CHEMISTRY
A EUROPEAN JOURNAL

DOI: 10.1002/chem.200501410

Cp*Rh-Based Indicator-Displacement Assays for the Identification of Amino Sugars and Aminoglycosides

Friederike Zaubitzer, Andrey Buryak, and Kay Severin*[a]

Abstract: Indicator-displacement assays based on the organometallic complex [{Cp*RhCl$_2$}$_2$] (Cp* = pentamethylcyclopentadienyl) and the dye gallocyanine were used to sense amino sugars and aminoglycosides in buffered aqueous solution by conducting UV-visible spectroscopy. The data of three assays at pH 7.0, 8.0, and 9.0 were sufficient to distinguish between the amino sugars galactosamine, glucosamine, mannosamine and the aminoglycosides kanamycin A, kanamycin B, amikacin, apramycin, paromomycin, and streptomycin. Furthermore, the assays were used to characterize mixtures of aminoglycosides and obtain quantitative information about the respective analytes.

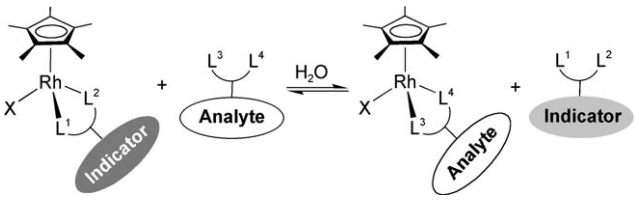
Keywords: amino sugars · aminoglycosides · organometallic complexes · rhodium · sensors

Introduction

Recently, indicator-displacement assays (IDAs) employing synthetic receptors have been used extensively for analytical sensing.[1] In these assays, an indicator competes with the analyte for the noncovalent binding to a receptor. This provides quantitative information about the analyte, given that the free and the bound indicator differ in their optical properties. The noncovalent attachment of the signaling unit makes IDAs very flexible, because the nature of the indicator as well as the indicator/receptor ratio[2] can be varied according to the sensing problem.

Transition-metal complexes with available coordination sites are frequently used as receptors, because they can display a high binding affinity to Lewis basic analytes, even in competitive solvents, such as water. The selectivity of such receptors can be modulated by variation of the steric and electronic properties of permanently attached coligands. So far, transition-metal-based IDAs have been employed successfully to sense phosphates,[3] phosphoesters,[4] cyanide,[5] nitric oxide,[6] carbonate,[7] amino acids,[8,9] citrate,[10] carboxylate anions,[11] and thiols,[12] amongst others.[1]

In the context of investigating applications of organometallic halfsandwich complexes as sensors,[13,14] we have recently reported that Cp*Rh-based IDAs (Cp = pentamethylcyclopentadienyl) can be used to sense amino acids and peptides in aqueous solution (Scheme 1).[15,16] The sensing ensembles were obtained by mixing the organometallic complex [{Cp*RhCl$_2$}$_2$] (**1**) with metal-binding dyes. Complex **1** is commercially available, not air-sensitive and soluble in water. An important advantage over the commonly employed three-dimensional transition-metal complexes is that the Cp*Rh complex displays very high binding constants, in particular for analytes with N- or S-donor ligands.[17] This enables the IDAs to be performed at very low analyte concentrations. Peptides with histidine or methionine residues close to the N-terminus, for example, could be sensed at concentrations as low as 300 nM.[15] An apparent disadvantage of Cp*Rh-based IDAs is that structural modifications of the permanently attached cyclopentadienyl ligand are difficult. The selectivity is, thus, dictated by the intrinsic affinity



Scheme 1. Schematic representation of an indicator-displacement assay based on an organometallic Cp*Rh complex. L = donor group.

[a] F. Zaubitzer, A. Buryak, Prof. K. Severin
Institut des Sciences et Ingénierie Chimiques
École Polytechnique Fédérale de Lausanne (EPFL)
1015 Lausanne (Switzerland)
Fax: (+41) 21-693-9305
E-mail: kay.severin@epfl.ch

of the Cp*Rh fragment for the different analytes. We have shown, however, that this limitation can be compensated by using sensor-array technology.[16]

Here we demonstrate that Cp*Rh-based IDAs can be used to discriminate with high fidelity between simple amino sugars, such as galactosamine and mannosamine, as well as aminoglycosides. Aminoglycosides are an important class of antibiotics that contain two or more amino sugars linked to an aminocyclitol unit by glycosidic bonds. They are particularly active against aerobic, gram-negative bacteria. Despite the introduction of newer, less-toxic antimicrobial agents, aminoglycosides continue to play a useful role in the treatment of serious infections.[18] Due to their pharmacological relevance, there has been tremendous interest in developing analytical tools for these compounds.[19] To obtain quantitative data from complex samples, such as blood or food, immunoassays or LC/MS measurements are commonly performed. For the analysis of aminoglycosides in samples without other interfering analytes, a number of spectroscopic methods have been reported.[19,20] These methods provide quantitative data, but are mostly nonspecific. For example, aminoglycosides have been quantified by fluorescence spectroscopy upon addition of fluorescent probes that react with the primary amine group.[20] The utilization of IDAs for aminoglycoside analysis represents a new approach in this context.[21] As shown below, this method is technically simple, but also very powerful: even structurally similar analytes, such as kanamycin A, kanamycin B, and amikacin, can be distinguished. Furthermore, it is possible to obtain quantitative information and to differentiate mixtures of aminoglycosides from pure samples.

