

Kinetic effects on the interactions of Rh(III) with humic acids as determined using size exclusion chromatography (SEC)

Antonio Cobelo-García

Marine Biogeochemistry Research Group. Instituto de Investigacións Mariñas (CSIC). 36208 Vigo, Spain

*Corresponding author:

e-mail: acobelo@iim.csic.es

Tel. +0034 986 231930 Ext. 145

Fax: +0034 986 292762

Abstract

The anthropogenic inputs of Rh in the environment – together with other platinum group elements – have increased considerably during the last 20-30 years. However, thermodynamics and kinetics on the interaction of Rh with natural organic and inorganic ligands are still poorly characterized. Here we report the time-dependent speciation of rhodium chlorides spiked to model freshwater with and without the presence of humic substances. Rhodium species were determined using size exclusion chromatography – inductively coupled plasma mass spectrometry (SEC-ICPMS). Results indicate that organic matter can effectively bind rhodium, but the extremely slow reaction kinetics of Rh plays a significant role on its speciation in natural waters. Accordingly, formation of Rh-organic complexes from spiked rhodium chlorides required around 15 days to reach equilibrium; this should be taken into account in those laboratory experiments where the biological interactions of Rh, using spiked samples, are studied. Regarding Rh inorganic speciation in freshwater, the available thermodynamic constants predict the dominance of the neutral tri-hydroxo and negatively-charged tetra-hydroxo rhodium complexes over typical pHs (6-8); our results, however, indicate only the presence of negatively-charged hydroxocomplexes at pH 7. Re-examination of the Rh stability constants suggest that these hydroxylated rhodium complexes may also dominate its inorganic speciation in seawater.

Keywords

Rhodium, speciation, natural waters, humic acids, size exclusion chromatography

Kinetic effects on the interactions of Rh(III) with humic acids as determined using size exclusion chromatography (SEC)

1. Introduction

Rhodium represents one of the least abundant trace elements in the environment, with a crustal concentration of around 0.06 ng g^{-1} (Wedepohl 1995) and below 0.4 ng L^{-1} in water bodies (Ravindra et al. 2004). In anthropogenically impacted areas, especially those close to roads and urban areas, a 10^2 - 10^3 fold enrichment has been found leading to a significant scientific effort on the development of reliable analytical methodologies for the determination of its environmental concentrations, mobility, reactivity and biological impact (e.g. Rauch et al. 2000; Jarvis et al. 2001; Moldovan et al. 2001; Whiteley and Murray 2005; Zereini et al. 2007; Dahlheimer et al. 2007; Colombo et al. 2008; Cobelo-Garcia et al. 2008). This enhanced Rh environmental concentration has been linked to its use (together with Pt and Pd) as catalysts in vehicle exhaust systems, which were introduced in the past 20-30 years. Here, platinum group elements (PGE: Pt, Pd, and Rh) convert noxious compounds, such as hydrocarbons, carbon monoxide and nitrogen oxides, into more innocuous compounds like carbon dioxide, nitrogen and water (Moldovan et al. 2006). The relative and absolute concentrations of PGE in the catalyst depend on a number of factors, including manufacturer, vehicle type, engine and catalytic functions required, although values of total PGE are roughly around 0.5% (w/w) of the catalyst (Reinhard 2006). During abrasion and aging of the washcoat layer of the catalyst, PGE are emitted as particles at rates of 6-161 (Pt), 8-54 (Pd), and 3-77 (Rh) ng km^{-1} , depending on the type and age of catalyst, type of engine, vehicle speed, status of the vehicle, way of driving and meteorological conditions (Moldovan et al. 2006).

Rhodium speciation in natural waters is not well characterized, despite the well-known dependence of the biological availability (including toxicity) of trace elements with their chemical speciation (e.g. Campbell 1995). The available information indicates that inorganic speciation of Rh is dominated by aquo-hydroxo, aquo-chloro and mixed aquo-hydroxo-chloro complexes in the form of $[\text{RhCl}_x(\text{OH})_y(\text{H}_2\text{O})_{6-x-y}]^{3-x-y}$, $x, y = 0$ to 6 and $x+y \leq 6$ (Buslaeva and Simanova 1999), and there is evidence for interaction of the metal with dissolved organic matter (Menzel et al. 2001; Cobelo-Garcia et al. 2007). There is, however, no complete or reliable dataset for kinetic and thermodynamic parameters of Rh(III) complexes with relevant inorganic (e.g. Cl^- , OH^-) and organic ligands. This may be attributed to the determination of constants at different temperatures and/or ionic strengths, and under conditions that are not environmentally relevant (Buslaeva and Simanova 1999), and the remarkably slow kinetics involved in the Rh(III) ligand replacement reactions, thereby casting doubt onto how close experimental systems have been (Gerber et al. 2010).

Lack of reliable thermodynamic and kinetic information on the interaction of rhodium with natural organic and inorganic ligands is a major weakness for the assessment of the reactivity, transport and biological impacts of rhodium in the environment. To this end, we have conducted a series of time-dependent experiments to monitor the speciation of Rh spiked – as its chloride forms in order to mimic the conditions normally used in Rh bioaccumulation experiments (e.g. Sures and Zimmermann 2007) – into model freshwater with and without the presence of humic substances. Results are discussed in terms to (i) compare the experimental observations obtained with speciation calculations using available thermodynamic data, (i) the implications for Rh reactivity and mobility in the environment, and (ii) the critical kinetic issues that need to be considered in the experimental approaches of Rh bioaccumulation studies.

2. Materials and Methods

2.1 Apparatus and Reagents

Potassium perchlorate (KClO_4), disodium phosphate (HNa_2PO_4), and monosodium phosphate (H_2NaPO_4) were purchased from Fluka (Biochemika Ultra, $\geq 95.5\%$). A highly hydrophilic synthetic polymer stationary phase (porous spherical polyhydroxymethacrylate gel) column was used (PolySep-GFC-P 2000 from Phenomenex, Macclesfield, Cheshire, UK). The separation range specified by the manufacturer is 0.1-10 kDa. The system consisted of a solvent delivery system, a Rheodyne six-port PEEK injection valve, and a quadrupole ICP-MS (VG Plasmaquad 3, Thermo Elemental, Winsford, UK). A PEEK sample loop of 200 μL was used. In order to correct for instrumental drift during the duration of the chromatograms, an internal standard (In at a concentration of 10 $\mu\text{g L}^{-1}$ each) was added prior to the introduction of the sample in the ICP-MS at a flow rate of 0.5 mL min^{-1} with the aid of a T-junction using a peristaltic pump.

2.2 Mobile Phase and Calibration of the Column

Mobile phase consisted of organic-free artificial freshwater (prepared as given in Errecalde et al. 1998) to which a non-metal complexing background electrolyte (10 mM KClO_4) and phosphate buffer (5 mM) were added. Phosphate buffer was used to adjust the mobile phase at the desired pH (7.0 ± 0.1). The column was calibrated using the following organic substances (Figure S1): ethylene glycol (MW=62 Da), L-Cysteine (MW=121 Da), HEPPSO (MW=268 Da), and two polystyrenesulfonic acid salts of MW=6800 (PPS1) and MW=4300 (PSS2) Da, respectively. The void volume was calculated using Blue Dextran (MW=2 10^6 Da). A flow rate of 0.8 mL min^{-1} was used in all experiments.

2.3 SEC-UV of the Humic Material

The UV chromatogram of the AHA (Aldrich humic acids; 100 mg L^{-1} in mobile phase) was recorded for the characterization of the size distribution of the organic matter used in the experiments (Figure 1). The humic material starts to elute at 8.4-8.5 min, i.e. 0.3-0.4 min before the void volume of the column indicating that a fraction of the organics has a MW higher than the exclusion limit of the column (10^4 Da) and/or some rejection between the organic molecules and the stationary phase was occurring at the mobile phase composition used. The peak of humic acids was centered at 9.2 min, resulting in an approx. size of the average organic molecules of $\log \text{MW } 3.67$, which is in reasonable agreement for the number-averaged molecular weight of the AHA reported in the literature ($\log \text{MW } 3.21\text{-}3.49$; Becket et al. 1987; Chin et al. 1994).

