Electronic Supplementary Information

Comment on "Investigation of Zr(IV) and 89Zr(IV) complexation with hydroxamates: progress towards designing a better chelator than desferrioxamine B for immuno-PET imaging" by F. Guérard, Y.-S. Lee, R. Tripier, L. P. Szajek, J. R. Deschamps, M. W. Brechbiel, Chem. Commun. 2013, 49, 1002.

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Potentiometric measurements

Preparation of Zr(IV) stock solution

Commercial $ZrCl_4$ (5 g, 99.99%, Sigma-Aldrich) was dissolved in 200 ml 0.1 M HCl aqueous solution: such manipulation has been done carefully into a glove box.

The obtained stock solution (0.1 M approx.) has been standardized against 1.000 M EDTA solution (commercial Titriplex kit of Na_2-EDTA from Merck) using standard procedures (50° C, xylenol orange indicator).

The result of 7 independent titrations experiments provided a 0.1079(1) mol/kg concentration for the stock solution.

Standard procedure adopted for Zr(IV)-Desferrioxamine and Zr(IV)-hydroxo potentiometric titrations

Stock solution of $ZrCl_4$ in 0.1 M HCl (see above) and solid acetohydroxamic acid (Sigma-Aldrich) have been used in the measurements.

Potentiometric (pH-metric) titrations, used to determine ligand protonation and complex stability constants, were performed in 0.1 M NMe_4Cl at 298.1 ± 0.1 K using an automated apparatus and a procedure previously described.[1] The combined Metrohm 6.0262.100 electrode was calibrated as a hydrogen-ion concentration probe by titrating previously standardized amounts of HCl with CO_2-free NaOH solutions and determining the equivalent point by Gran's method,[2] which gives the standard potential, E°, and the ionic product of water (pKw = 13.83(1) in 0.1 M NMe_4Cl at 298.1 K). Calibration was performed before and after each measurement to ensure constancy of cell parameters. Computer program HYPERQUAD[3] was used to calculate ligand protonation and complex stability constants from potentiometric data.

Six titrations were performed with metal to ligand ratios ranging from 1:1 to 1:8 in the pH range 2.3-11.2.

Zr(IV)/AHA solutions are allowed to equilibrate pre-emptively in the measurement cell until no further emf variations are observed. Despite complex formation in this case being rather fast, a precautionary margin of 40 minutes was allowed nonetheless.

Acidic branch of the titrations is fast: it can be reliably recorded by waiting 1-2 minutes per point between titrant injection and emf readings (0.05 mV maximum allowed standard deviation, 0.1 mV/min maximum allowed drift).

As we approach pH 7, starting from about pH 5 and ending above pH 9, the system requires longer waiting time between injection and emf reading. It is unclear whether the slowness is related to polynuclear species, or if the simultaneous presence of many species at equilibrium in an already delicate pH range imposes a slightly longer waiting time to collect high quality data. For this reason, in this pH region, measurements were conducted with longer holding times (10 to 20 minutes depending on Zr:AHA ratio) between injection and emf readings. This allowed for the collection of points with the same quality parameters (0.05 mV maximum allowed standard deviation, 0.1 mV/min maximum allowed drift).

After pH 9.5, measurements become relatively fast again, and points can be collected within a 1-2 minutes timeframe each.

The protonation constant of acetohydroxamate was redetermined under our experimental conditions by using the same apparatus and procedure. The obtained value is reported in Table S1. Table S1 also includes values of the previously determined[4] hydrolysis constants of Zr(IV) that were used in the calculation of complex stability constants.