Results and Discussion

The basic requirement for the construction of an IDA that aims to discriminate between different analytes is that the indicator and the various analytes have approximately the same affinity for the receptor. If the relative affinity of the analytes is too low, no significant replacement of the indicator would be achieved. On the other hand, if the relative binding constants of the analytes are much higher than that of the indicator, all analytes would give the same signal. However, only an *approximate* match between the binding affinities is required, as differences can be counterbalanced by adjusting the receptor/indicator/analyte ratio.[2,15]

We initially examined competition reactions of various commercially available dyes and D-mannosamine by using the Cp*Rh complex **1** as the receptor (H_2O, 100 mM phosphate buffer, pH 7.0). A small-scale screening exercise suggested gallocyanine (**2**) as a suitable candidate for the construction of Cp*Rh-based IDAs for amino sugars.[22] Gallocyanine forms a 1:1 complex with the Cp*Rh fragment, as evidenced by results of UV-visible titration experiments (Figure 1). The complexation is accompanied by a significant change in color, with a strong reduction in absorption at 620 nm and a strong increase in the region between 700 and 800 nm. Fitting of the data to a 1:1 binding algorithm gave a complexation constant of $K = 2.3 \times 10^6\ M^{-1}$.[16]

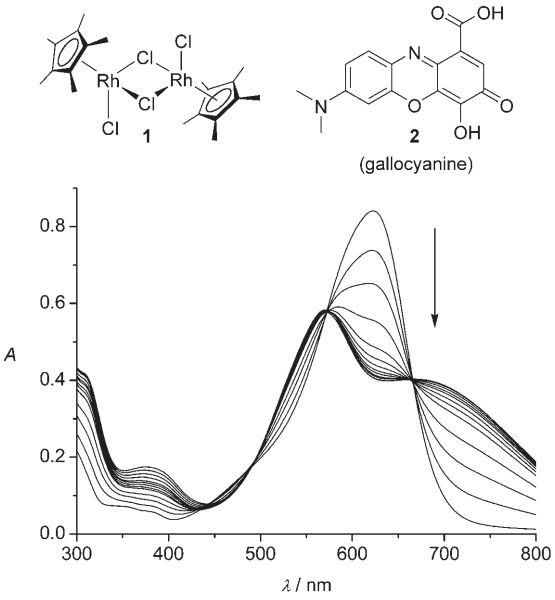


Figure 1. UV-visible absorption spectra of a solution of **2** (25 μM) upon addition of complex **1** (final Rh concentration: 0, 5.0, 10.0, 15.0, 20.0, 22.5, 25.0, 27.5, 30.0, 35.0, 40.0, 45.0, and 50.0 μM). The spectra were recorded in H_2O (100 mM phosphate buffer, pH 7.0) after equilibration.

The UV/Vis spectra obtained from IDAs employing the indicator **2**, the organometallic receptor **1** and the three amino sugars D-glucosamine, D-galactosamine, and D-mannosamine are depicted in Figure 2 ([Rh] = [**2**] = 25 μM, 100 mM phosphate buffer, pH 7.0). Whereas the titration

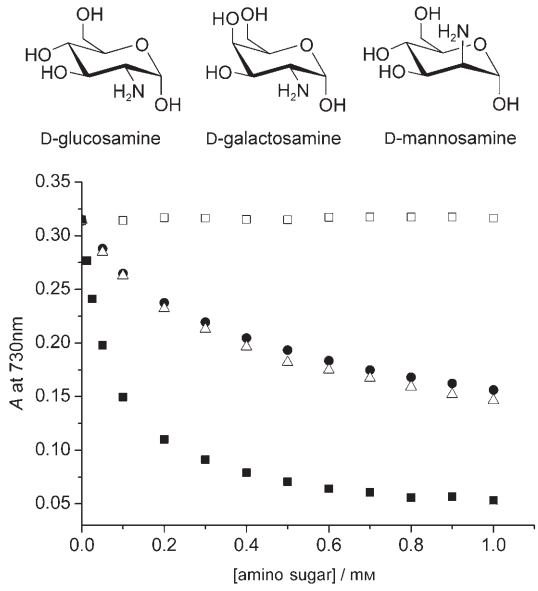


Figure 2. Absorbance at 730 nm for solutions containing complex **1**, the indicator **2**, and various amounts of D-glucosamine (●), D-galactosamine (△), D-mannosamine (■) or N-acetyl-D-glucosamine (□) ([Rh] = [**2**] = 25 μM, 100 mM phosphate buffer, pH 7.0). The data represent averaged values from two independent measurements.