2.4 Incubation Experiments

The experimental procedure was as follows: a stock solution of 10,000 $\mu\text{g L}^{-1}$ Rh^{3+} was prepared in 1M HNO_3 from the commercial standard (10,000 mg L^{-1} of Rh(III) in 10% HCl). Immediately after (<1 min) preparation of the stock solution, a volume (generally 1 μL per mL of sample) was spiked to the sample to give a concentration of approx. 100 nM Rh(III) ($\sim 10 \mu\text{g L}^{-1}$), and the pH was brought back to 7.0 by appropriate addition of NaOH. This rhodium concentration used in the incubation experiments is higher than those present in typical natural waters (generally below 0.4 ng L^{-1} ; Ravindra et al. 2004), but was chosen to allow a direct determination using ICP-MS and is low enough to avoid formation of polynuclear complexes normally present at higher concentrations (Maslei et al. 1976).

Rhodium speciation was then monitored with respect to time by means of SEC-ICP-MS. Three different types of samples (matrices) were used: (i) mobile phase (see composition above), pH 7.0; (ii) mobile phase, pH 7.0 + 100 mg L^{-1} AHA; and (iii) mobile phase, pH 7.0 + 10 mg L^{-1} AHA. In these experiments, the sample was not passed through the UV detector in order to decrease the length of the tubes with the aim of minimize the dispersion of the sample and therefore the width of the peaks. A schematic representation of the experimental layout is given in Figure 2.

2.5 Speciation Calculations

Speciation diagrams were obtained using the Medusa software (<http://www.kemi.kth.se/medusa/>) and the stability constants reported in Tables S1 and S2 of the supplementary information.

3. Results and Discussion

3.1 Kinetic effects on the speciation of Rh(III) spiked to freshwater

Ideally, separation of different molecules using size exclusion chromatography relies only on the molecular weight; in practice, interactions of the analytes with the stationary phase may lead to early or retarded elution (e.g. Piatina and Hering 2000). We took advantage of this phenomenon to separate the different Rh(III) species depending on their charge, which determines the affinity or repulsion with the stationary phase. The column used in this study is packed with a hydrophilic polyhydroxymethacrylate gel, containing hydroxyl groups (and potentially residual carboxyl groups); negatively charged Rh(III) complexes are expected to suffer from early elution due to charge exclusion with the hydroxyl groups, whereas positively charged complexes are likely to suffer from an opposite behavior. Peak assignment to Rh(III) species was done based on charge interactions and Rh(III) kinetic parameters, as discussed below.

The chromatograms recorded at short incubation times (5-275 min, Figures 3-5) show a similar pattern independently of the presence and amount of humic material. The first Rh(III) species (peak 3) appears at an elution time of 10.5 min., which corresponds – in the theoretical case of no analyte-stationary phase interaction – with a molecule weight of approx. 1.7 kDa. This value is one order of magnitude higher than the molecular weight of the possible Rh(III) species present in solution (0.2-0.3 kDa): $[\text{RhCl}_x(\text{OH})_y(\text{H}_2\text{O})_z]^{3-x-y}$, where $x \leq 6$; $0 \leq y \leq 6-x$; $z = 6-x-y$. If we assume that Rh(III) species in the commercial stock solution – supplied at a HCl concentration of approx. 3.0M – are at thermodynamic equilibrium, then ~60% of Rh(III) will be in the form of $[\text{RhCl}_5(\text{H}_2\text{O})]^{2-}$ + $[\text{RhCl}_6]^{3-}$ and the remaining ~40% as *cis*- $[\text{RhCl}_4(\text{H}_2\text{O})_2]^-$ (Gerber et al. 2010). The pseudo-first order aquation rate constants at zero ionic strength – extrapolated from the data given by Gerber et al. (2010) – are 0.56 min^{-1} for $[\text{RhCl}_6]^{3-}$ ($t_{1/2} = 1.2 \text{ min}$) and 0.018 min^{-1} for $[\text{RhCl}_5(\text{H}_2\text{O})]^{2-}$ ($t_{1/2} = 39 \text{ min}$). It is therefore expected that almost no $[\text{RhCl}_6]^{3-}$ will be present in the sample solutions after 5 minutes of equilibration and $[\text{RhCl}_5(\text{H}_2\text{O})]^{2-}$ will disappear during the first few hours. We can therefore assign peak 3 to the rhodium pentachloride species.

The major peak, centered at 11.6 min (peak 1), is assigned to *cis*- $[\text{RhCl}_4(\text{H}_2\text{O})_2]^-$, which results from the aquation of $[\text{RhCl}_5(\text{H}_2\text{O})]^{2-}$; these species have a pseudo-first order aquation rate constant of $1.27 \cdot 10^{-4} \text{ min}^{-1}$ (at 0.1 M HCl ionic strength; $t_{1/2} = 3.8 \text{ days}$) to yield *fac*- $[\text{RhCl}_3(\text{H}_2\text{O})_3]^0$, which appears at 14.1 min (peak 2). Since this *fac*- $[\text{RhCl}_3(\text{H}_2\text{O})_3]^0$ is a non-charged polar molecule, it should behave ‘ideally’ from the point of view of size exclusion chromatography. Its elution time corresponds to a MW of 90 Da, which is lower than the actual MW of 263 Da but closer to the real value than those for the other charged molecules. After one day of equilibration, the amount of *cis*- $[\text{RhCl}_4(\text{H}_2\text{O})_2]^-$ has decreased significantly and a fourth peak (peak 2a), appearing at a retention time (15.3 min), higher than the permeation time of the column, is observed. Following the aquation pathway (Palmer and Harris 1975; Gerber et al. 2010) these species correspond to the cationic *cis*- $[\text{RhCl}_2(\text{H}_2\text{O})_4]^+$. These cationic species are likely to have affinity with the stationary phase of the column through charge interactions with the hydroxyl and carboxyl groups, explaining the retention time beyond the permeation limit.

Chromatographic differences between the three samples appear after 6 days of equilibration (Figures 3-5). A major Rh(III) peak appears at short elution time for the three types of samples, with minor contributions of *cis*- $[\text{RhCl}_4(\text{H}_2\text{O})_2]^-$ and *cis*- $[\text{RhCl}_2(\text{H}_2\text{O})_4]^+$. However, the elution time of the dominant peak for the three samples differ, indicating the presence of different Rh(III) species. Accordingly, in the absence of humic acids, this peak (peak 4 in Figure 3) is centered at 10.4 min, similar to the elution time obtained for $[\text{RhCl}_5(\text{H}_2\text{O})]^{2-}$. Assuming no polymerization of the rhodium

complexes, which is unlikely at the working concentrations used (Maslei et al. 1976), this would indicate the presence of a negatively-charged hydroxylated Rh(III) complexes in solution. We suspect that acid hydrolysis yielding the final Rh-hydroxide complexes starts before the two chlorides can be substituted by water molecules, as this process seems to be more rapid than aquation (Palmer and Harris 1975) and is thermodynamically favorable; accordingly, the ratio $[\text{RhCl}_2(\text{OH})(\text{H}_2\text{O})_3]^0/\text{cis-}[\text{RhCl}_2(\text{H}_2\text{O})_4]^+$ is approx. 20 at the sample pH (7.0). Also, the final species start to appear just after formation of $\text{cis-}[\text{RhCl}_2(\text{H}_2\text{O})_4]^+$ and no intermediate complex is observed during the experiments. In the presence of 100 mg L^{-1} of AHA, after 6 days of incubation, the dominant peak of Rh(III) is observed at short retention times (9.6 min; peak 5 in Figure 4), and becomes the only peak after 15 days. At longer incubation times (22 and 31 days), this peak elutes at slightly lower retention times (9.4 min) and the front of the peak becomes more vertical. The fact that this elution time is different than in the absence of organic matter (that appeared at 10.4 min) and very similar to the elution time of the humic substances (9.2 min; Figure 2) indicates that this peak corresponds to Rh(III) bound to organic matter. When lower concentrations of humic acids (10 mg L^{-1}) are present, results are intermediate of those obtained in the absence of organic matter and with 100 mg L^{-1} AHA (Figure 5): a dominant broad peak is observed after 6 days of equilibration, starting to elute at the same retention time than the organic peak of Rh(III) observed in the presence of 100 mg L^{-1} (approx. 8.8 min) but with the maximum intensity at approx. 10.0 min (Figure 5) which is within the retention times of the two experiments reported above. This indicates that, even after a long equilibration time of 31 days, the co-existence of organic complexes and negatively-charged hydroxo species of Rh(III) in the sample is evident.

