ESI-MS study on the aldol reaction catalyzed by L-proline†

Cesar Marquez and Jürgen O. Metzger*

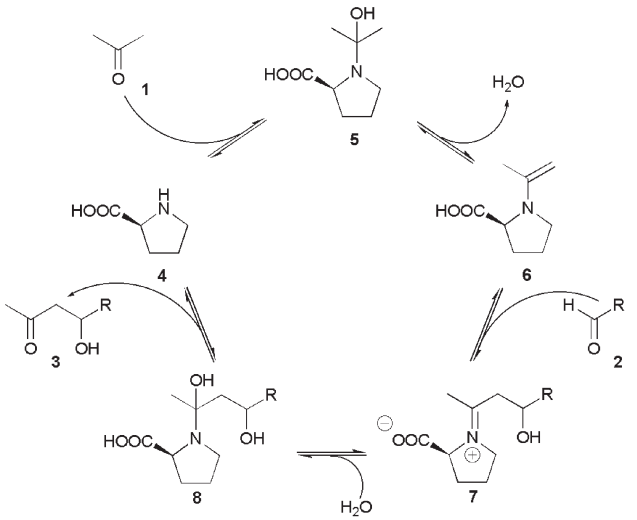
Received (in Cambridge, UK) 4th January 2006, Accepted 14th February 2006
First published as an Advance Article on the web 28th February 2006
DOI: 10.1039/b518288c

The aldol reaction catalyzed by L-proline has been monitored and the accepted mechanism confirmed by intercepting and characterizing, for the first time, all the proposed intermediates by ESI-MS/MS.

Certain small metal-free organic compounds are able to catalyze organic reactions in a fashion similar to enzymes or transition metals. As clear advantages, these compounds tend to be less expensive, easier to obtain and do not require the specific reaction conditions of the enzymes. At the same time, their chemistry is cleaner than that of the transition metals. These factors have provoked an increased interest in the scientific community in organocatalytic processes and their reaction mechanisms.[1]

Electrospray ionization (ESI)[2,3] is an interesting technique of mass spectrometry (MS), as both positive and negative ions formed in solution can be transferred directly to the gas phase.[4] Due to a variety of distinctive characteristics, ESI-MS and its tandem version ESI-MS/MS are rapidly becoming suitable tools for the detection and mass spectrometric characterization of reaction intermediates directly from solution,[5–9] providing advances in mechanistic studies in chemistry and biochemistry[10] and in high throughput screening of homogeneous catalysis reactions.[11] Recently, Santos *et al.* reported their ESI-MS studies of the organocatalytic Baylis–Hillman reaction in which most intermediates could be intercepted as protonated species and characterized by ESI-MS/MS.[9]

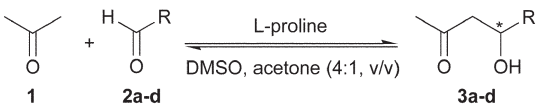
The aldol reaction is recognized as a powerful tool for the construction of new carbon–carbon bonds,[12] and its organocatalysis has been studied thoroughly.[13–18] However, it is noteworthy that, to the best of our knowledge, the intermediates have not been unequivocally intercepted and characterized. The catalytic cycle currently accepted for this reaction has been proposed to proceed *via* the formation of an adduct intermediate **5** between the L-proline and the acetone **1**, producing an enamine **6** by elimination of water (Scheme 1).[14–16,19–22] The enamine **6** interacts with the aldehyde **2** forming adduct **7**. Addition of water to **7** produces a transient **8** that dissociates to yield the aldol product **3**, leaving the L-proline free to continue with the cycle. As the aldol reaction is a reversible process, all intermediates **5–8** are in principle present in the equilibrated solution, and should be detectable by MS as protonated and/or cationized ions.



Scheme 1 Proposed mechanism of the L-proline-catalyzed aldol reaction.

We studied the reaction of acetone **1** and the benzaldehydes **2a–d** to form the aldols **3a–d** catalyzed by L-proline **4** (Scheme 2) in accordance with the protocol of List *et al.*,[14] focusing on the direct MS detection and characterization of the intermediates involved using a microreactor coupled on-line to the ESI mass spectrometer.[6,7]