curves of D-glucosamine and D-galactosamine are very similar, D-mannosamine was found to bind more strongly to the Cp*Rh complex. On assumption that the systems can be described by a simple competition reaction, the data was used to calculate the binding constants for the three analytes: K(glucosamine) = 5.6 ($\pm$0.5) $\times 10^4\,M^{-1}$, K(galactosamine) = 6.6 ($\pm$0.5) $\times 10^4\,M^{-1}$, K(mannosamine) = 6.6 ($\pm$0.5) $\times 10^5\,M^{-1}$. Notably, the titration data can be used as calibration curves for quantification of the respective amino sugar within the range 20–400 μM.

To test the selectivity of the assay, analogous titration experiments were performed with N-acetyl-D-glucosamine (Figure 2) and D-glucose. Neither of the two sugars caused a significant displacement of the indicator. This showed that the presence of a free amino group is of importance for the success of the assay. Interestingly, however, cyclohexylamine was not able to displace gallocyanine. Apparently, the amine and at least one of the hydroxy groups of the amino sugars are required for complexation to the Cp*Rh complex. It is likely that five-membered N,O-chelates with hydroxy groups adjacent to the amine are formed. Unfortunately, attempts to obtain decisive information from X-ray crystallographic studies were unsuccessful.

To increase the discriminative power of IDAs, it is possible to use sensor-array technology. In a sensor array, the response of several nonselective sensors, which show a differential response to a given analyte, is used to identify the analyte by a multivariate analysis.[23,24] Due to their intrinsic flexibility, IDAs are ideally suited for the construction of sensor arrays.[16,25] Our particular approach is based on the fact that the response of an IDA can be modulated by a simple change of pH, given that the metal-donor groups of the indicator and the analyte differ in their pK_b values (Scheme 2). For example, if the donor group of the analyte is more basic than that of the indicator, a decrease of the pH will shift the equilibrium in favor of the metal–indicator complex. On the other hand, a more basic pH will favor the metal–analyte complex. Thus, for the construction of a sensor array it is sufficient to perform IDAs at different pH values.

A sensor-array approach of this kind was evaluated for the analysis of amino sugars and aminoglycosides. As analytes, we again used the three amino sugars D-glucosamine, D-galactosamine, and D-mannosamine, as well as the following six aminoglycosides: kanamycin A, kanamycin B, amikacin, apramycin, paromomycin, and streptomycin. The structures of these antibiotics are depicted in Figure 3.

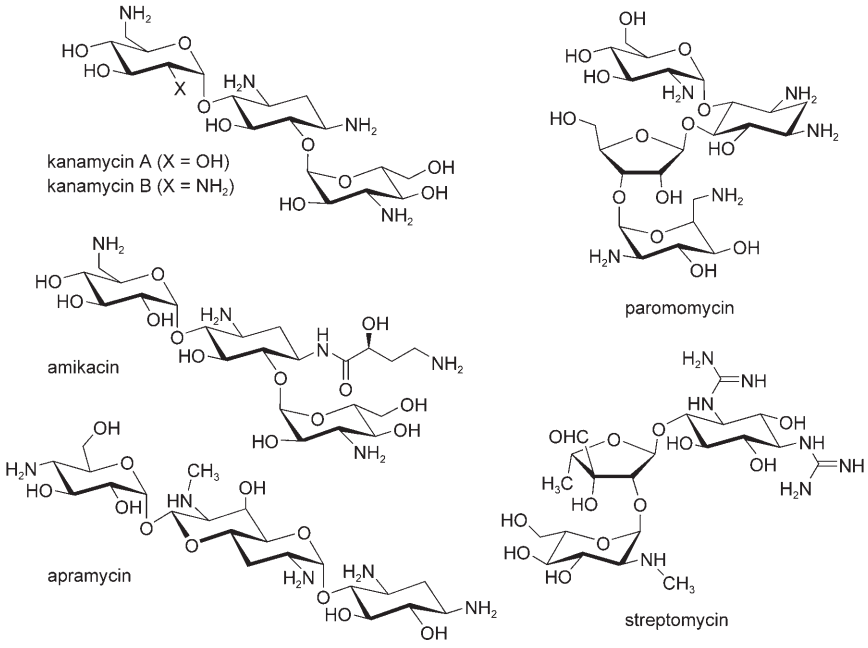
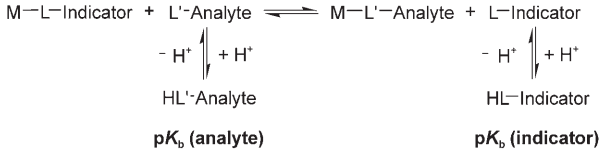


Figure 3. Aminoglycosides used as analytes.