3.2 Comparison of Rh(III) inorganic speciation with thermodynamic calculations

Speciation calculations of Rh with inorganic ligands in freshwater ($1 \cdot 10^{-3} \text{ M}$ chloride) were carried out using the stability constants available in the literature which are compiled in Tables S1 and S2 of the supplementary information. Results predict that rhodium is dominated by hydroxylated species over the entire pH range; under typical freshwater pHs (6-8), rhodium inorganic speciation is composed by $[\text{Rh}(\text{OH})_3(\text{H}_2\text{O})_3]^0$ and $[\text{Rh}(\text{OH})_4(\text{H}_2\text{O})_2]^-$ (Figure 6). These results arise from the stability constants of the hydroxo-complexes of rhodium given by Maslei et al. (1976) as chloride complexes are not significant at environmentally relevant pHs. The existence of two rhodium species does not agree with the chromatographic results, which suggest the presence of a single dominant type of species. The possibility of rhodium polymerization leading to the single peak observed seems unlikely as the concentrations in our experiments ($0.1 \cdot 10^{-6} \text{ M}$ Rh) are one order of magnitude lower than those used by Maslei et al. (1976; $1\text{-}5 \cdot 10^{-6} \text{ M}$) where they reported no polymerization under such conditions. Therefore no polymeric hydroxo complexes of rhodium should be present in our experiments. A possible explanation for this situation is that Maslei et al. (1976) did not use final equilibrium pHs in their calculations. They used $\text{Rh}(\text{ClO}_4)_3$ as the starting species, and upon acid hydrolysis decrease the initial sample pH, and this is especially critical at near-neutral values. If that was the case, the predominance area for each of the rhodium-hydroxide complexes would shift towards lower pHs, in agreement with our proposed speciation.

Regarding chloride complexes, which may be important in saline waters, Gerber et al. (2010) have showed that the available constants (see Table S2) are incorrect as they predict complexes with higher chloride content than those obtained in their experimental results undertaken under equilibrium conditions in chloride solutions. Although it is evident that the stability of chloride and hydroxide complexes of rhodium need further refinements, we used them to calculate a 'tentative' Rh(III) speciation in seawater. Resulting speciation indicates that under typical pH conditions, Rh(III) is dominated by hydroxylated complexes ($[\text{Rh}(\text{OH})_4(\text{H}_2\text{O})_2]^-$ and $[\text{Rh}(\text{OH})_5(\text{H}_2\text{O})]^{2-}$), while chlorides are only relevant at $\text{pH} < 7$.

3.3 Implications for the speciation and reactivity of Rh(III) in natural waters

1 The study and prediction of the reactivity and mobility of Rh(III) in the environment requires a
2 knowledge on its reaction kinetics and thermodynamic data with naturally-occurring inorganic and
3 organic ligands. The extremely slow kinetics of Rh(III) reactions may control to a significant extent
4 its speciation in natural waters. This can be especially critical in rapid changing environments (e.g.
5 estuaries) where the concentration of inorganic and organic ligands varies in a short time scale. Here,
6 ambient speciation may reflect past and present scenarios as suggested for other kinetically impeded
7 elements (e.g. Pt; Cosden and Byrne 2003). Also, the interactions of Rh with organic matter, as shown
8 in this study, may control its transport or fixation in organic-rich aquatic systems with elevated
9 rhodium concentrations derived from vehicular traffic pressure (e.g. urban lakes and rivers, road
10 run-off, etc.).

11 Chromatographic results here obtained suggests that inorganic rhodium speciation in freshwaters is
12 dominated by negatively-charged hydroxides. It is also interesting that tentative speciation
13 calculations in seawater using reported stability constants also suggest the predominance of
14 hydroxylated species. These species, having a negative charge, are likely to have little attraction for
15 the negatively charged surface of the marine particles. In our previous studies (Cobelo-García et al.
16 2007; 2008) we observed the precipitation of Rh spiked to seawater under typical pHs, and this was
17 interpreted as the salt-induced destabilization of polymerized Rh complexes formed at such
18 concentrations ($10 \mu\text{g L}^{-1}$) at alkaline conditions. It is, however, unlikely that this polymerization
19 occurs at typical concentrations ($<0.4 \text{ ng L}^{-1}$). Accordingly, Bertine et al. (1996) observed that Rh is
20 remarkably stabilized in seawater compared to its crustal abundance; the suggested Rh speciation as
21 negatively-charged hydroxylated complexes in seawater would explain such behavior.
22
23

24 **3.4 Recommendations for laboratory experiments on the reactivity and bioavailability of Rh**

25 The kinetic impediments of Rh are critical for those laboratory experiments where the reactivity and
26 bioavailability of rhodium is studied after being spiked with different amounts of this element –
27 generally, as in the present work, as its chloride complexes (e.g. Sures and Zimmermann 2007; Diehl
28 and Gagnon 2007; Mulholland and Turner 2011). Here we have shown that the equilibrium
29 speciation of Rh is not rapidly attained; given that metal bioavailability and reactivity greatly
30 depends on its speciation, samples should be allowed to equilibrate after spiking (>15 days) before
31 the experiment is started to let rhodium reach equilibrium with the inorganic and organic ligands
32 present. Otherwise, results would not reflect the real environmental speciation and may lead to
33 inaccurate estimations of Rh biological interactions.
34
35
36
37
38
39

40 **4. Acknowledgments**

41 We gratefully acknowledge the financial support by the European Union under the Marie Curie
42 fellowship scheme (Contract No. MEIF-CT-2005-009152) and the P.I. project (ref. 2008830I150)
43 from the CSIC (*Consejo Superior de Investigaciones Científicas*) granted to A.C.G. This work benefited
44 from the fruitful comments and suggestions given by Prof. G.E. Millward and Dr. A. Turner
45 (University of Plymouth, UK).
46
47
48
49
50
51

52 **5. References**

53 Beckett R, Jue Z, Giddings JC (1987) Determination of molecular-weight distributions of fulvic and
54 humic acids using flow field-flow filtration. *Environ Sci Technol* 21: 289-295.
55

56 Bertine KK, Koide M, Goldberg ED (1996). Comparative marine chemistries of some trivalent metals
57 – bismuth, rhodium and rare earth elements. *Mar Chem* 53: 89-100.
58
59
60
61
62
63
64
65

1 Buslaeva TM, Simanova SA (1999) Platinum metals in aqueous hydrochloric and chloride solutions:
2 palladium, platinum, rhodium, and iridium. *Russ J Coord Chem* 25: 151-161.

3 Campbell PGC (1995) Interactions between trace metals and aquatic organisms: a critique of the
4 free-ion activity model. In Tessier A and Turner DR (eds) *Metal Speciation and Bioavailability in*
5 *Aquatic Systems*. John Wiley and Sons Ltd, Chichester, pp. 45-102.

6
7 Chin YP, Aike G, O'Loughlin E (1994) Molecular weight, polydispersity, and spectroscopic properties
8 of aquatic humic substances. *Environ Sci Technol* 28: 1853-1858.

9
10 Cobelo-García A, Turner A, Millward GE, Couceiro F (2007) Behaviour of palladium(II), platinum(IV),
11 and rhodium(III) in artificial and natural waters: influence of reactor surface and geochemistry on
12 metal recovery. *Anal Chim Acta* 585: 202-210.

13
14 Cobelo-García A, Turner A, Millward GE (2008) Fractionation and reactivity of platinum group
15 elements during estuarine mixing. *Environ Sci Technol* 42: 1096-1101.

16
17 Colombo C, Monhemius AJ, Plant JA (2008) The estimation of the bioavailabilities of platinum,
18 palladium and rhodium in vehicle exhaust catalysts and road dusts using a physiologically based
19 extraction test. *Sci Total Environ* 389: 46-51.

20
21 Cosden JM, Byrne RH (2003) Comparative geochemistries of Pd^{II} and Pt^{II}: formation of mixed
22 hydroxychloro and chlorocarbonato-complexes in seawater. *Geochim Cosmochim Acta* 67: 1331-
23 1338.

