BERGISCHE UNIVERSITÄT WUPPERTAL

Physical & Theoretical Chemistry

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Non target TOF-analysis of oxidation products of aromatic amino acids with chlorine dioxide as potential marker of cell degradation

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Introduction

Chlorine dioxide is a widely used chemical for disinfection purposes applied mostly in water. The mechanisms of how it interacts with organisms are plentiful. That leads to complex interactions with the proteome. There the most reactive amino acids seem to be the aromatic ones [1]. This raises the question if some of them can be used as markers for reaction between chlorine dioxide and biological matter.
Bacteria seem to "explode" after the interaction with chlorine dioxide, as such particular interest is put on the membrane of the organisms (**Figure 2, 4**). However, technically used chlorine dioxide is mostly used as a chemical and not as a well-defined solution out of many highly oxidative chlorine species. Therefore, there is the need to look only on the behavior of chlorine dioxide.

Methods

Chlorine dioxide is synthesized by the mixing of sodium chlorite and sodium peroxydisulfate. This method is equal to the commercially available product *Clorious2* distributed by Brenntag AG. The released gas is then stripped trough Millipore water. The concentration is determined by photometric measurements. For each measurement, the chlorine dioxide concentration is 0.05 mg/l. Only the concentration of the amino acid changes. This concentration is the highest legal concentration in drinking water in Germany.
The reaction time is 15 minutes. In some cases, the solutions got a light red shade. After the reaction time the solutions were mixed with acetonitrile and formic acid producing a mixture of 50:50:0.1; Water:ACN:FA. This was then measured with a flowrate of 7 µl/min with ESI and a Bruker micrOTOF.

Conclusion

For tyrosine no products could be found (**Figure 6**). But interestingly the whole intensity is way higher with added chlorine dioxide solution than without. That leads to the conclusion, that the processes in the ion source need to be further investigated.
For tryptophane all expected m/z-ratios were found (**Figure 5**) [2]:
Furthermore, an analysis of the better researched products of tryptophane shows that there should be a bias in the protonation process towards the products with an intact indole system like DIA and OIA (**Figure 4**).
It can thus be established that tryptophane is much more likely to get oxidated than tyrosine and thus seems to be a valid marker for oxidations with chlorine dioxide. Further research should be conducted using histidine.

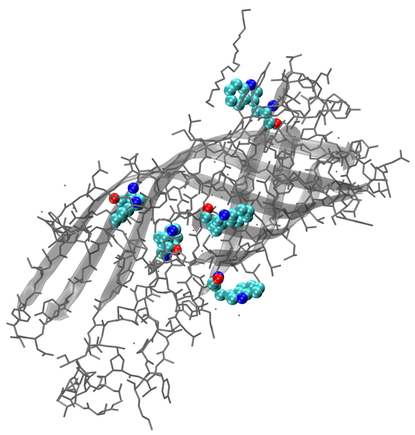
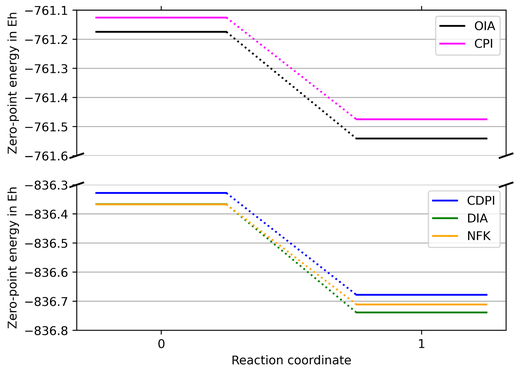
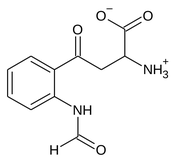
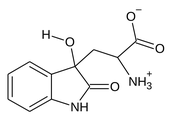


