Reply to the 'Comment on "Investigation of Zr(IV) and 89 Zr(IV) complexation with hydroxamates: progress towards designing a better chelator than desferrioxamine B for immuno-PET imaging"' by A. Bianchi and M. Savastano, Chem. Commun., 2020, 56, D0CC01189D

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COMMUNICATION

Reply to the 'Comment on "Investigation of Zr(IV) and ^{89}Zr(IV) complexation with hydroxamates: progress towards designing a better chelator than desferrioxamine B for immuno-PET imaging"' by A. Bianchi and M. Savastano, Chem. Comm 2020, 56, D0CC01189D.

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Abstract. The alternative analysis of A. Bianchi and M. Savastano is a valuable contribution to the understanding of the complex systems at stake in the complexation chemistry of Zr^{4+} by considering polynuclear species. Placed in the context of nuclear medicine where such aggregates are unlikely and considering recent literature data, this however points out that no clear agreement exists to describe such complex formation.

We read with attention the comment article by A. Bianchi and M. Savastano[1] in which they revisited the potentiometric study of the complexation of Zr^{4+} by hydroxamate ligands as we reported on in 2013.[2] Our communication was then, to our knowledge, the first reporting on structural and thermodynamic data of **Zr^{4+}** complexes relevant to nuclear medicine, and one of the few studies on the complexation of Zr^{4+} in aqueous medium. Available data from the literature was very scarce and often incomplete, when considering the purpose of our study, which limited comparisons that are now more possible to achieve. Guided by complimentary data we obtained by X-ray crystallographic studies, DFT calculations, and especially results from radiolabeling experiments with ^{89}Zr, we proposed an analysis of potentiometric data that was certainly simpler than in reality. Nonetheless, the main conclusions were shortly later corroborated by a number of reports which demonstrated the significantly higher stability of complexes formed with ligands comprised of four hydroxamate groups compared to only three in DFO.[3–8]

As potentiometry is a technique based on the fitting of experimental curves and various models, a large number of combinations is possible, even with a rigorous application of known parameters. The alternative interpretation of Bianchi and Savastano is based on the larger predominance of hydroxylated species and the non-negligible existence of polynuclear species such as $[Zr_3(AHA)_3OH_7]^{2+}$ and $[Zr_3(AHA)_3OH_8]^{+}$). We acknowledge that the proposed combination in fitting resolves the unusual trend in equilibrium constant we reported and that this new interpretation is probably a good improvement for the study of zirconium(IV) complexation with this ligand. Nonetheless, we would like to point out that in the context of nuclear medicine, it should be noted that polynuclear species implying Zr^{4+} as the only metal are much less likely to occur than in potentiometric titration conditions. As a matter of fact, radiolabeling chemistry implies the use of ^{89}Zr at trace concentrations, several hundreds of times lower than the 1 mM concentration used by Bianchi **and** Savastano and us for potentiometric titrations. In addition, the presence of metallic impurities such as Fe^{3+} that may compete in the complexation reaction must also be kept in mind. Considering a ^{89}Zr effective molar activity of 37 MBq/nmol reported for current production processes,[9] effective Zr concentration can be estimated to ≈ 2 µM in our study discussed herein,[2] which is in the range of the concentrations used in the typical procedure reported for radiolabeling monoclonal antibodies for human use (from 0.5 to 2.5 µM).[10,11] Thus, application of speciation diagrams should be handled with due consideration when translation to the context of trace concentrations, such as in radiolabelling conditions, is considered. Similar divergent analyses can be found in a closely related context, with the recent work of the same authors on the complexation of Zr^{4+} by DFO,[12] that was published almost simultaneously by Toporivska and Gumienna-Kontecka.[13] While Bianchi and co-authors proposed a quite complex speciation diagram, including polynuclear species, only mononuclear species were described in the second study. This result points out there is no clear consensus on how to treat the data and fit speciation curves while also maintaining a link to concrete applications such as nuclear medicine.

In conclusion, it is indisputable that the new analysis proposed by Bianchi and M. Savastano provides an improvement over our previous proposal and that the new equilibrium constants will contribute to the development in this growing field of research. This, however, does not alter the main conclusions of our original communication. It is now admitted that hydroxamate ligands are reference compounds for the complexation of ^{89}Zr and we are

excited to observe that a growing number of research groups from various specialty have focused their efforts in the study of this complex chemistry since our pioneer works.

Conflicts of interest

There are no conflicts to declare.

Notes and references

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