24
25 Dahlheimer SR, Neal CR, Fein JB (2007) Potential mobilization of platinum-group elements by
26 siderophores in surface environments. *Environ Sci Technol* 41: 870-875.

27
28 Diehl, DB, Gagnon ZE (2007) Interactions between essential nutrients with platinum group metals in
29 submerged aquatic and emergent plants. *Water Air Soil Pollut* 184: 255-267.

30
31 Errecalde O, Seidl M, Campbell PGC (1998) Influence of a low molecular weight metabolite (citrate)
32 on the toxicity of cadmium and zinc to the unicellular green alga *Selenastrum capricornutum*: an
33 exception to the free-ion model. *Water Res* 32: 419-429.

34
35 Gerber WJ, Koch KR, Rohwer HE, Hosten EC, Geswindt TE (2010) Separation and quantification of
36 [RhCl_n(H₂O)_{6-n}]³⁻ⁿ (n=0-6) complexes, including estereoisomers, by means of ion-pair HPLC-ICP-MS.
37 *Talanta* 82: 348-358.

38
39 Jarvis KE, Parry SJ, Piper JM (2001) Temporal and spatial studies of autocatalyst-derived platinum,
40 rhodium and palladium and selected vehicle derived trace elements in the environment. *Environ Sci*
41 *Technol* 35: 1031-1036.

42
43 Maslei, NN, Nabivanets BI, Yantso EA (1976) Mononuclear complexes of rhodium(III). *Ukrainskiy*
44 *Khimicheskiy Zhurnal* 3: 247-251.

45
46 Menzel CM, Berner Z, Stüben D (2001) Coupling size-exclusion chromatography and ICP-MS to
47 investigate the speciation of platinum-group elements in environmental samples. *Geostandard*
48 *Newslett* 25: 239-251.

49
50 Moldovan M, Rauch S, Gómez M, Palacios MA, Morrison GM (2001) Bioaccumulation of palladium,
51 platinum and rhodium from urban particulates and sediments by the freshwater isopod *Asellus*
52 *Aquaticus*. *Water Res* 35: 4175-4183.

53
54 Moldovan M, Gómez-Gómez MM, Palacios-Corvillo MA (2006) Release of particulate and acid soluble
55 palladium from catalytic converters into the environment. In Zereini F and Alt F (eds) *Palladium*
56 *Emissions in the Environment*. Springer-Verlag Berlin Heidelberg, pp. 25-38.

57
58
59
60
61
62
63
64
65

1 Mulholland R, Turner A (2011) Accumulation of platinum group elements by the marine gastropod
2 *Littorina littorea*. Environ Pollut 159: 977-982.

3 Palmer DA, Harris GM (1975) Kinetics, mechanism, and stereochemistry of the aquation and chloride
4 anation reactions of fac- and mer-trichlorotriaquorhodium(III) complexes in acidic aqueous solution.
5 A complete reaction scheme for complex ions of the general formula $[\text{RhCl}_n(\text{OH}_2)_{6-n}]^{3-n}$. Inorg Chem
6 14: 1316-1321.

7
8 Piatina TB, Hering JG (2000) Direct quantification of metal-organic interactions by size-exclusion
9 chromatography (SEC) and inductively-coupled plasma mass spectrometry (ICP-MS). J Environ Qual
10 29: 1839-1845.

11
12 Rauch S, Motelica-Heino M, Morrison GM, Donard OFX (2000). Critical assessment of platinum group
13 elements determination in road and urban river sediments using ultrasonic nebulisation and high
14 resolution ICP-MS. J Anal Atom Spectrom 15: 329-334.

15
16 Ravindra K, Bencs L, Van Grieken R (2004) Platinum group elements in the environment and their
17 health risk. Sci Total Environ 318: 1-43.

18
19 Reinhard B (2006) Automotive catalysts. In Zereini F and Alt F (eds) Palladium Emissions in the
20 Environment. Springer-Verlag Berlin Heidelberg, pp. 3-23.

21
22 Sures B, Zimmermann S (2007) Impact of humic substances on the aqueous solubility, uptake and
23 bioaccumulation of platinum, palladium and rhodium in exposure studies with *Dreissena*
24 *polymorpha*. Environ Pollut 146: 444-451.

25
26 Whiteley JD, Murray F (2005) Autocatalyst-derived platinum, palladium and rhodium (PGE) in
27 infiltration basin and wetland sediments receiving urban runoff. Sci Total Environ 341: 199-209.

28
29 Zereini F, Wiseman C, Puettmann W (2007) Changes in palladium, platinum, and rhodium
30 concentrations, and their spatial distribution in soils along a major highway in Germany from 1994
31 to 2004. Environ Sci Technol 41: 451-456.
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

1
2 **Figure Captions**
3

4 Figure 1. UV chromatogram of 100 mg L⁻¹ AHA prepared in mobile phase. For composition of the
5 mobile phase and chromatographic conditions see section 2.
6

7 Figure 2. Schematic representation of the experimental procedure. (a) Preparation of the
8 incubation experiments; (b) layout of the analytical SEC-UV-ICPMS system and (c) injection
9 valve during sample loading (i) and injection (ii).
10

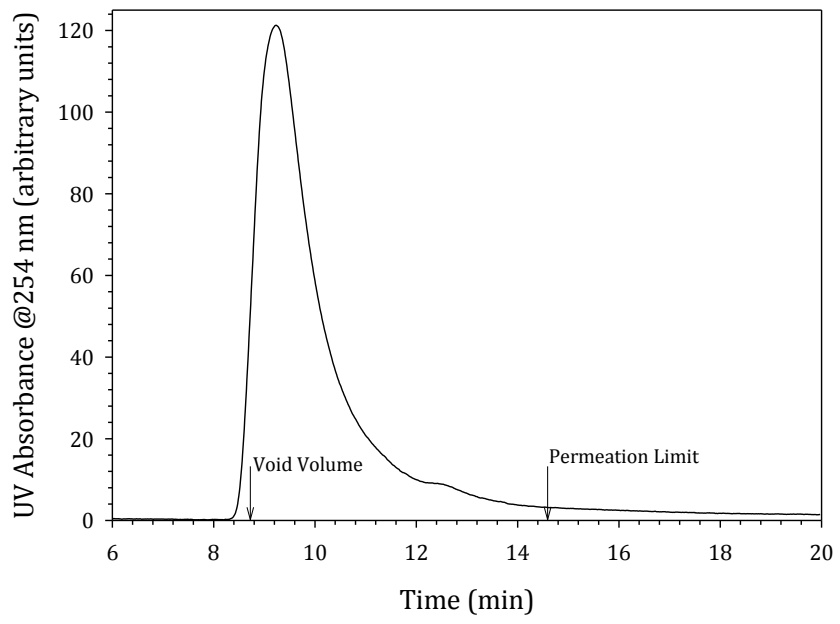
11
12 Figure 3. Time dependent size-exclusion chromatograms of 100 nM Rh(III) prepared from a
13 commercial standard stock solution in organic-free artificial freshwater (pH 7.0).
14

15
16 Figure 4. Time dependent size-exclusion chromatograms of 100 nM Rh(III) prepared from a
17 commercial standard stock solution in artificial freshwater (pH 7.0) containing 100 mg L⁻¹ of
18 humic acids.
19