Fitting of the whole set of six titration curves was performed by using a starting model comprising the $[Zr(AHA)_n]^{n-4}$ (n = 1-4) species and their hydrolysed forms. After this first trial, the acidic branches of the curves were satisfactorily fitted with a complexation model composed of $[Zr(AHA)_n]^{n-4}$ (n = 1-4), $[Zr(AHA)OH]^{2+}$ and $[Zr(AHA)(OH)_2]^{+}$. Fitting of the alkaline branches of the titration curves was achieved by insertion of the trinuclear $[Zr_3(AHA)_3(OH)_7]^{2+}$ and $[Zr_3(AHA)_3(OH)_8]^{+}$ species into the complexation model. Better or alternative models were pursued in vain by introduction of additional and/or different species. We only observed, that satisfactory fitting was also possible by replacing $Zr_3(AHA)_3(OH)_7]^{2+}$ and $[Zr_3(AHA)_3(OH)_8]^{+}$ with pairs of higher order oligomers such as $Zr_6(AHA)_6(OH)_{14}]^{4+}$ and $[Zr_6(AHA)_6(OH)_{16}]^{2+}$ or $Zr_9(AHA)_9(OH)_{21}]^{6+}$ and $[Zr_9(AHA)_9(OH)_{24}]^{3+}$, with no changes (within the experimental errors) of the stability constants of the other complex species. Each individual curve was satisfactorily fitted by the final model.

The final refinement was made by considering the model with the trinuclear complexes. The values of the obtained constants are listed in Table S1.

Table S1. Equilibrium constants of the AHA^- protonation and Zr^{4+} complex formation AHA^- determined in this work in 0.1 M NMe_4Cl at 298.1 K. Hydrolysis constants of Zr(VI) taken for ref. 4.

Equilibrium	log β
$AHA^- + H^+ = HAHA$	9.239(4)
$Zr^{4+} + AHA^- = [ZrAHA]^{3+}$	14.57(9)
$Zr^{4+} + 2AHA^- = [Zr(AHA)_2]^{2+}$	26.43(8)
$Zr^{4+} + 3AHA^- = [Zr(AHA)_3]^{+}$	35.4(1)
$Zr^{4+} + 4AHA^- = [Zr(AHA)_4]$	39.87(6)
$Zr^{4+} + AHA^- + H_2O = [Zr(AHA)OH]^{2+} + H^+$	12.77(8)
$Zr^{4+} + AHA^- + 2H_2O = [Zr(AHA)(OH)_2]^{+} + 2H^+$	10.68(8)
$3Zr^{4+} + 3AHA^- + 8H_2O = [Zr_3(AHA)_3(OH)_8]^{+} + 8H^+$	23.21(5)
$3Zr^{4+} + 3AHA^- + 7H_2O = [Zr_3(AHA)_3(OH)_7]^{2+} + 7H^+$	31.8(1)
$Zr^{4+} + H_2O = ZrOH^{3+} + H^+$	-1.71(6)
$Zr^{4+} + 2H_2O = Zr(OH)_2^{2+} + 2H^+$	-3.81(3)
$Zr^{4+} + 3H_2O = Zr(OH)_3^{+} + 3H^+$	-6.67(4)
$Zr^{4+} + 4H_2O = Zr(OH)_4 + 4H^+$	-11.51(9)