The reaction solution was fed continuously into the mass spectrometer through a mixing tee that allowed the dilution of the mixture with methanol (50 : 1) before entry to the ionization source. The mass spectrum of the reaction solution using benzaldehyde **2a** after a reaction time of one hour is provided in Fig. 1. The signals *m*/*z* 156 and *m*/*z* 178 corresponding to the cationized enamine **6**·$\mathbf{H}^+$ (*m*/*z* 156) and **6**·$\mathbf{Na}^+$ (*m*/*z* 178) and/or the isomeric iminium ion and oxazolidinone,[20,22] respectively, being the most intensive peaks, and the signals of substrate **2a**·$\mathbf{Na}^+$ (*m*/*z* 186) and of product **3a**·$\mathbf{Na}^+$ (*m*/*z* 244), are clearly recognizable. All ions were characterized by high-accuracy mass spectrometry. The signals of the intermediates **5**, **7a** and **8a** are expected to have a much lower concentration and will appear in the chemical noise.



a: R=(*p*-NHCOCH$_3$)-C$_6$H$_4$; b: R=(*p*-NO$_2$)-C$_6$H$_4$; c: R=(*o*-Cl)-C$_6$H$_4$; d: R=*i*Pr.

Scheme 2 Aldol reaction of acetone **1** and aldehydes **2a–d** catalyzed by L-proline to give aldol **3a–d**.

Institut für Reine und Angewandte Chemie, Carl von Ossietzky Universität, 26111, Oldenburg, Germany.
E-mail: Juergen.Metzger@uni-oldenburg.de; Fax: +49441 798 3329; Tel: +49441 798 3718

† Electronic supplementary information (ESI) available: Experimental procedures and MS/MS spectra for intermediates in the catalytic cycle. See DOI: 10.1039/b518288c

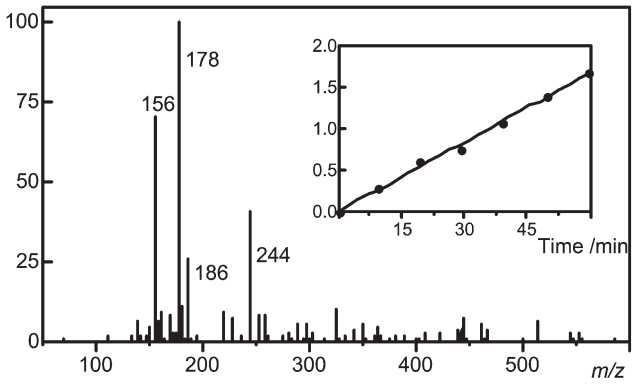


Fig. 1 ESI-MS spectrum of the reaction solution of acetone **1** and aldehyde **2a** after 60 min: **6**·H^+ (*m*/*z* 156), **6**·Na^+ (*m*/*z* 178), **2a**·Na^+ (*m*/*z* 186), **3a**·Na^+ (*m*/*z* 244). Ordinate: relative intensity. Shown in the insert is the evolution over time of the ratio between **3a**·Na^+ (*m*/*z* 244) and **2a**·Na^+ (*m*/*z* 186).

However, these intermediates could be detected using the MS/MS technique to filter out the signal of interest from baseline noise.

It was possible to follow the reaction over time by monitoring the decrease of the intensity of the signal of **2a** and the increase of that of **3a** (see insert in Fig. 1). The conversion of **2a** is approximately 65% after one hour of reaction, assuming the relation signal intensity *versus* concentration in solution is similar for both **2a** and **3a**.

The formation of the species giving signals *m*/*z* 156 and *m*/*z* 178 is a fast reaction. When a solution of L-proline **4** in methanol is mixed with acetone in continuous-flow this species is formed in less than 6 s. The ratio of these signals and **4**·H^+ (*m*/*z* 116) varies with the concentration of the ketone in the medium. Using a ratio of methanol/acetone 4 : 1 v/v, they were ten times stronger than that of the L-proline **4**·H^+. The fragmentation pattern observed for the signal *m*/*z* 156 formed under continuous-flow MS/MS (Fig. 2b) is coincident with that observed in the reaction medium at different times. It consists mainly in the elimination of formic acid of 46 u giving the fragment ion of *m*/*z* 110. This fragmentation is clearly expected for the protonated enamine **6**·H^+, as it is the only one observed in the MS/MS of the protonated L-proline **4**·H^+ producing the fragment with *m*/*z* 70.[23]

The protonated intermediate **5**·H^+ (*m*/*z* 174) was detected in the reaction solution after 6 s and was characterized by MS/MS (Fig. 2a). It fragments to produce **4**·H^+ and **6**·H^+. Notice that the latest fragment differentiates this species from a non-covalent complex of L-proline with acetone.