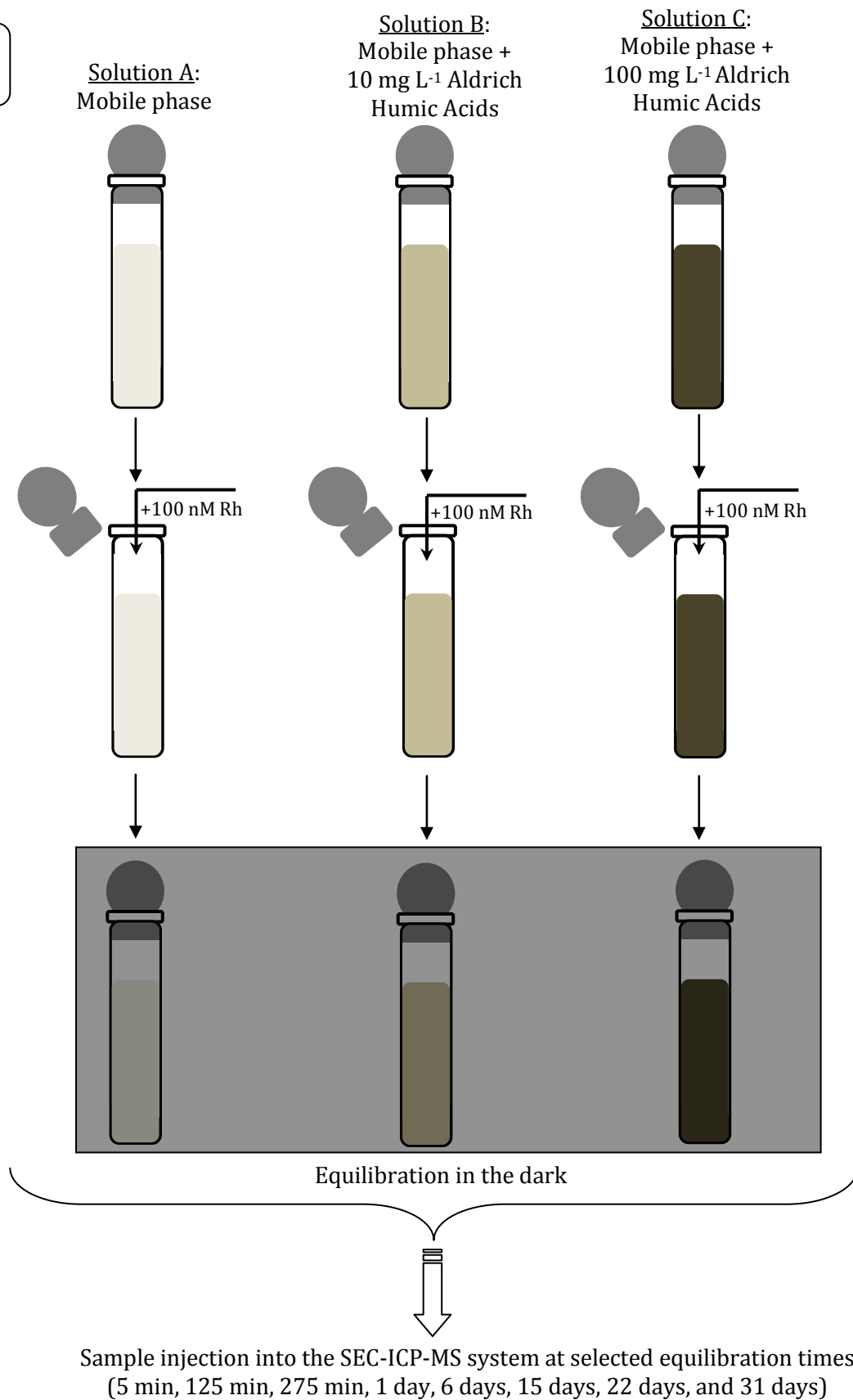
20
21 Figure 5. Time dependent size-exclusion chromatograms of 100 nM Rh(III) prepared from a
22 commercial standard stock solution in artificial freshwater (pH 7.0) containing 10 mg L⁻¹ of
23 humic acids.
24

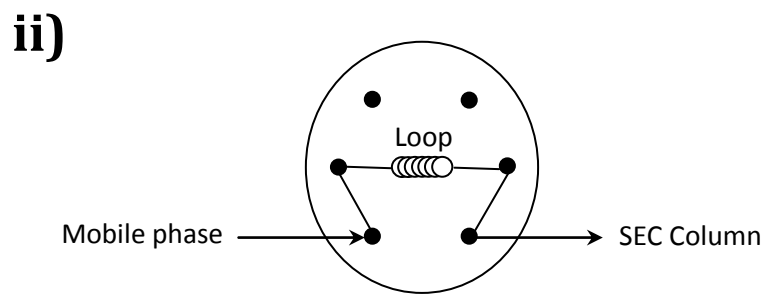
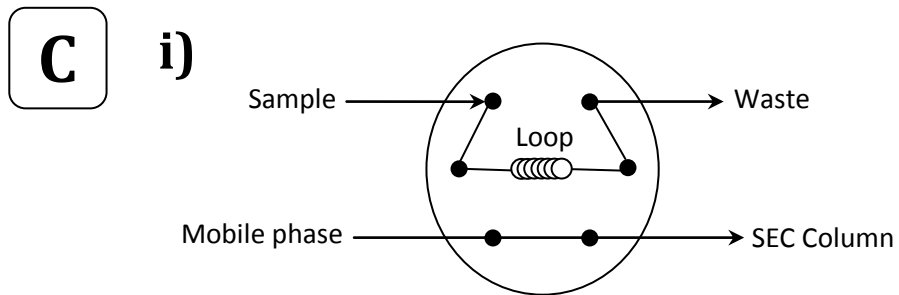
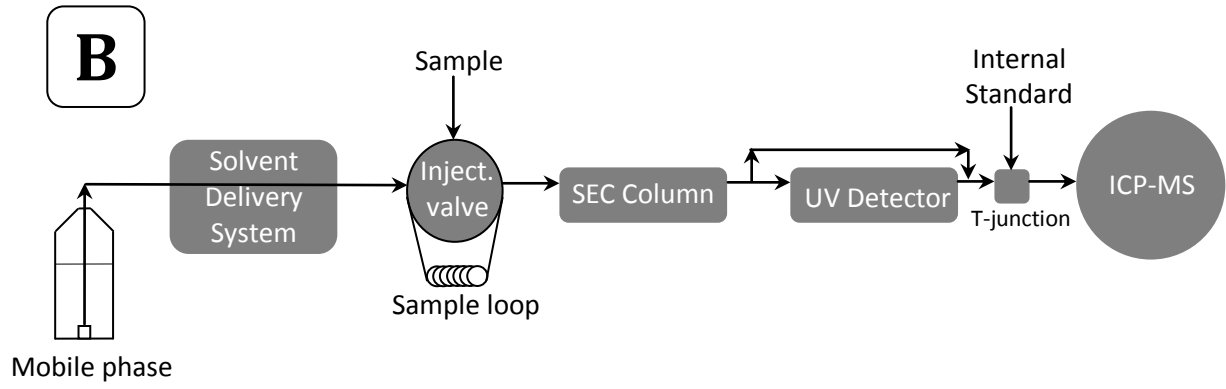
25
26 Figure 6. Calculated Rh(III) speciation in freshwater using the stability constants reported in
27 Table S1 and S2 of the supporting information. The shaded area represents the pH of the samples
28 studied in this work.
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

