ChemComm

The TpZn–OH/CS_2 reaction: theoretical and preparative visualization of an essential bioinorganic reaction path

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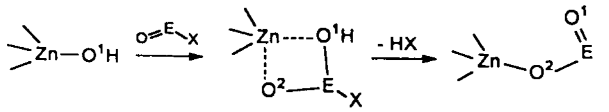
Received (in Basel, Switzerland) 10th February 2000, Accepted 7th March 2000
Published on the Web 27th March 2000

The reaction between L_3Zn–OH complexes (L = N_3 based ligand) and CS_2 which is an analogue of the carbonic anhydrase functionality was computed at the B3LYP/6-311 + G* level to proceed *via* stable four-center intermediates [L_3Zn–SC(S)OH or L_3Zn–SC(O)SH] to give L_3Zn–SH and COS; in agreement with these calculations, the chemical reaction of $Tp^{Ph,Me}$Zn–OH with CS_2 resulted in the quantitative formation of $Tp^{Ph,Me}$Zn–SH and COS; in the presence of 1 equivalent of MeOH the reaction yielded $Tp^{Ph,Me}$Zn–SC(S)OMe, thus also supporting the existence of the four-center intermediate L_3Zn–SC(S)OH.

It is now generally accepted that hydrolytic zinc enzymes (carbonic anhydrase, peptidases, phosphatases, esterases, nucleases) owe their high efficiency to their double functionality, in that they allow electrophilic activation by metal coordination to a substrate oxygen (C=O, P=O) and they provide the attacking nucleophile in the form of the Zn–OH unit.[1] When only one zinc ion is present as in carbonic anhydrase or in the many matrix metalloproteases it performs both functions. This means that during the catalytic turnover zinc changes its coordination number from four to five and then back to four, and the essential intermediate for the general case of O=E–X [E = CR, $P(OR)_2$; X = OR, NHR] hydrolysis is a four-center species like **A**. Several computational studies have considered the details of this process,[2–4] and one of us has recently published structural evidence for the corresponding reaction trajectory.[5]

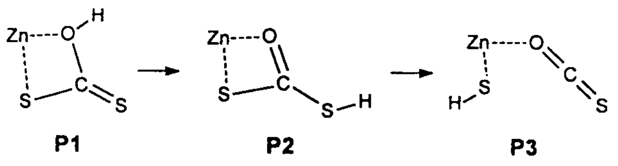


A

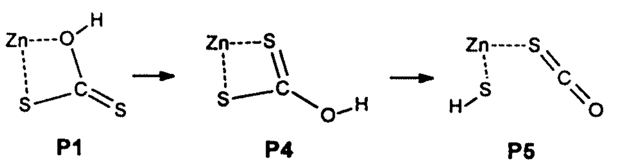
While one would assume that in passing through the intermediate **A**, the Zn–O¹ bond is broken and the Zn–O² bond is formed, *i.e.* hydroxide is the leaving group, experimental proof for this reaction course is still missing. Major obstacles to discerning O¹ and O² by isotopic labelling lie in the mobility of the OH proton, in the reversibility of the reactions, and in the high lability of zinc complexes, which for instance leads to rapid scrambling of ^{17}O from $C^{17}O_2$ over all positions in the TpZn–OH/CO_2 system [Tp = tris(pyrazolyl)borate].[6] It was therefore attractive to investigate a reaction system where homologous substitution, *i.e.* of O by S, renders distinguishable the atoms in question. We chose for this purpose the L_3Zn–OH/CS_2 system (L = N based ligand) based on previous experience in both the computational treatment of the L_3Zn–OH/CO_2 system[7] and reactivity studies of pyrazolylborate complexes TpZn–OH with CO_2 and similar heterocumulenes.[8] This has enabled us to present a case where the computational results came first and were only subsequently verified by the experiments.

The quantum chemical investigation on the B3LYP/6-311 + G* level of theory was carried out for the potential energy surface of the reaction system comprised of naked $[Zn–OH]^+$ and CS_2 (*cf.* Schemes 1 and 2). It confirmed what might have been expected from thermodynamic considerations, namely that the global energy minimum corresponds to $[Zn–SH]^+$ and COS, and it gave some insight into possible reaction mechanisms leading there. Scheme 1 presents the relevant intermediates of a reaction course which corresponds to the Lipscomb mechanism[9] of carbonic anhydrase action. It consists of subsequent proton transfers and implies that the oxygen of Zn–OH becomes part of O-bound COS (**P3**) which is then expelled. Scheme 2 shows a reaction sequence according to the Lindskog mechanism,[10] the essence of which is an internal rotation of the coordinated dithiocarbonate (**P1**) from the O,S- to the S,S-bound state without a proton transfer. A subsequent proton transfer then yields Zn–SH and COS which is now S-bound (**P5**) before it is expelled.