A "mini array" of three IDAs at the pH values 7.0, 8.0, and 9.0 was employed for the analysis. The individual IDAs were performed by mixing solutions of complex **1**, indicator **2**, and the respective analyte ([Rh] = [**2**] = [analyte] = 100 μM, 100 mM phosphate or 2-(cyclohexylamino)ethanesulfonic acid (CHES) buffer). Eight independent measurements were carried out for each analyte at every pH. The data was collected in parallel by using a 96-well plate in combination with a UV-visible microplate reader. The results for the nine different analytes are depicted in Figure 4.

Each sugar gives rise to a characteristic signal pattern. As expected for analytes with donor groups of different basicity,[26] variations are found not only for the absolute values, but also for the relative values. Glucosamine, galactosamine, and paromomycin are difficult to distinguish at pH 7.0 and 8.0, but show a distinct response at pH 9.0. Mannosamine, kanamycin A, and kanamycin B, on the other hand, show



Scheme 2. If the donor groups L and L′ have different basicities, a change in pH will affect the competition between indicator and analyte for coordination to the metal (M).

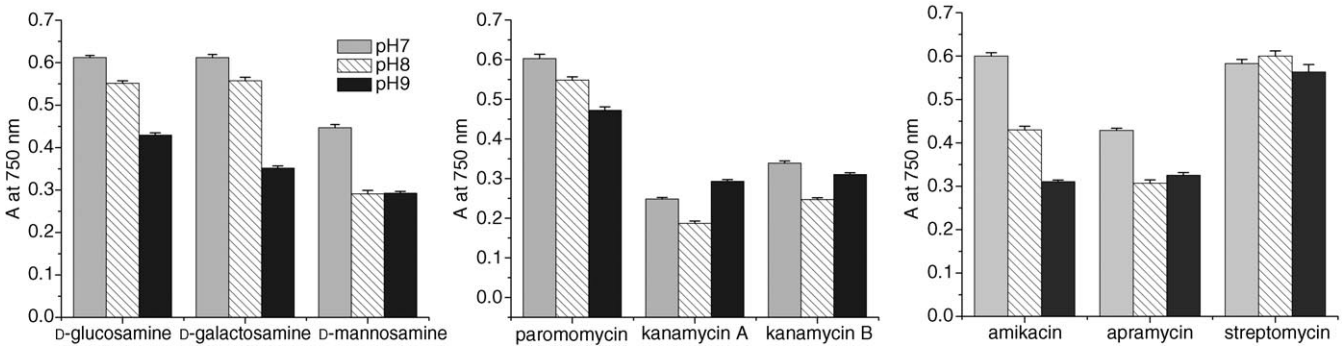


Figure 4. Absorbance at 750 nm for solutions containing complex **1**, the indicator **2**, and the respective amino sugar or aminoglycoside at three different pH values ([Rh] = [**2**] = [analyte] = 100 μM, 100 mM phosphate or CHES buffer). The data represent averaged values from eight independent measurements.

nearly the same absorption at pH 9.0, but differ characteristically at pH 7.0 and 8.0.

To confirm that the data from the IDA mini array was sufficient for the classification of all nine analytes, a linear discriminant analysis (LDA) was performed.[27] A graphic representation of this analysis in the form of a score plot is given in Figure 5. All analytes can clearly be distinguished,

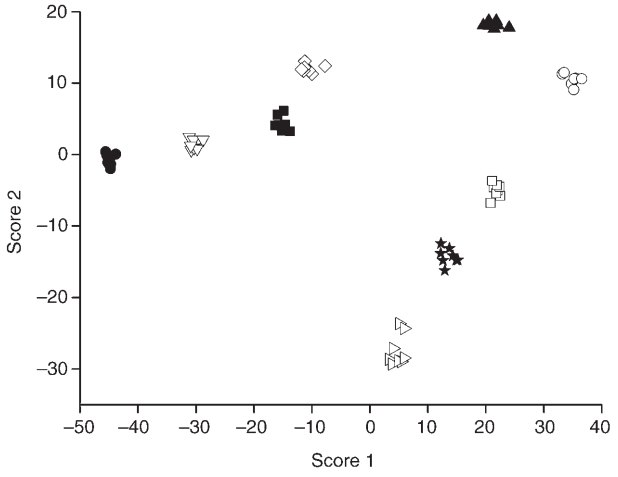


Figure 5. Two-dimensional LDA score plot for the analysis of glucosamine (□), galactosamine (○), mannosamine (◇), kanamycin A (●), kanamycin B (▽), amikacin (▲), apramycin (■), paromomycin (★), and streptomycin (▷).

even closely related compounds, such as kanamycin A and kanamycin B, which differ in only one functional group (OH versus NH_2), or the stereoisomers glucosamine, galactosamine, and mannosamine.