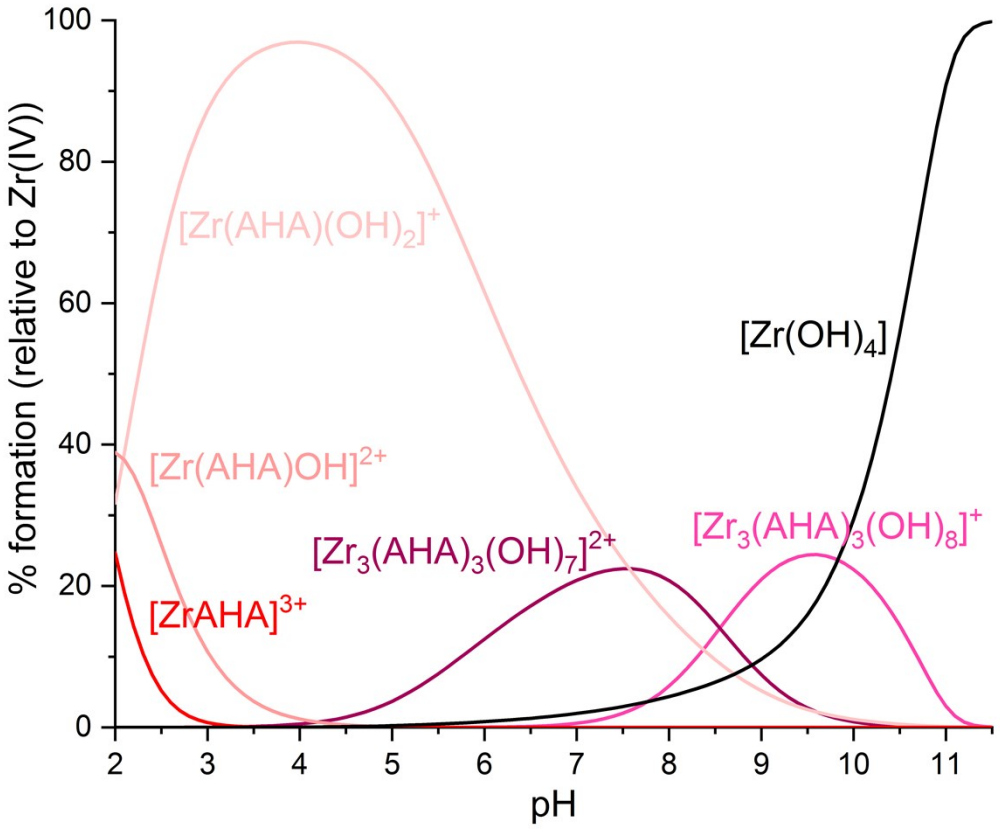

Fig. S1 Distribution diagram of the species formed as a function of pH in the Zr^{4+}/AHA system in aqueous solution. $[Zr^{4+}] = [AHA^{-}] = 1mM$. Species formed in less than 5% are not represented.