The intermediates **7** in the reaction of preformed **6** with the different aldehydes **2a–d** were also intercepted using continuous-flow methodology after only 2 s reaction time. When **2a** was used, it was possible to intercept the transient **7a** as ion **7a**·H^+ (*m*/*z* 319) and **7a**·Na^+ (*m*/*z* 341), respectively. The main fragmentations of **7a**·H^+ were those yielding **4**·H^+ and **6**·H^+ (Fig. 2c).

Finally, it was possible to intercept the adduct ion **8a**·H^+ (*m*/*z* 337) and **8a**·Na^+ formed by the addition of water to **7a** in the ongoing reaction. The intermediate **8a**·H^+ fragmented to **4**·H^+ of *m*/*z* 116 and neutral **3a** without applying collision energy. Additionally, cleavage of 64 u (HCOOH + H_2O) and 82 u (HCOOH + 2 H_2O) was observed (Fig. 2d). Studying ion **8c**·H^+, the major fragmentation was the one that produced **4**·H^+ and neutral **3c**. Ion **8a**·Na^+

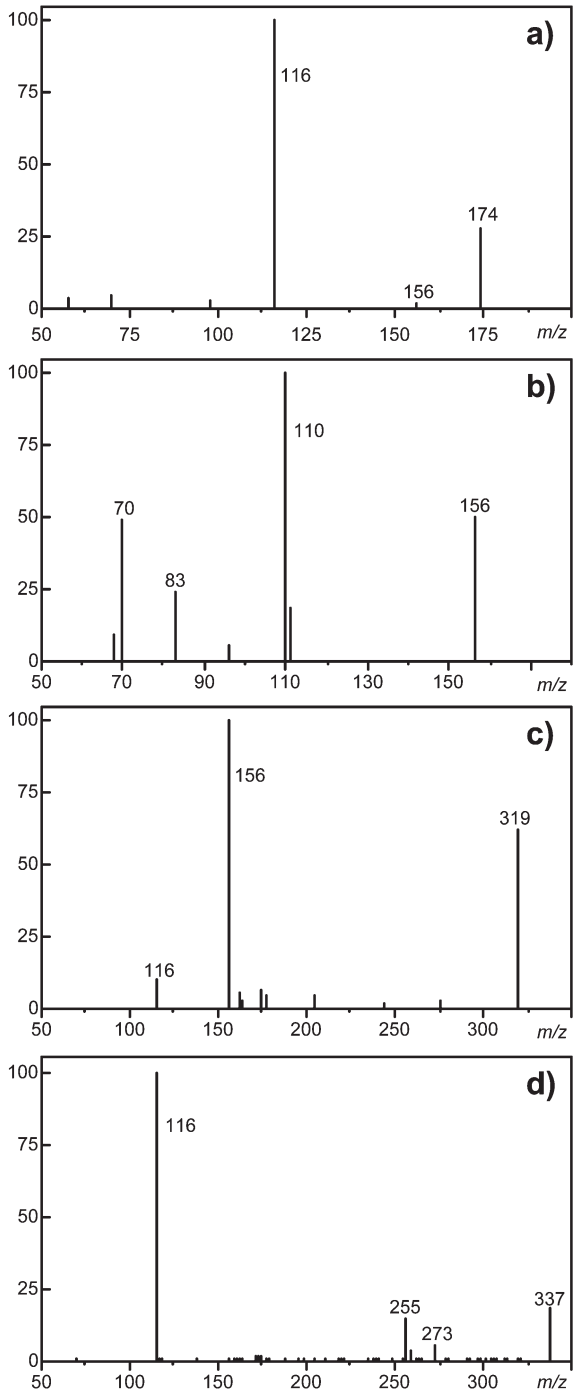


Fig. 2 ESI-MS/MS spectra of the intermediates a) **5**·H^+, b) **6**·H^+, c) **7a**·H^+, d) **8a**·H^+ (see Scheme 1). Ordinate: relative intensity.

showed the same fragmentation to give product **3a** and proline **4**. In contrast to **8a**·H^+ the ion **3a**·Na^+ (*m*/*z* 244) was observed. Clearly, **3a** is better able to chelate Na^+ than proline **4**, whereas **4** is more basic and will be protonated in the fragmentation of **8a**·H^+. Interestingly, a loss of water was clearly observed for **8b**·Na^+ (*m*/*z* 347), yielding a fragment ion of *m*/*z* 329.