Three factors are likely to be of importance for the differentiation of the analytes: i) the number of potential 1,2-amino alcohol binding sites per analyte; ii) the differences in the pK_b values of the corresponding amino groups, which will influence the binding equilibria as outlined in Scheme 2; iii) the accessibility of the 1,2-amino alcohol binding sites. The overall response of the IDA will depend on all three parameters, which are characteristic features of the respective analyte.

In a second set of experiments, we investigated the influence of the analyte concentration by using the aminoglycosides kanamycin A and kanamycin B as examples. Indicator-displacement assays were again performed at pH 7.0, 8.0, and 9.0. In addition to assays with an analyte concentration of 100 μM, competition reactions with kanamycin concentrations of 50, 80, and 120 μM were carried out (four measurements each). A score plot for the LDA of the resulting data is shown in Figure 6. Interestingly, both the analyte *and* its concentration can be distinguished. Thus, three UV-visible measurements are sufficient to tell apart the closely related antibiotics kanamycin A and kanamycin B, and to obtain an estimate of the concentration within the range 50–120 μM.[28]

We were interested in whether our new methodology was able to distinguish pure samples from mixtures. To address this, IDAs were performed at three pH values (7.0, 8.0, and 9.0) with the aminoglycosides kanamycin A, paromomycin, and amikacin (100 μM) and equimolar mixtures of all three

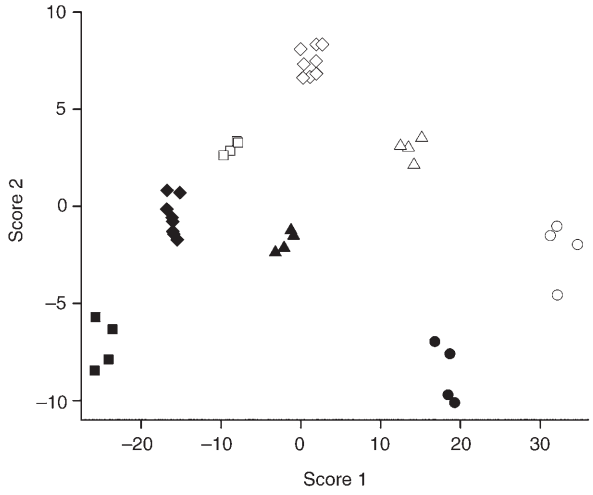


Figure 6. Two-dimensional LDA score plot for the analysis of kanamycin A (filled symbols) and kanamycin B (open symbols) at different concentrations: 50 μM (circle), 80 μM (triangle), 100 μM (rhombus), and 120 μM (square).

combinations (50 μM each). The corresponding score plot is shown in Figure 7. All data sets were well separated, indicating that the mixtures can indeed be differentiated from the pure samples.

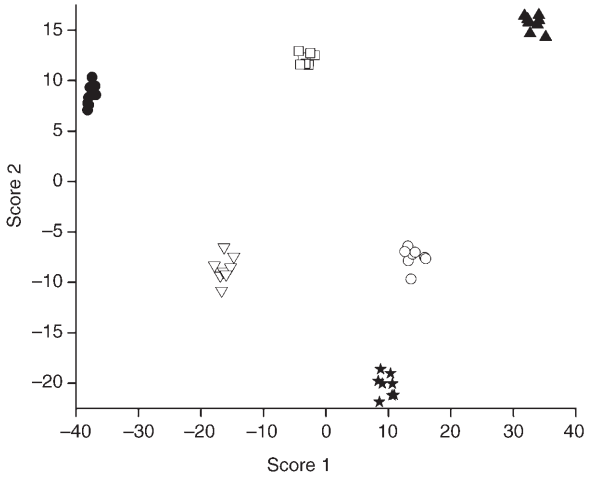


Figure 7. Two-dimensional LDA score plot for the analysis of kanamycin A (●), amikacin (▲), paromomycin (★), and equimolar mixtures of kanamycin A/amikacin (□), amikacin/paromomycin (○), and kanamycin A/paromomycin (▽).

Conclusion

Organometallic complexes have been used increasingly for bioanalytical purposes.[29] The present work describes a new application in this context: the colorimetric sensing of amino sugars and aminoglycosides in aqueous solution by using indicator-displacement assays. The sensing ensemble comprises the complex $[\{Cp^*RhCl_2\}_2]$ and the dye gallocyanine, both of which are commercially available. Measurements at three different pH values were shown to be sufficient to identify amino sugars and aminoglycosides with high fidelity. Furthermore, the assays can be used to characterize mixtures of aminoglycosides and to obtain quantitative information about the respective analytes. The simplicity of the assays should make it appealing for the analysis of these pharmacologically important analytes. From a more general perspective, it is interesting that IDA arrays can be constructed by simply changing the pH. This method should be applicable to other types of IDAs and may provide a means to easily increase the analytical power of such analyses.