Our calculations have shown that both pathways are feasible because their steps have low to moderate activation barriers, and referred to the separated reactants the overall activation barrier is +2.4 (Scheme 2) or −3.9 (Scheme 1) kcal mol^{-1}. In order to corroborate the results obtained for this very simple model system we recalculated all minima of the potential energy surface for the two well accepted model systems $[(NH_3)_3ZnOH]^+$ and $[(Him)_3ZnOH]^+$ (Him = imidazole). A comparison of the relative energies of all three model complexes is given in Table 1. As expected, the inclusion of a complete ligand sphere levels the energy differences and yields no minimum for species like **P3** or **P5**. As compared with the results for the $[ZnOH]^+$/CS_2 system, the relative energies of the **P2**/**P4** intermediates change to a marked preference for the **P2** structures. Furthermore **P2** is now similar in energy to the separated reaction products L_3ZnSH and COS, as is **P1** to **P4**, supporting the notation that four-center intermediates like **P1**,



Scheme 1



Scheme 2

DOI: 10.1039/b001149p

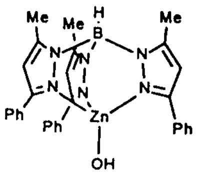
Table 1 Relative energies of the minima for the reaction of the three model complexes with CS_2 (energies in kcal mol^{-1})

	P1	**P2**	**P4**	Product(s)
$[ZnOH]^{+}$[a]	−28.49	−41.65	−46.87	−72.47/−69.90[c]
$[(NH_3)_3ZnOH]^{+}$[a]	−5.36	−20.61	−10.86	−21.82[d]
$[(Him)_3ZnOH]^{+}$[b]	−6.61	−20.84	−6.68	−24.80[d]

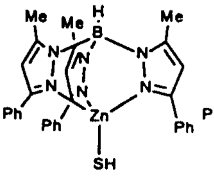
[a] B3LYP/6-311 + G*//B3LYP/6-311 + G*. [b] B3LYP/6-311 + G*//HF/6-311 + G*; Him = imidazole. [c] **P3**/**P5**. [d] L_3ZnSH + free COS.

P2 and **P4** are relevant entities in the reaction course for the CS_2 just like for the CO_2 reaction system.

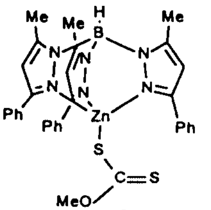
The chemical investigation of this reaction system, carried out with the phenyl-substituted TpZn–OH complex **1**, resulted in full agreement between the calculated and observed reaction. In water-free dichloromethane **1** and a stoichiometric amount of CS_2 were converted quantitatively to the TpZn–SH complex **2** and COS within 12 h at room temperature.† The analysis of the gaseous reaction products† revealed that CO_2 had formed in addition to COS. This pointed to the fact that COS is capable of undergoing the same interconversion, as was verified subsequently by reacting **1** with COS.



1

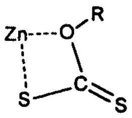


2

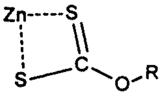


3

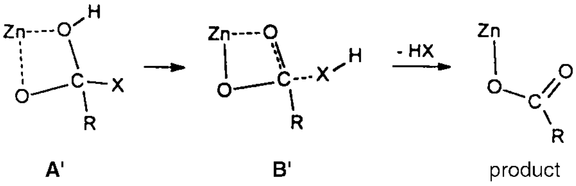
An indication of possible reaction intermediates was found when MeOH was present accidentally in the reaction system: a 1:1:1 mixture of **1**, CS_2 and MeOH then yielded the xanthogenate complex **3**, again quantitatively.† The formation of **3** can be understood as a CS_2 addition to the methoxide complex TpZn–OMe (which can exist in very small quantities in equilibrium with **1** and methanol[11]), in analogy to the formation of alkali metal xanthogenates from alkali and CS_2. This proposal was verified by reacting TpZn–OMe, the methoxide derivative of **1**,[12] with CS_2 in dichloromethane, which produced **3** fast and quantitatively.† The corresponding intermediate, in analogy to **P1**, should be **P1a**. Alternatively, intermediate **P4** might be esterified with MeOH, yielding an intermediate **P4a**. We favor the insertion reaction *via* **P1a**, because we see no reason why **P4** should be esterified faster than being interconverted to **P5**. On the other hand, and in agreement with our calculations,[13] the methoxide in TpZn–OMe should be a stronger nucleophile than the hydroxide in TpZn–OH and hence capable of outracing the latter in the reaction with CS_2 even when present in much smaller concentrations. We have observed similar alcohol incorporations before when reacting the cumenyl substituted TpZn–OH with CO_2 and CS_2.[8]



P1a



P4a



Scheme 3

The considerations applied and verified here for the CO_2/CS_2 system are applicable in the same way to the hydrolytic cleavage reactions of O=C(R)–X with intermediate **A** (Scheme 3). **A′** corresponds to **P1**, a conceivable proton transfer to X creates a possible intermediate **B′** corresponding to **P2**. Thereafter the course of events is different: HX is eliminated while HS in **P3** stays zinc-bound, and while OCS in **P3** leaves, its equivalent RCOO in **B′** becomes part of the reaction product, implying that the Zn–O^1 bond in **A** is broken. If one believes in the wisdom '*natura non facit saltus*' this means that the general pathway for all zinc-catalyzed hydrolytic reactions follows the mechanism in Scheme 1, *i.e.* the Lipscomb mechanism.

This work was supported by the Deutsche Forschungsgemeinschaft (Va 29/15 and GRK 307, Freiburg, and SFB 436, Jena) and by the Fonds der Chemischen Industrie. Financial support by the Thüringer Ministerium für Wissenschaft, Forschung und Kultur is gratefully acknowledged. Furthermore, we thank the Konrad-Zuse-Zentrum für Informationstechnik Berlin for generously providing us with computing time.

Notes and references

† The new complexes were characterized by elemental analyses, IR and NMR spectra in comparison with reference compounds.[8] COS and CO_2 were identified by GC–MS. An X-ray structure determination was carried out for **2**.

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