESI chamber by a Cole-Parmer 74900 syringe pump (Cole Parmer Instrument Company, Vernon Hills, IL).

3. Results and discussion

We synthesized some polyamide/peptide conjugates. The polyamide is believed to specifically bind DNA and the dipeptide has been demonstrated to induce DNA cleavage. We also synthesized some polyamide/histidine conjugates in order to obtain polyamide/dipeptide conjugates then compared their activity. The MS/MS spectral data of the $[M+H]^+$ ions, and of the most significant fragment ions of the five compounds, are summarized in Tables 1 and 2. The main fragmentation are cleavages labeled as *a*, *b* and *m* (Scheme 1). The structure of compounds **1**–**5** is similar. They all have polyamide, 1,6-hexanediamine and His group. The most striking feature of the MS/MS spectra of the $[M+H]^+$ ions of them is the fragment ions Y_5, Y_6, Y_6, Y_5, X_1/Y_6, X_2/Y_6 at *m/z* 513, 650, 1056, 919, 853 and 524, all due to cleavage of the CO–NH bond. Other important features of the MS/MS spectra of compounds **1**–**5** are the fragment ions a_4, a_3, X_1/Y_5, X_2/Y_6, X_1, X_2, X_3, all due to cleavage of CO–C bond (*a* cleavage).

* Corresponding authors. Tel.: +86 371 67767050; fax: +86 371 67767051.
E-mail address: yeyong03@tsinghua.org.cn (Y. Ye).

1387-3806/$ – see front matter © 2006 Published by Elsevier B.V.
doi:10.1016/j.ijms.2006.02.005