Figure

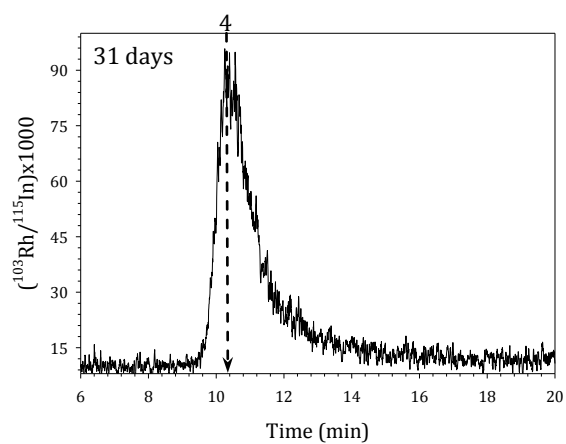
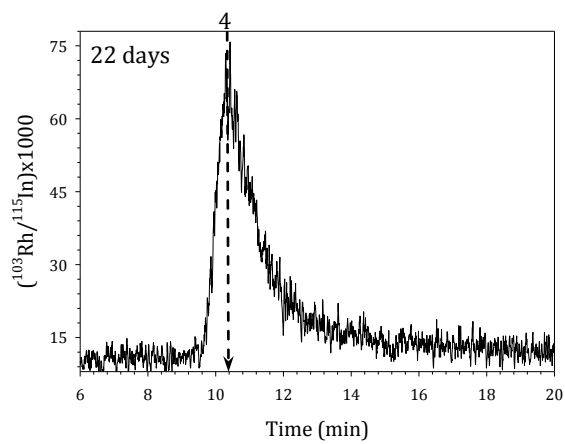
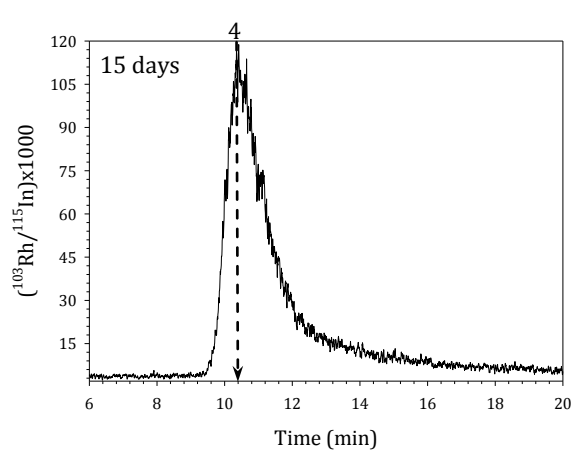
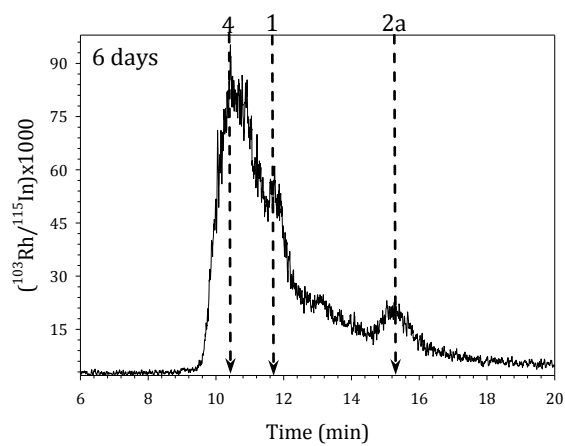
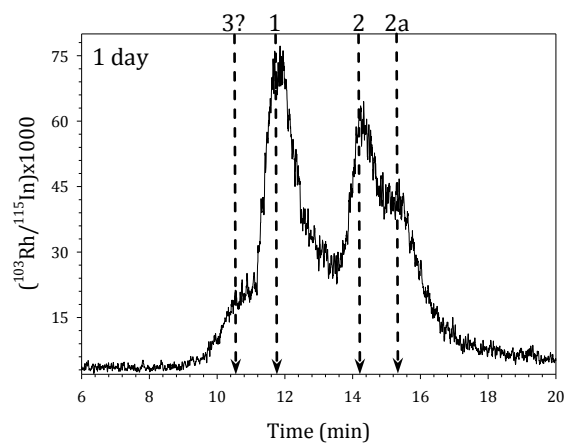
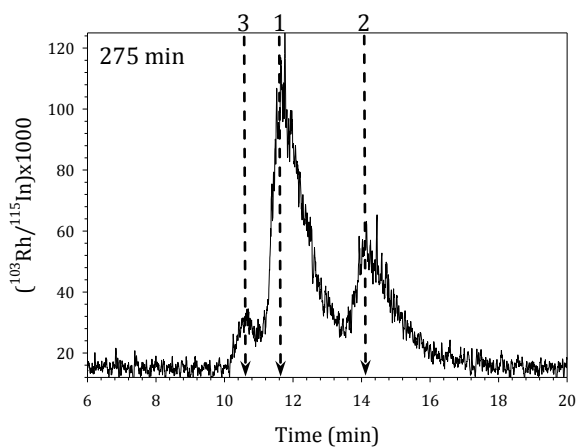
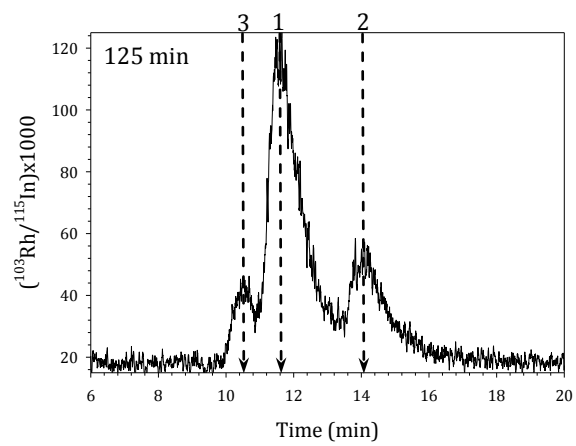
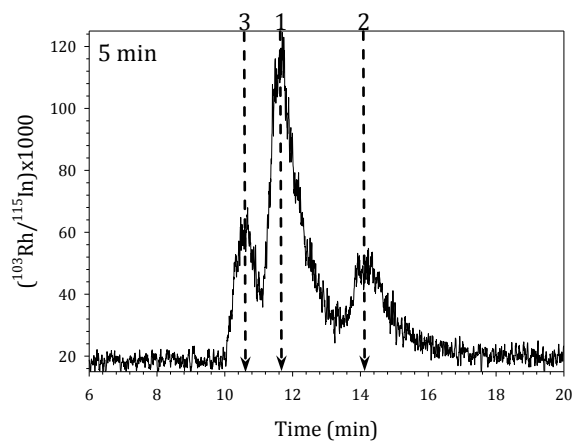