Figure 1, right: Schematic of the effect of chlorine dioxide
Figure 2, top left: Structure of OmpA membrane protein; tryptophane molecules as explicit structures.
Figure 3, bottom left: Zero-point energies of the oxidation products of tryptophane as soluted substance and protonated species
Figure 4, bottom: Schematic of a bacteria membrane with OmpA, tryptophane molecules in red

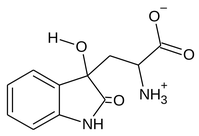




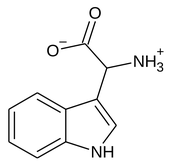
N-formylkynurenine (NFK)
MW = 236.22



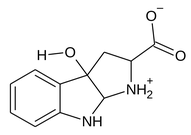
Dioxindolylalanine (Dia)
MW=236.22



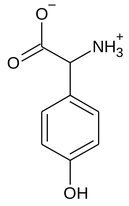
Oxindolylalanin (Oia)
MW = 220.22



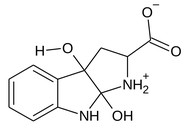
Tryptophan (Trp)
MW=204.23



2-carboxy-3a-hydroxy-1,2,3,3a,8,8a-hexahydropyrrollol[2,3-b]indole (CPI)
MW = 220.22



Tyrosin (Tyr)
MW=181.19



2-carboxy-3a,8a-dihydroxy-1,2,3,3a,8,8a-hexahydropyrollo[2,3b]indole(CDPI)
MW = 236.22

Computational Methods

All structures were preoptimized with the CREST toolset. Afterwards the protonated species get protonated with CREST [4]. The energy optimizations are done with Psi4 with B3LYP/631++G [5].

References

[1] Navalon S, Alvaro M, Garcia H. Chlorine dioxide reaction with selected amino acids in water. J Hazard Mater. 2009 May 30;164(2-3):1089-97.
[2] Stewart DJ, Napolitano MJ, Bakhmutova-Albert EV, Margerum DW. Kinetics and mechanisms of chlorine dioxide oxidation of tryptophan. Inorg Chem. 2008 Mar 3;47(5):1639-47.
[3] Pautsch A, Schulz GE. Structure of the outer membrane protein A transmembrane domain. Nat Struct Biol. 1998 Nov;5(11):1013-7
[4] Pracht, P.; Bohle, F.; Grimme, S.; Automated exploration of the low-energy chemical space with fast quantum chemical methods, Phys. Chem. Chem. Phys., 2020, 22, 7169-7192.
[5] Janesko BG. Systematically Improvable Generalization of Self-Interaction Corrected Density Functional Theory. J Phys Chem Lett. 2022 Jun 30;13(25)

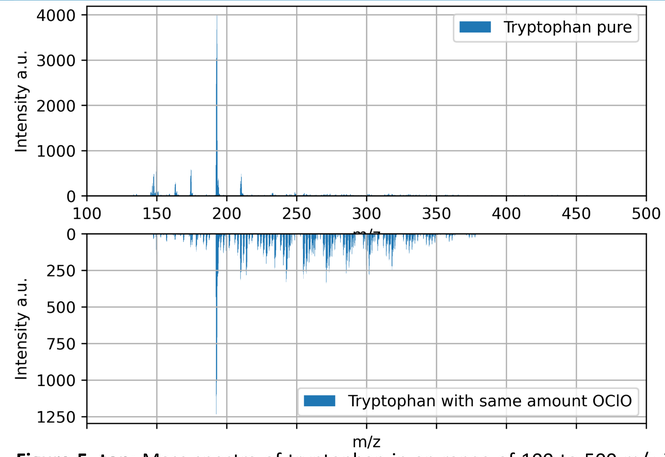


Figure 5, top: Mass spectra of tryptophan in an range of 100 to 500 m/z in an concentration of 0.7 mmol/l; chlorine dioxide in the same concentration
Figure 6, bottom: Mass spectra of tyrosin in an range of 100 to 500 m/z in an concentration of 0.7 mmol/l; chlorine dioxide in the same concentration