Table 1
MS^2 and MS^3 spectra data of the conjugates

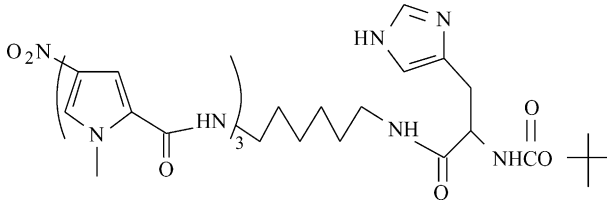
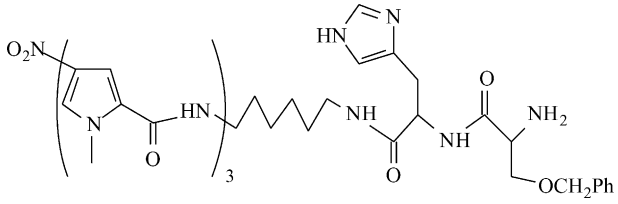
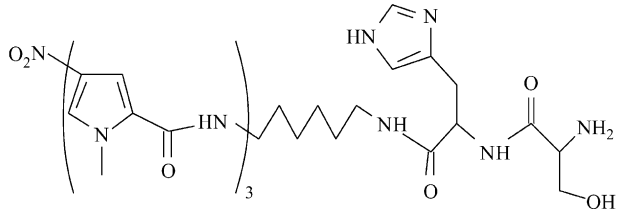
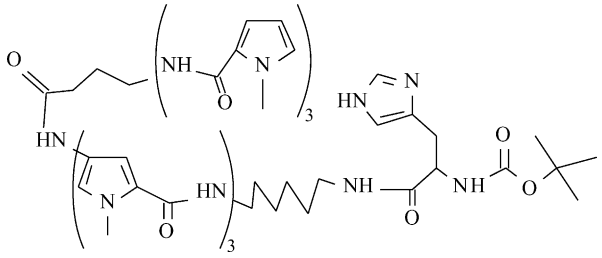
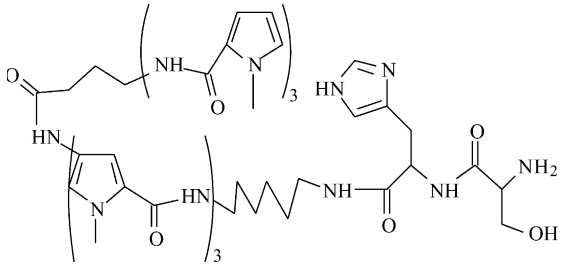
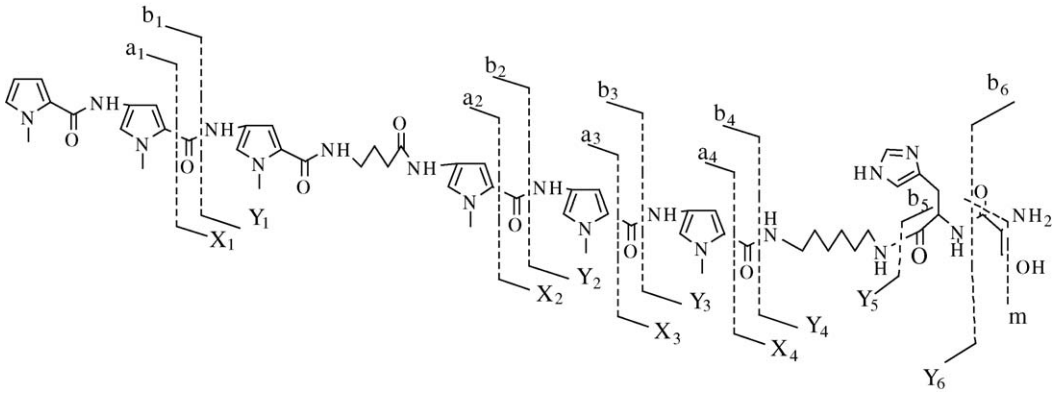
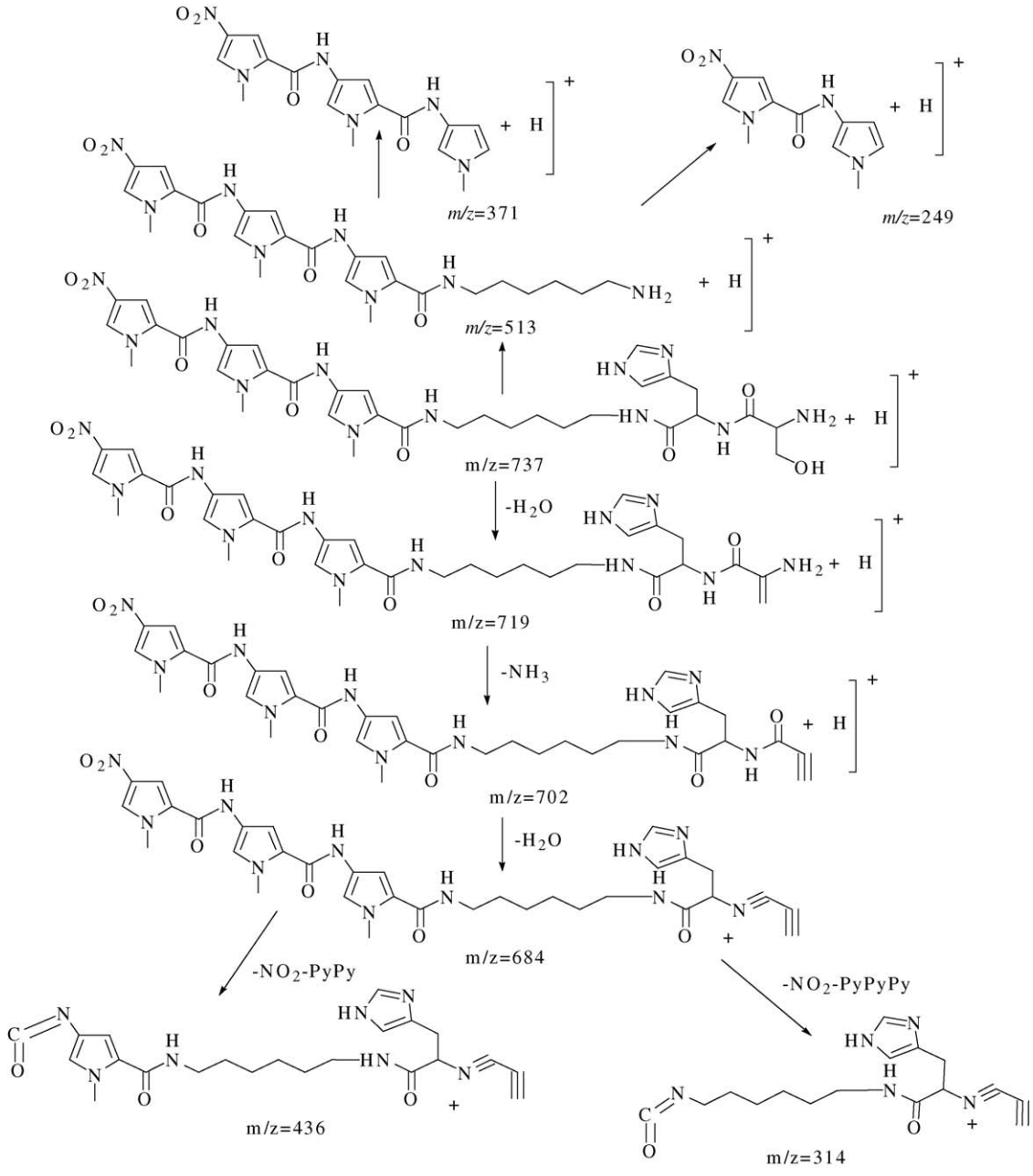
No.	Chemical structures	Precursor	Fragment ions (rel, int)					
			Y_6	Y_5	a_4	a_3	b_3	m
1		750(71) 650(57) 513(92)	**650(100)**	**513(3)** **513(100)**	**371(36)** **371(100)**		**275(6)** **275(16)**	
2		827(79) 513(31) 371(66)		**513(100)**	**371(18)** **371(100)**	**249(17)** **249(100)**	**275(4)** **275(61)**	**684(25)**
3		737(19) 513(100) 684(70) 371(55)	**650(4)**	**513(100)**	**371(21)** **371(49)**	**249(13)** **249(93)**	**275(100)**	**684(9)**
4		1156(37) 1056(49)	**1056(100)**	**919(100)**				
5		1144(78) 941(97) 919(100) 716(100) 611(41)		**919(100)**				