Experimental Section

General: The complex $[\{Cp^*RhCl_2\}_2]$ (**1**) was prepared according to a literature procedure[30]. Gallocyanine (hydrochloride) and D-galactosamine (hydrochloride, 99%) were purchased from Acros Organics. D-Glucosamine (hydrochloride, 98%), kanamycin A (sulfate, 97%), and N-acetyl-D-glucosamine (99%) were purchased from Sigma. D-Mannosamine (hydrochloride, 98%), D-glucose (99%), paromomycin (sulfate, 98%), kanamycin B (sulfate, 99%), amikacin (disulfate, 99%), apramycin (sulfate, 95%), streptomycin (sesquisulfate, 95%) as well as CHES and phosphate buffers were purchased from Fluka (BioChemika). All solutions were prepared by using bidistilled water. Either phosphate buffer (100 mM, pH 7.0 or 8.0) or CHES buffer (100 mM, pH 9.0) were employed for stock solutions of complex **1** (0.25 or 0.50 mM) and the dye (1.0 mM). The stock solutions of D-glucosamine, D-galactosamine, D-mannosamine, D-glucose, and N-acetyl-D-glucosamine (5.0 or 10.0 mM) were prepared in phosphate buffer (100 mM, pH 7.0). For sensor-array experiments, stock solutions of all amino sugars and aminoglycosides (1.0 mM) were prepared in pure water. Solutions of gallocyanine were freshly prepared prior to each experiment and the equilibration of reaction mixtures was performed in the dark. All UV/Vis titration curves were recorded by using a Lambda 40 spectrometer (Perkin–Elmer). The array was analyzed by using a PowerWave microplate reader (BIO-TEK) with 96-well plates.

UV/Vis titration experiments with $[\{Cp^*RhCl_2\}_2]$ (1) and gallocyanine (2): Stock solutions of complex **1** and dye **2** in phosphate buffer (100 mM, pH 7.0) were used to prepare 13 solutions of various Cp*Rh concentrations and constant dye concentration (25 μM). The final Cp*Rh concentrations were 0, 5.0, 10.0, 15.0, 20.0, 22.5, 25.0, 27.5, 30.0, 35.0, 40.0, 45.0, and 50.0 μM. After equilibration for 1.5 h in the dark, the absorption curve was recorded within the range 300–800 nm. The binding constant was determined by fitting the titration data to a 1:1 binding algorithm by using the program Gepasi, version 3.30.[31]

UV/Vis titration experiments with D-glucosamine, D-galactosamine, D-mannosamine, D-glucose, and N-acetyl-D-glucosamine: Stock solutions of complex **1**, dye **2**, and the respective analyte in phosphate buffer (100 mM, pH 7.0) were used to prepare a series of solutions with constant Cp*Rh and dye concentrations (25 μM each), but with various analyte concentrations. The final D-glucosamine and D-galactosamine concentrations were 0, 50, 100, 200, 300, 400, 500, 600, 700, 800, 800, and 1000 μM. The final D-mannosamine concentrations were 0, 12.5, 25, 50, 100, 200, 300, 400, 500, 600, 700, 800, 900, and 1000 μM. For D-glucose and N-acteyl-D-glucosamine, the analyte concentration was raised in intervals of 100 μM. In all cases, the dye and the analyte were mixed prior to the addition of complex **1**. After equilibration for 2 h in the dark, the absorption curve was recorded within the range 300–800 nm. The binding constants of the metal–amino sugar complexes were determined indirectly from the binding constant of the Cp*Rh–dye complex ($2.3\times10^6\,M^{-1}$), assuming that the systems can be described by a simple competition reaction. The titration data was fitted by using the program Gepasi, version 3.30.

Sensor arrays—identification of amino sugars and aminoglycosides: Stock solutions of complex **1** (0.50 mM) and dye **2** (1.0 mM) in phosphate buffer (100 mM, pH 7.0 or 8.0) or CHES buffer (100 mM, pH 9.0), along with aqueous solutions of the respective analyte (1.0 mM) were employed. In each well of a 96-well plate, a mixture (200 μL) of complex **1**, the dye **2**, and the respective analyte was prepared (final conc.: [Rh] = [**2**] = [analyte] = 100 μM). In all cases, the dye and the analyte were mixed prior to the addition of complex **1**. After equilibration for 6 h in the dark, the absorption at 750 nm was measured. Eight independent measurements were performed for each amino sugar or aminoglycoside at each pH value. The resulting data was used to perform a linear discriminant analysis by using the statistical software SYSTAT 11.

Sensor arrays—identification of kanamycin A and kanamycin B at various concentrations: Stock solutions of complex **1** (0.50 mM) and dye **2** (1.0 mM) in phosphate buffer (100 mM, pH 7.0 or 8.0) or CHES buffer (100 mM, pH 9.0), along with aqueous solutions of kanamycin A and kanamycin B (1.0 mM) were employed. In each well of a 96-well plate, a mixture (200 μL) of complex **1**, the dye **2**, and the respective analyte was prepared (final conc.: [Rh] = [**2**] = 100 μM; [analyte] = 50, 80, 120 μM). In all cases, the dye and the analyte were mixed prior to the addition of complex **1**. After equilibration for 6 h in the dark, the absorption at 750 nm was measured. Four independent measurements were performed for both aminoglycosides at each pH value. The resulting data in combination with the values obtained for kanamycin A and kanamycin B at a concentration of 100 μM (see above) was used to perform a linear discriminant analysis by using the statistical software SYSTAT 11.