A

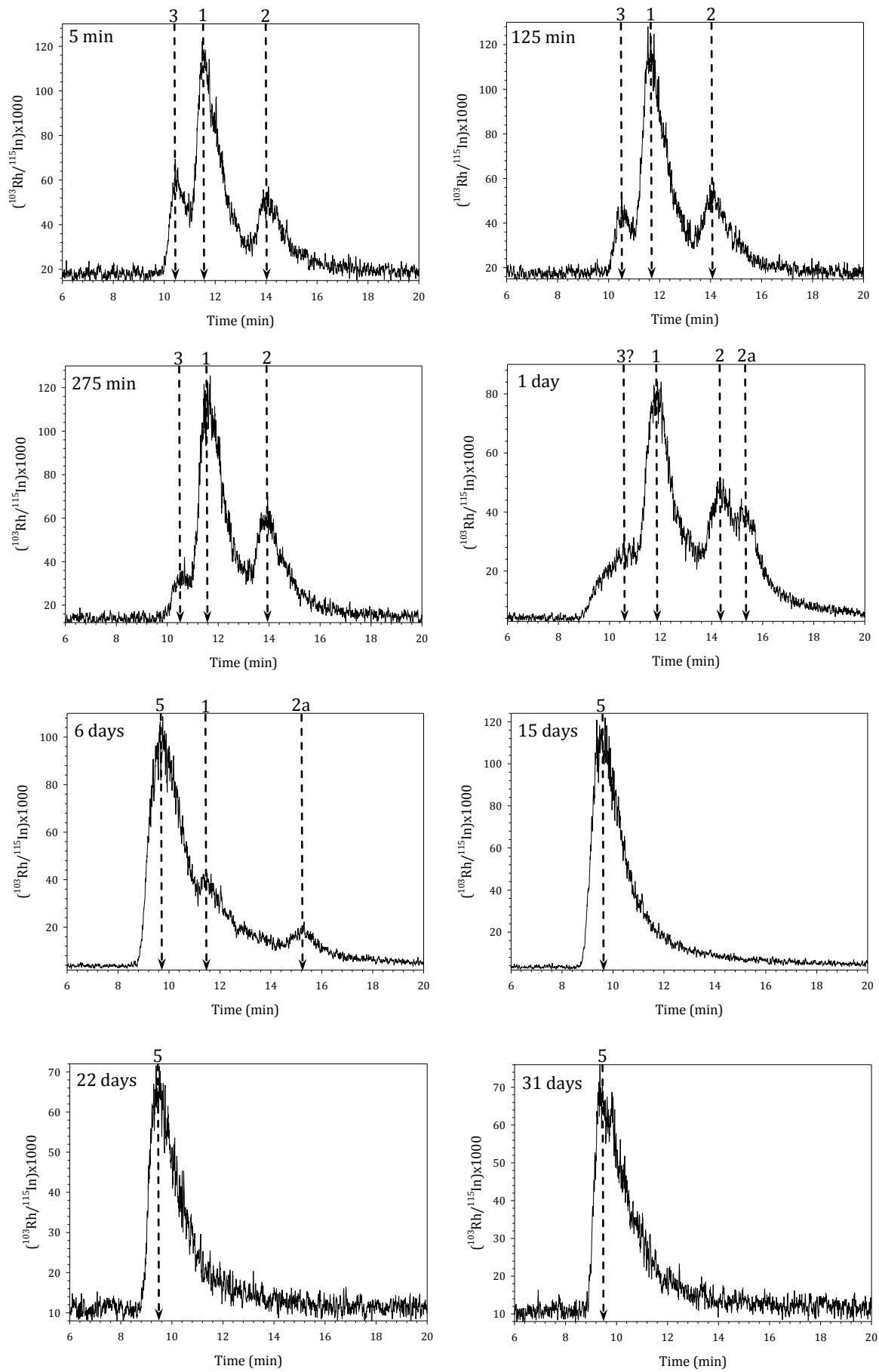




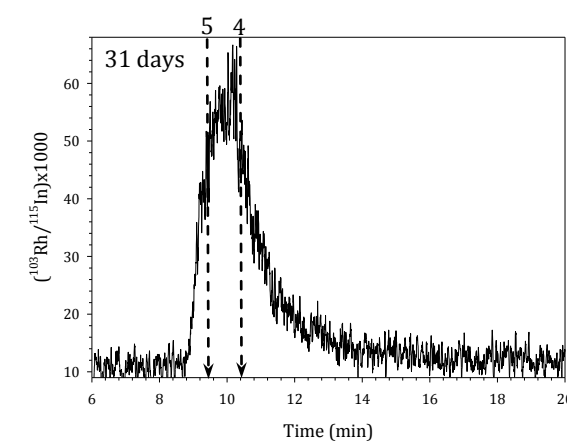
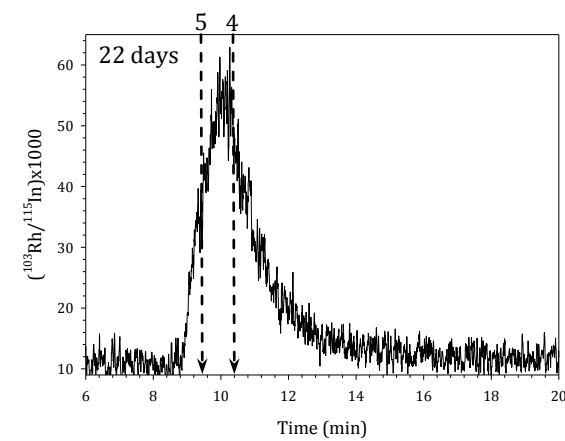
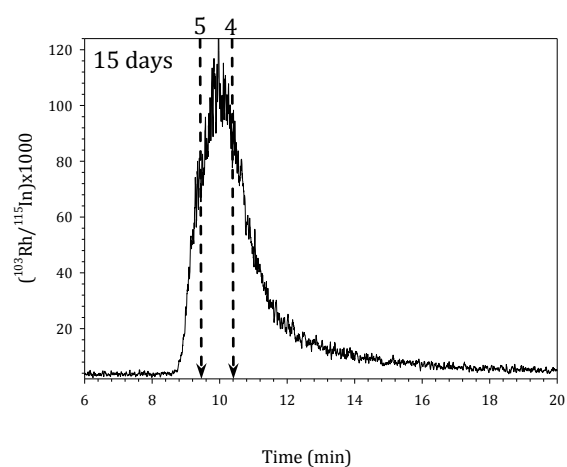
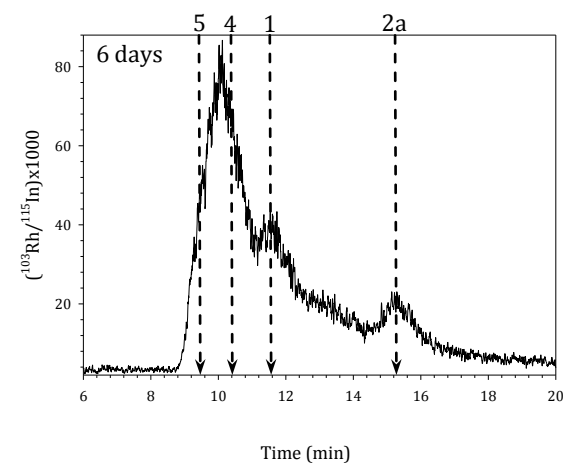
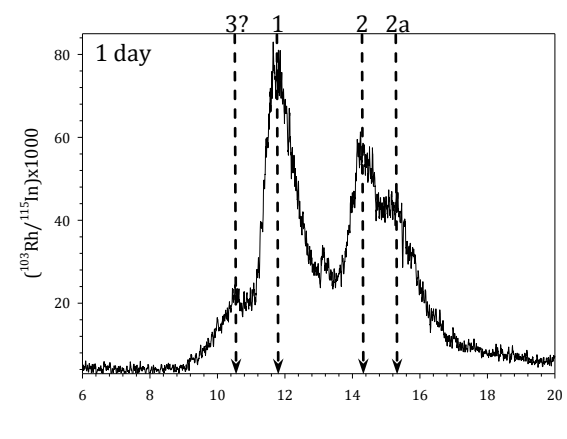
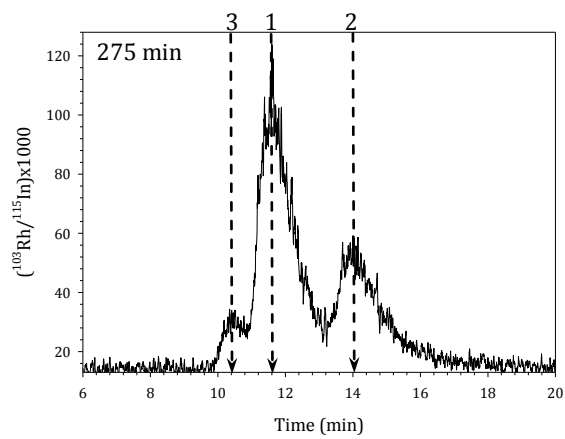
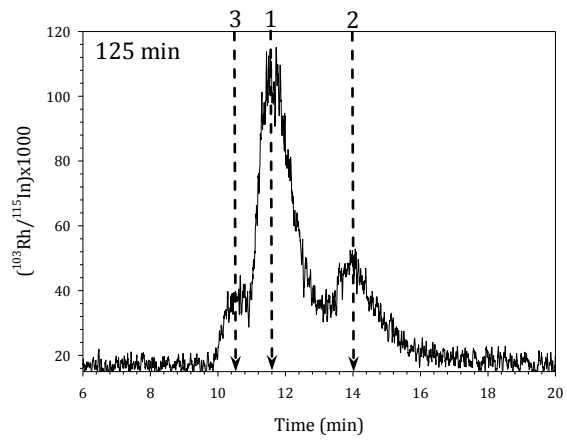
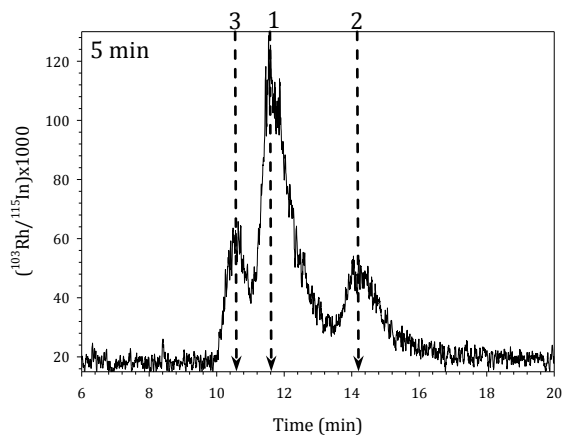
Figure