Table 2
MS^2 and MS^3 spectra data of the conjugates (continue)

No.	Fragment ions (rel, int)											
	X_1	X_2	X_3	X_3/m	X_4/m	X_1/Y_6	X_1/Y_5	X_2/Y_6	X_1/b_3	X_1/a_2	X_1/a_4	X_2/Y_5
1												
2												
3			**489(6)**									
				436(100)	**314(45)**							
4						**853(52)**	**716(16)**	**524(26)**				
5	**941(21)**	**611(8)**	**489(7)**				**716(15)**					
						853(18)	**716(100)**		**478(24)**	**330(13)**	**574(28)**	
							716(24)	**524(21)**				
											574(28)	**387(28)**
												387(100)



Scheme 1. Main fragmentation modes.



Scheme 2. Fragmentation pathway of the compound **3**.

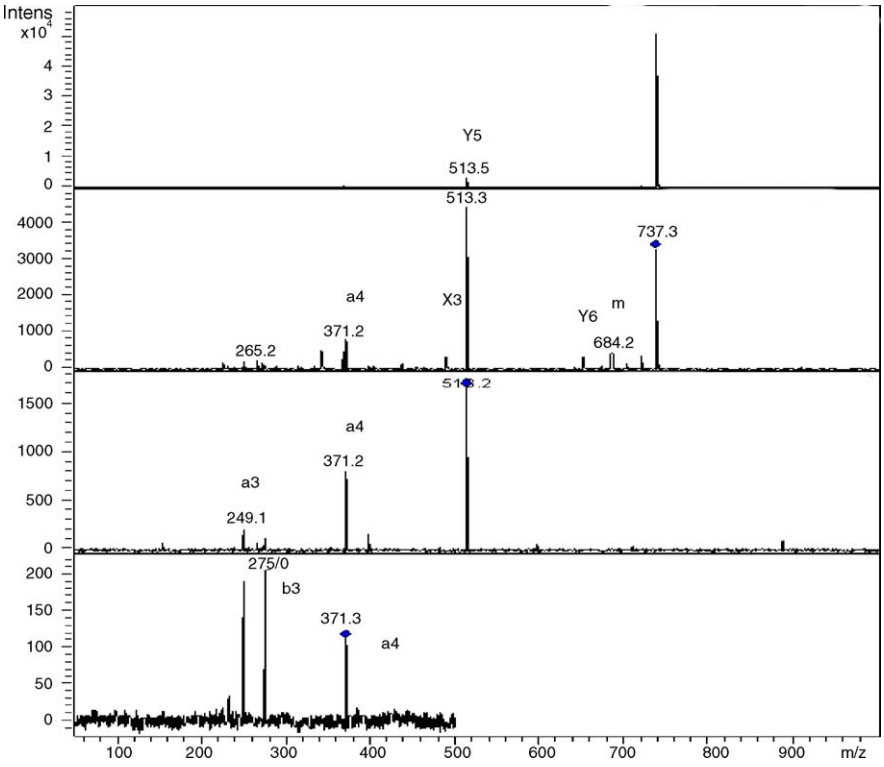


Fig. 1. The MS and MS^n spectra of compound **3**.

The fragment ions Y_5 at *m/z* 513 in the MS/MS spectra of the $[M+H]^+$ ions of compounds **1**–**3** are due to expulsion of His group (Scheme 1); the ions a_4 at *m/z* 371 correspond to cleavage of the C–CO bond between the ring and carbonyl group and the ions b_3 at *m/z* 275 correspond to cleavage of the CO–NH bond between the two rings.

In order to better understand the fragmentation mechanisms of these polyamides/peptide conjugate, the MS/MS/MS spectra of compound **3** was recorded. Fragmentation pathway is shown in Scheme 2. The decomposition of $[M+H\text{-}224]^+$ yields ions at *m/z* 513 by an a path cleavage of CO–NH amide bonds between the NH groups of 1,6-hexanediamine and His group. The formation of the ions at *m/z* 249 and *m/z* 371 occurs by the *a* path cleavage between the ring and carbonyl group with rearrangement of one hydrogen atom, while the ions observed at *m/z* 314 and *m/z* 436 result from an *m* path cleavage (Scheme 2). The CO–NH amide bond of His–Ser dipeptide cleavages produce the ions at *m/z* 650 (Fig. 1).

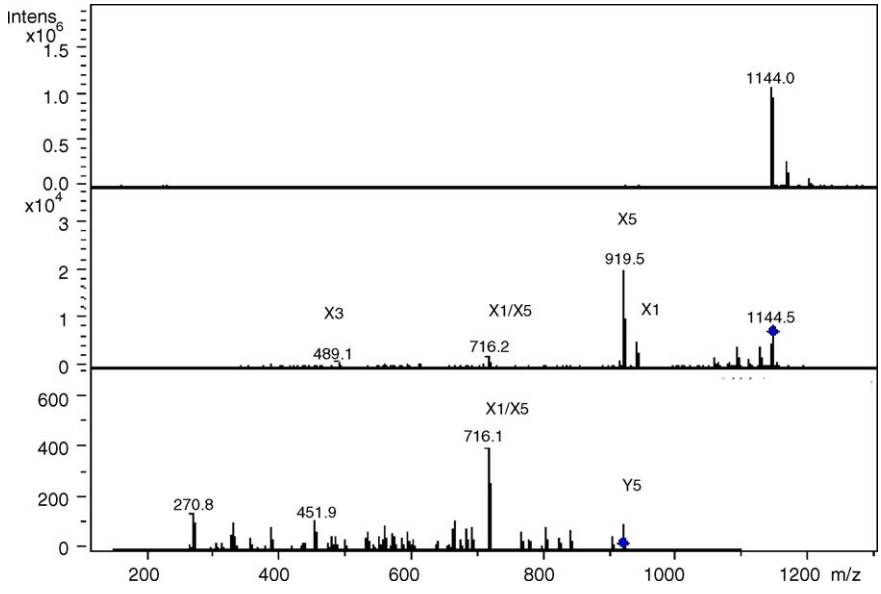


Fig. 2. The MS and MS^n spectra of compound **5**.