Sensor arrays—discrimination between mixtures of aminoglycosides and pure samples: Stock solutions of complex **1** (0.50 mM) and dye **2** (1.0 mM)

in phosphate buffer (100 mM, pH 7.0 or 8.0) or CHES buffer (100 mM, pH 9.0), along with aqueous solutions of paromomycin, kanamycin A, and amikacin (1.0 mM) were employed. In each well of a 96-well plate, a mixture (200 μL) of complex **1**, the dye **2**, and an equimolar mixture of two aminoglycosides was prepared (final conc.: $[Rh]=[\mathbf{2}]=[analyte]_{total}=100\ \mu M$). In all cases, the dye and the analyte were mixed prior to the addition of complex **1**. After equilibration for 6 h in the dark, the absorption at 750 nm was measured. Eight independent measurements were performed for all three mixtures at each pH value. The resulting data in combination with the values obtained for the pure samples (see above) was used to perform a linear discriminant analysis by using the statistical software SYSTAT 11.

Acknowledgements

This work was supported by the Swiss National Science Foundation, the EPFL, and by a Fellowship of the Dr. Carl Duisberg Foundation..

[1] For reviews, see: a) F. Mancin, E. Rampazzo, P. Tecilla, U. Tonellato, *Chem. Eur. J.* **2005**, *12*, 1844–1854; b) L. Fabbrizzi, M. Licchelli, A. Taglietti, *Dalton Trans.* **2003**, 3471–3479; c) C. Suksai, T. Tuntulani, *Chem. Soc. Rev.* **2003**, *32*, 192–202; d) S. L. Wiskur, H. Aït-Haddou, J. J. Lavigne, E. V. Anslyn, *Acc. Chem. Res.* **2001**, *34*, 963–972.

[2] A. M. Piątek, Y. J. Bomble, S. L. Wiskur, E. V. Anslyn, *J. Am. Chem. Soc.* **2004**, *126*, 6072–6077.

[3] a) R. G. Hanshaw, S. M. Hilkert, J. Hua, B. D. Smith, *Tetrahedron Lett.* **2004**, *45*, 8721–8724; b) S. L. Tobey, E. V. Anslyn, *Org. Lett.* **2003**, *5*, 2029–2031; c) Z. Zhong, E. V. Anslyn, *Angew. Chem.* **2003**, *115*, 3113–3116; *Angew. Chem. Int. Ed.* **2003**, *42*, 3005–3008; d) M. S. Han, D. H. Kim, *Angew. Chem.* **2002**, *114*, 3963–3965; *Angew. Chem. Int. Ed.* **2002**, *41*, 3809–3811; e) L. Fabbrizzi, N. Marcotte, F. Stomeo, A. Taglietti, *Angew. Chem.* **2002**, *114*, 2665–2668; *Angew. Chem. Int. Ed.* **2002**, *41*, 3811–3814.

[4] a) R. G. Hanshaw, E. J. O'Neil, M. Foley, R. T. Carpenter, B. D. Smith, *J. Mater. Chem.* **2005**, *15*, 2707–2713; b) T. Zhang, E. V. Anslyn, *Tetrahedron* **2004**, *60*, 11117–11124.

[5] C.-F. Chow, M. H. W. Lam, W.-Y. Wong, *Inorg. Chem.* **2004**, *43*, 8387–8393.

[6] a) M. H. Lim, S. J. Lippard, *Inorg. Chem.* **2004**, *43*, 6366–6370; b) S. A. Hilderbrand, M. H. Lee, S. J. Lippard, *J. Am. Chem. Soc.* **2004**, *126*, 4972–4987.

[7] L. Fabbrizzi, A. Leone, A. Taglietti, *Angew. Chem.* **2001**, *113*, 3156–3159; *Angew. Chem. Int. Ed.* **2001**, *40*, 3066–3069.