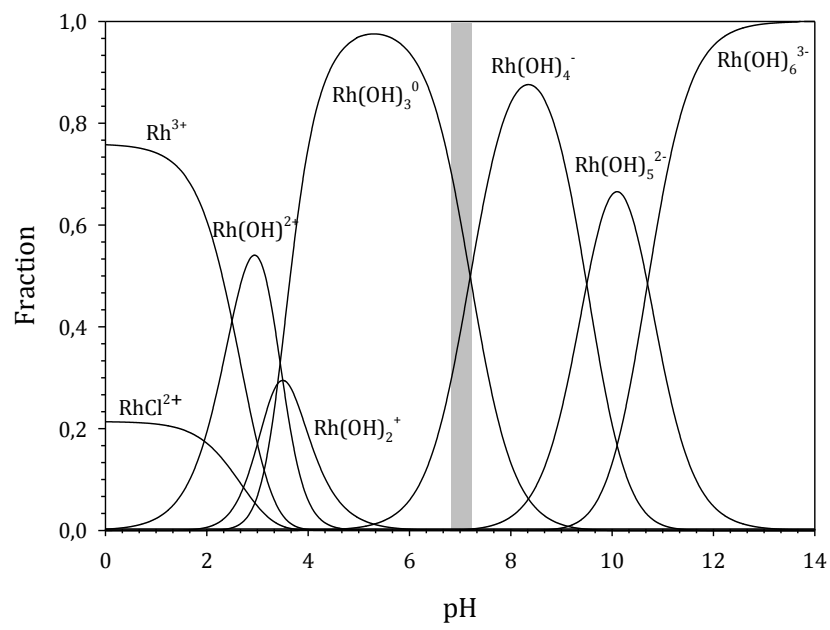
Figure



Figure



Figure



Supporting Information

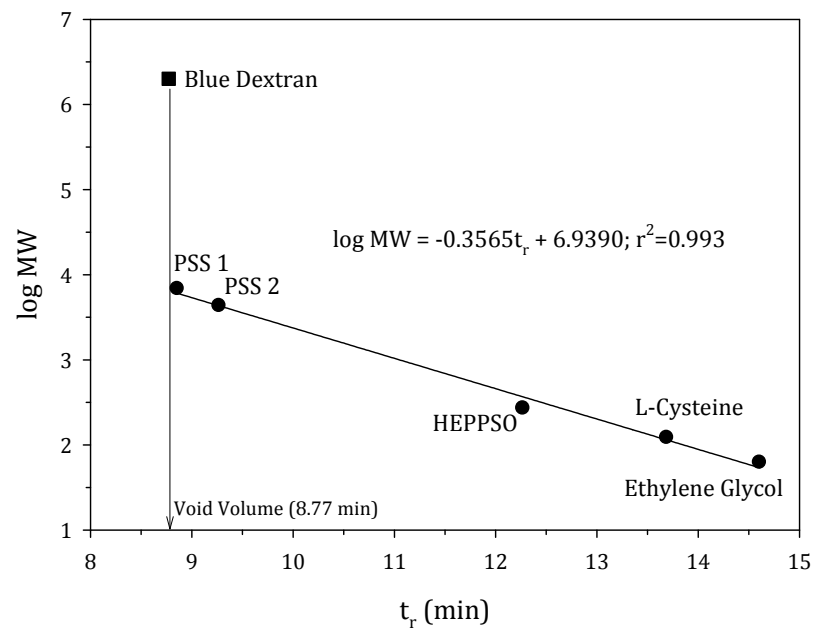


Figure S1. Calibration of the PolySep GFC-P 200 column using a flow rate of 0.8 mL min⁻¹ and a mobile phase prepared in artificial freshwater (Errecalde et al. 1998) with 10 mM KClO₄ + 5 mM phosphate buffer (pH 7.0)

Supporting Information

Table S1. Comparison of cumulative hydrolysis constants for Rh(III) reported in the literature^a.

Reference	T (°C), I	log β ₁ [*]	log β ₂ [*]	log β ₃ [*]	log β ₄ [*]	log β ₅ [*]	log β ₆ [*]	log β _{Rh(OH)₃(s)} [*]
1	25, NaClO ₄	-3.4 (I variable)	-	-	-	-	-	-9.1 (I variable)
2	25, NaClO ₄	-3.2 to -3.4 (I variable)	-	-	-	-	-	-
3	18, HClO ₄ +NaOH	-3.0 (I=0.1)	-6.5 (I=0.1)	-10.5 (I=0.1)	-17.7 (I=0.1)	-26.9 (I=0.1)	-37.2 (I=0.1)	-4.5 (I=0.1)
		-2.5 (I=0) [*]	-6.0 (I=0) [*]	-9.4 (I=0) [*]	-16.6 (I=0) [*]	-26.1 (I=0) [*]	-36.8 (I=0) [*]	-
4	25, -	-2.5 (I=0)	-	-	-	-	-	-

¹Forrester and Ayres (1959); ²Plumb and Harris (1964), Swaminathan and Harris (1966); ³Maslei et al. (1976), (*) extrapolated to zero ionic strength by the authors; ⁴Sassani & Shock (1998), predicted

^aConditional complexation reactions and solubility product are referred to the general form:



References

Forrester JS, Ayres GH (1959) Rhodium(III) in aqueous solutions. *J Phys Chem* 63: 1979-1981.

Maslei, NN, Nabivanets BI, Yantso EA (1976) Mononuclear complexes of rhodium(III). *Ukrainskiy Khimicheskiy Zhurnal* 3: 247-251.

Plumb W, Harris GM (1964) Kinetics of the exchange of water between oxygen-18-labeled solvent and aquorhodium(III) cation. *Inorg Chem* 3: 542-545.

Sassani DC, Shock EL (1998) Solubility and transport of platinum-group elements in supercritical fluids: Summary and estimates of thermodynamic properties for ruthenium, rhodium, palladium, and platinum solids, aqueous ions, and complexes to 1000 °C and 5 kbar. *Geochim Cosmochim Acta* 62: 2643-2671.

Swaminathan K, Harris GM (1966) Kinetics and mechanism of reaction of chloride ion with hexaaquarhodium(III) ion in acidic aqueous solutions. *J Am Chem Soc* 88: 4411-4414.

Supporting Information

Table S2. Comparison of cumulative complexation constants for Rh(III) chlorides and mixed hydroxy-chlorides reported in the literature^a.

Reference	T (°C), I	log β* ₁₀	log β* ₂₀	log β* ₃₀	log β* ₄₀	log β* ₅₀	log β* ₆₀	log β* ₂₁	log β* ₃₁	log β* ₄₁	log β* ₅₁
1	25, -	2.01 (I=0)	3.28 (I=0)	3.30 (I=0)	3.25 (I=0)	-	-	-	-	-	-
2	25,	2.45	4.54	5.92	7.08	8.68	8.36	-	-	-	-
3		2.62	4.38	5.97	7.42	8.79	-	-	-	-	-
4		-	-	-	-	-	-	-	1.1	1.1	1.4
5	25, ≈0	-	-	-	-	-	-	-1.2			

¹Sassani & Shock (1998), predicted; ²Cozzi and Pantani (1958); ³Mihailov et al. (1974), obtained from re-evaluation of data given in Cozzi and Pantani (1958); ⁴Buslaeva and Simanova (1999) and using the chloride stability constants given by Cozzi and Pantani (1958); ⁵Palmer and Harris (1975)

^aConditional complexation reactions are referred to the general form:



References

Buslaeva TM, Simanova SA (1999) Platinum metals in aqueous hydrochloric and chloride solutions: palladium, platinum, rhodium, and iridium. *Russ J Coord Chem* 25: 151-161.

Cozzi D, Pantani F (1958) The polarographic behaviour of rhodium(III) chlorocomplexes. *J Inorg Nucl Chem* 8: 385-398.

Mihailov MH, Mihailova VT, Khalkin VA (1974) A correlation between the overall stability constants of metal complexes – II. *J Inorg Nucl Chem* 36: 115-120.

Palmer DA, Harris GM (1975) Kinetics, mechanism, and stereochemistry of the aquation and chloride anation reactions of fac- and mer-trichlorotriaquorhodium(III) complexes in acidic aqueous solution. A complete reaction scheme for complex ions of the general formula $[\text{RhCl}_n(\text{OH}_2)_{6-n}]^{3-n}$. *Inorg Chem* 14: 1316-1321.

Sassani DC, Shock EL (1998) Solubility and transport of platinum-group elements in supercritical fluids: Summary and estimates of thermodynamic properties for ruthenium, rhodium, palladium, and platinum solids, aqueous ions, and complexes to 1000 °C and 5 kbar. *Geochim Cosmochim Acta* 62: 2643-2671.