Although the polyamide/peptide conjugates **2** and **3** are structurally similar to each other, they have different fragmentation pathways. The precursor ion *m*/*z* 737 of compound **3** produced fragment ions *m*/*z* 650 by the *b* cleavage. However, the *b* cleavage product *m*/*z* 650 was not observed in the MS/MS spectra of $[M+H]^+$ ions of compound **2**. The compounds **3** and **5** are structurally similar also. But they follow some different fragmentation pathways. Because it contains more polyamide structure, compound **5** has many more cleavage positions such as fragment ions X_1 and X_2 .The MS and MS^n spectra of compound **5** is shown in Fig. 2. Fragment ions X_1, Y_5, X_1/Y_5, X_3 is due to CO–NH bond cleavage.

4. Conclusion

In this study, the ESI-MS spectra of polyamide/peptide conjugates provide abundant structural information. The main fragmentation pathways involve the cleavage of the C–CO between rings and carbonyl groups (*a* cleavage), CO–NH amide bonds (*b* cleavage). The ESI-MS approach has proven to be an excellent method for the structural elucidation of this class of polyamide/peptide conjugates known to specifically cleave DNA.

Acknowledgement

We thank the National Natural Science Foundation of China (No. 20572061).

References

[1] W.S. Wade, M.M. Mrksich, P.B. Dervan, J. Am. Chem. Soc. 114 (1992) 8783.
[2] J.W. Trauger, E.E. Baird, P.B. Dervan, Nature 382 (1996) 559.
[3] S. White, E.E. Baird, P.B. Dervan, Chem. Biol. 4 (1997) 569.
[4] A. Blasko, K.A. Browne, T.C. Bruice, J. Am. Chem. Soc. 116 (1994) 3726.
[5] J.G. Pelton, D.E. Wemmer, Biochemistry 27 (1988) 8808.
[6] C.L. Kielkopf, S.E. White, J.W. Szewczyk, et al., Science 282 (1998) 111.
[7] Q. Zhang, T.J. Dwyer, V. Tsui, et al., J. Am. Chem. Soc. 126 (2004) 7958.
[8] M.R. Burns, C.L. Carlson, S.M. Vanderwerf, et al., J. Med. Chem. 44 (2001) 3632.
[9] I.S. Blagbrough, E. Moya, Tetrahedron Lett. 36 (1995) 9393.
[10] M.R. Ashton, E. Moya, I.S. Blagbrough, Tetrahedron Lett. 36 (1995) 9397.
[11] M.M. Becker, P.B. Dervan, J. Am. Chem. Soc. 101 (1979) 3664.
[12] P.B. Dervan, M.M. Becker, J. Am. Chem. Soc. 100 (1978) 1968.
[13] I.S. Blagbrough, E. Moya, S.P. Walford, Tetrahedron Lett. 37 (1996) 551.
[14] E. Moya, I.S. Blagbrough, Tetrahedron Lett. 36 (1995) 9401.
[15] Y.S. Li, Y.F. Zhao, et al., Bioorg. Med. Chem. 8 (2000) 2675.
[16] J. Chen, R. Wan, et al., Lett. Pept. Sci. 7 (2001) 325.
[17] Y. Li, S. Hatfield, J. Li, M. McMills, Y. Zhao, X. Chen, Bioorg. Med. Chem. 10 (2002) 667.
[18] R. Wan, N. Wang, G. Zhao, Y. Zhao, Chem. J. Chin. Univ. 21 (2000) 1864.
[19] X. Li, Y.F. Zhao, Chin. Chem. Lett. 8 (1997) 611.
[20] Y. Ye, M.Y. Niu, Q. Yin, L.F. Cao, Y.F. Zhao, J. Chem. Res. S 4 (2005) 252.