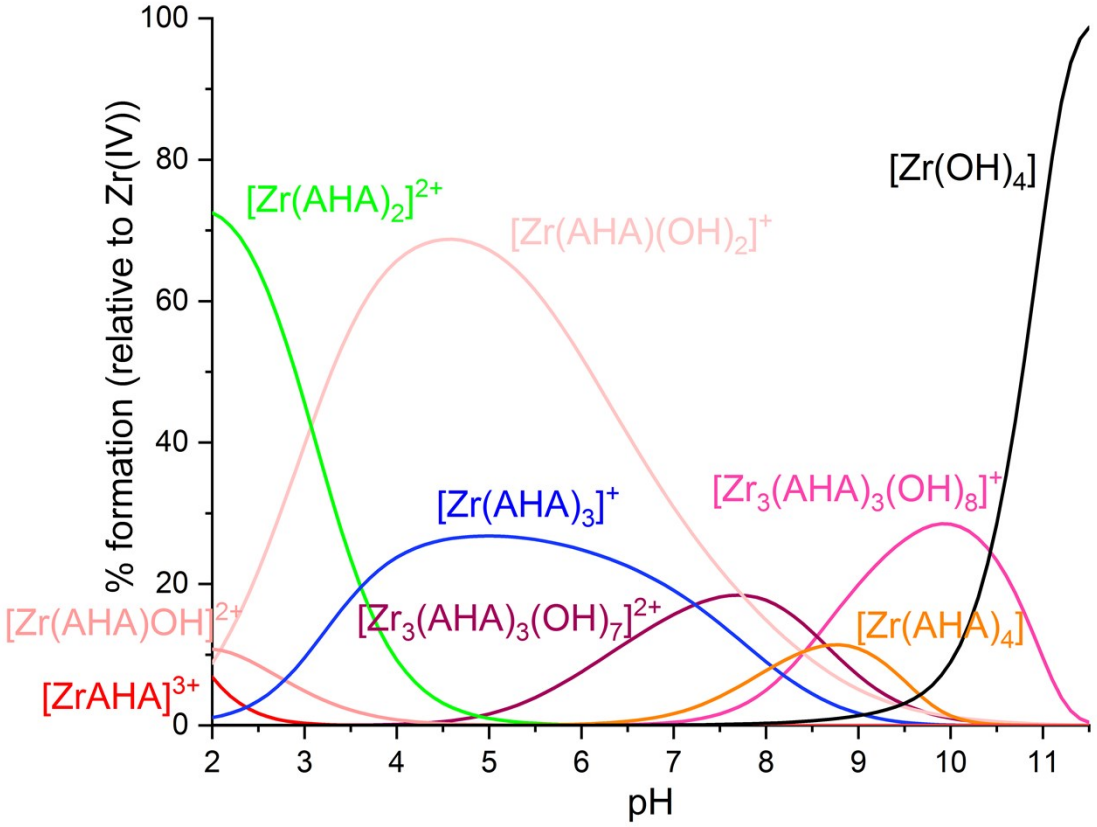

Fig. S2 Distribution diagram of the species formed as a function of pH in the Zr^{4+}/AHA system in aqueous solution. [Zr^{4+}] = 1mM, [AHA^-] = 2mM. Species formed in less than 5% are not represented.

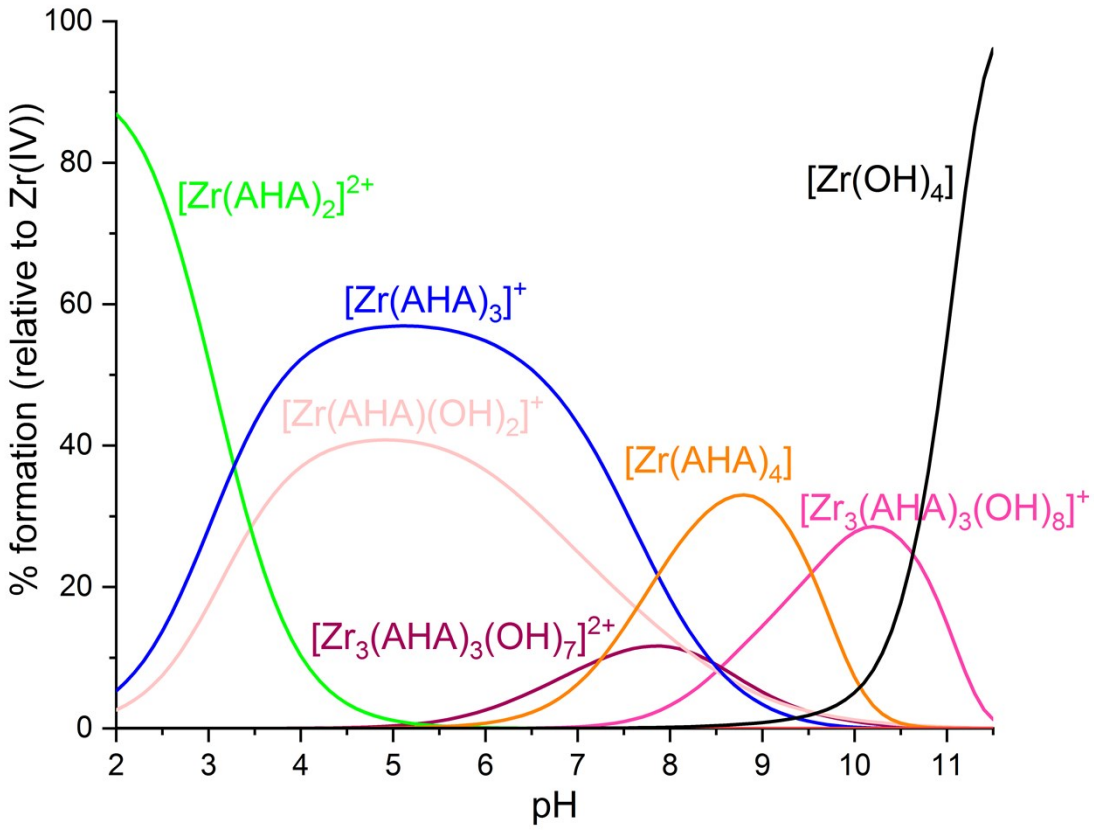

Fig. S3 Distribution diagram of the species formed as a function of pH in the Zr^{4+}/AHA system in aqueous solution. [Zr^{4+}] = 1mM, [AHA^-] = 3mM. Species formed in less than 5% are not represented.