In summary, the study by ESI-MS of the aldol reaction of acetone with aldehydes **2a–d** catalyzed by L-proline has allowed us not only to monitor the reaction evolution over time, but most interestingly, to characterize all the intermediates assumed for the

catalytic cycle. The presumed enamine **6** formation has been unequivocally confirmed by high-resolution measurement and MS/MS. The formerly discussed parasitic bicyclic oxazolidinone from acetone and L-proline was not observed.[20] Only the oxazolidinone of aldehyde **2d** could be detected in the reaction solution.[23] The three additional transients proposed in the aldol reaction including the L-proline/acetone adduct **5**, the enamine/aldehyde adduct **7** and the hydrated form of the latter **8** have been intercepted and characterized by MS/MS, confirming the mechanism currently accepted for this catalytic cycle.

We thank the German Research Association (DFG) for financial support of this project (SSP 1179).

Notes and references

1 A. Berkessel and H. Gröger, *Asymmetric Organocatalysis: From Biomimetic Concepts to Applications in Asymmetric Synthesis*, Wiley-VCH, London, 2005.
2 J. B. Fenn, M. Mann, C. K. Meng, S. F. Wong and C. M. Whitehouse, *Science*, 1989, **246**, 64.
3 C. M. Whitehouse, R. N. Dreyer, M. Yamashita and J. B. Fenn, *Anal. Chem.*, 1985, **57**, 675.
4 J. F. de la Mora, G. J. Van Berkel, C. G. Enke, R. B. Cole, M. Martinez-Sanchez and J. B. Fenn, *J. Mass Spectrom.*, 2000, **35**, 939.
5 (*a*) J. Griep-Raming, S. Meyer, T. Bruhn and J. O. Metzger, *Angew. Chem., Int. Ed.*, 2002, **41**, 2738; (*b*) S. Furmeier, J. Griep-Raming, A. Hayen and J. O. Metzger, *Chem.-Eur. J.*, 2005, **11**, 5545.
6 (*a*) S. Meyer and J. O. Metzger, *Anal. Bioanal. Chem.*, 2003, **377**, 1108; (*b*) S. Meyer and J. O. Metzger, *J. Am. Chem. Soc.*, 2004, **126**, 14485.
7 (*a*) X. Zhang, Y. Liao, R. Qian, H. Wang and Y. Guo, *Org. Lett.*, 2005, **7**, 3877; (*b*) R. Qian, H. Guo, Y. Liao, Y. Guo and S. Ma, *Angew. Chem., Int. Ed.*, 2005, **44**, 4771.
8 C. Trage, D. Schröder and H. Schwarz, *Chem.-Eur. J.*, 2005, **11**, 619.
9 L. S. Santos, C. H. Pavam, W. P. Almeida, F. Coelho and M. N. Eberlin, *Angew. Chem., Int. Ed.*, 2004, **43**, 4330.
10 L. S. Santos, L. Knaack and J. O. Metzger, *Int. J. Mass Spectrom.*, 2005, **246**, 84.
11 X. Y. Chen, X. Y. Zhang and P. Chen, *Angew. Chem., Int. Ed.*, 2003, **42**, 3798.
12 C. Palomo, M. Oiarbide and J. M. Garcia, *Chem. Soc. Rev.*, 2004, **33**, 65.
13 K. Sakthivel, W. Notz, T. Bui and C. F. Barbas, *J. Am. Chem. Soc.*, 2001, **123**, 5260.
14 B. List, R. A. Lerner and C. F. Barbas, *J. Am. Chem. Soc.*, 2000, **122**, 2395.
15 F. R. Clemente and K. N. Houk, *Angew. Chem., Int. Ed.*, 2004, **43**, 5766.
16 C. Allemann, R. Gordillo, F. R. Clemente, P. H. Y. Cheong and K. N. Houk, *Acc. Chem. Res.*, 2004, **37**, 558.
17 B. List, P. Pojarliev and C. Castello, *Org. Lett.*, 2001, **3**, 573.
18 W. Notz and B. List, *J. Am. Chem. Soc.*, 2000, **122**, 7386.
19 T. D. Machajewski and C. H. Wong, *Angew. Chem., Int. Ed.*, 2000, **39**, 1352.
20 B. List, L. Hoang and H. J. Martin, *Proc. Natl. Acad. Sci. USA*, 2004, **101**, 5839.
21 L. Hoang, S. Bahmanyar, K. N. Houk and B. List, *J. Am. Chem. Soc.*, 2003, **125**, 16.
22 A. Hartikka and P. I. Arvidsson, *Eur. J. Org. Chem.*, 2005, 4287.
23 The fragmentation pattern observed allows us to differentiate unambiguously the enamine $\mathbf{6}\cdot\mathbf{H}^+$ from the isomeric iminium ion and oxazolidinone in solution. A detailed discussion can be found in the supporting information.