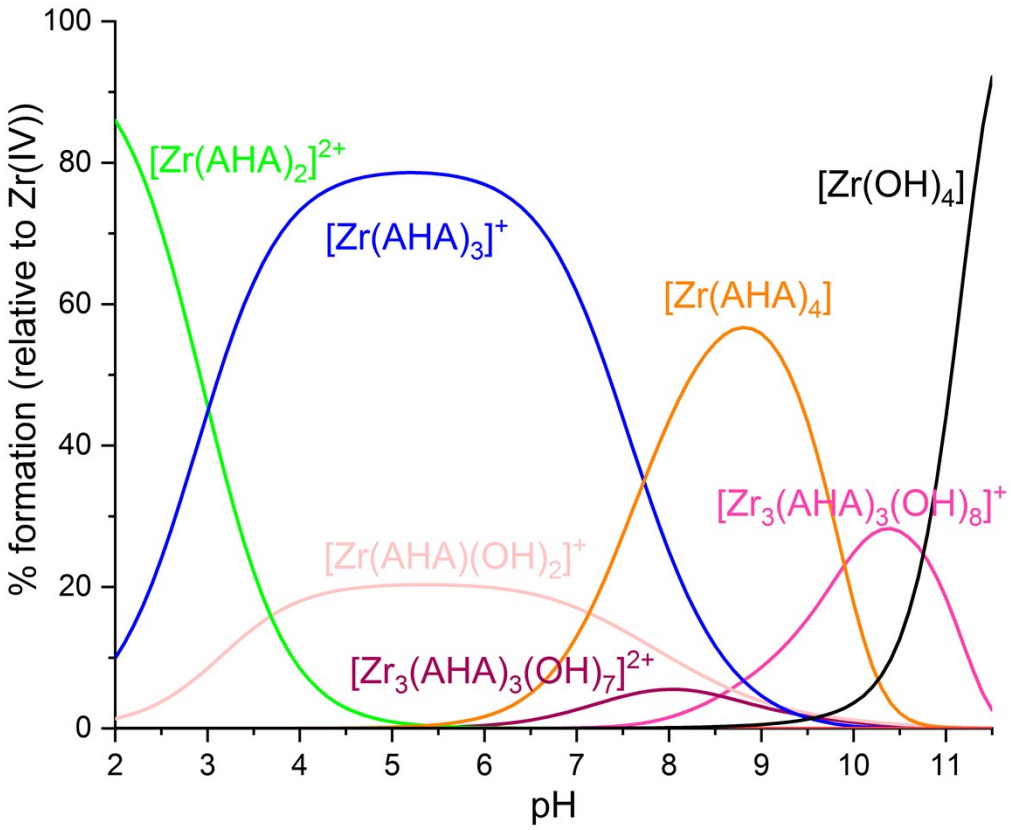

Fig. S4 Distribution diagram of the species formed as a function of pH in the Zr^{4+}/AHA system in aqueous solution. [Zr^{4+}] = 1mM, [AHA^-] = 4mM. Species formed in less than 5% are not represented.

References

1) C. Bazzicalupi, A. Bianchi, T. Biver, C. Giorgi, S. Santarelli and M. Savastano, *Inorg. Chem.* 2014, **53**, 12215–12224.

2) G. Gran, *Analyst* (London) 1952, **77**, 661–671.

3) P. Gans, A. Sabatini and A. Vacca, *Talanta* 1996, **43**, 1739−1753.

4) M. Savastano, C. Bazzicalupi, G., Ferraro, E. Fratini, P. Gratteri and A. Bianchi, *Molecules*, 2019, **24**, 2098.