[8] a) J. F. Folmer-Andersen, V. M. Lynch, E. V. Anslyn, *Chem. Eur. J.* **2005**, *11*, 5319–5326; b) C.-F. Chow, M. H. W. Lam, H. Sui, W.-Y. Wong, *Dalton Trans.* **2005**, 475–484; c) M. Bonizzoni, L. Fabbrizzi, G. Piovani, A. Taglietti, *Tetrahedron* **2004**, *60*, 11159–11162; d) M. S. Han, D. H. Kim, *Tetrahedron* **2004**, *60*, 11251–11257; e) C.-F. Chow, B. K. W. Chiu, M. H. W. Lam, W.-Y. Wong, *J. Am. Chem. Soc.* **2003**, *125*, 7802–7803; f) M. A. Hortalá, L. Fabbrizzi, N. Marcotte, F. Stomeo, A. Taglietti, *J. Am. Chem. Soc.* **2003**, *125*, 20–21; g) K. E. S. Dean, G. Klein, O. Renaudet, J.-L. Reymond, *Bioorg. Med. Chem. Lett.* **2003**, *13*, 1653–1656; h) H. Aït-Haddou, S. L. Wiskur, V. M. Lynch, E. V. Anslyn, *J. Am. Chem. Soc.* **2001**, *123*, 11296–11297; i) G. Klein, J.-L. Reymond, *Angew. Chem.* **2001**, *113*, 1821–1823; *Angew. Chem. Int. Ed.* **2001**, *40*, 1771–1773.

[9] For the colorimetric enantiodiscrimination of amino acids, see: J. F. Folmer-Andersen, V. M. Lynch, E. V. Anslyn, *J. Am. Chem. Soc.* **2005**, *127*, 7986–7987.

[10] L. Fabbrizzi, F. Foti, A. Taglietti, *Org. Lett.* **2005**, *7*, 2603–2606.

[11] M. Boiocchi, M. Bonizzoni, L. Fabbrizzi, G. Piovani, A. Taglietti, *Angew. Chem.* **2004**, *116*, 3935–3940; *Angew. Chem. Int. Ed.* **2004**, *43*, 3847–3852.

[12] D. H. Kim, M. S. Han, *Bioorg. Med. Chem. Lett.* **2003**, *13*, 2543–2546.

[13] For (arene)Ru complexes that can be used to sense lithium ions, see: a) Z. Grote, R. Scopelliti, K. Severin, *J. Am. Chem. Soc.* **2004**, *126*, 16959–16972; b) Z. Grote, M.-L. Lehaire, R. Scopelliti, K. Severin, *J. Am. Chem. Soc.* **2003**, *125*, 13638–13639; c) H. Piotrowski, K. Severin, *Proc. Natl. Acad. Sci. USA* **2002**, *99*, 4997–5000; d) H. Piotrowski, G. Hilt, A. Schulz, P. Mayer, K. Polborn, K. Severin, *Chem. Eur. J.* **2001**, *7*, 3197–3208; e) H. Piotrowski, K. Polborn, G. Hilt, K. Severin, *J. Am. Chem. Soc.* **2001**, *123*, 2699–2700.

[14] For a Cp*Ir complex that can be used to sense fluoride ions, see: M.-L. Lehaire, R. Scopelliti, H. Piotrowski, K. Severin, *Angew. Chem.* **2002**, *114*, 1477–1480; *Angew. Chem. Int. Ed.* **2002**, *41*, 1419–1422.

[15] A. Buryak, K. Severin, *Angew. Chem.* **2004**, *116*, 4875–4878; *Angew. Chem. Int. Ed.* **2004**, *43*, 4771–4774.

[16] A. Buryak, K. Severin, *J. Am. Chem. Soc.* **2005**, *127*, 3700–3701.

[17] Despite the high binding constants, ligand-exchange reactions at the three coordination sites opposite to the Cp* ligand are relatively fast: L. Helm, A. E. Merbach, *Chem. Rev.* **2005**, *105*, 1923–1959.

[18] S. B. Vakulenko, S. Mobashery, *Clin. Microbiol. Rev.* **2003**, *16*, 430–450.

[19] For a review, see: D. A. Stead, *J. Chromatogr. B* **2000**, *747*, 69–93.

[20] a) M. Rizk, S. S. Toubar, M. S. Abdel-Mottaleb, H. I. Ali, *Bull. Fac. Pharm.* **2003**, *41*, 141–148; b) Y. El-Shabrawy, *Spectrosc. Lett.* **2002**, *35*, 99–109; c) J. Kusnir, K. Barna, *Z. Anal. Chem.* **1974**, *271*, 288.

[21] For the detection of aminoglycosides by HPLC in combination with a ligand displacement reaction, see: M. Yang, S. A. Tomellini, *J. Chromatogr. A* **2001**, *939*, 59–67.

[22] Other indicators tested include arsenazo I, celestin blue, and Chicago sky blue. Arsenazo I was disregarded because amino sugars were not able to replace it from the Cp*Rh complex and celestin blue was rejected because of solubility problems. Chigaco sky blue is potentially suited for amino sugar IDAs, but it was found to bind more than one Cp*Rh fragment, which would complicate the analysis.

[23] For reviews of sensor arrays, see: a) A. T. Wright, E. V. Anslyn, *Chem. Soc. Rev.* **2006**, *35*, 14–28; b) K. L. Brogan, D. R. Walt, *Curr. Opin. Chem. Biol.* **2005**, *9*, 494–500; c) D. James, S. M. Scott, Z. Ali, W. T. O'Hare, *Microchim. Acta* **2005**, *149*, 1–17; d) J. J. Lavigne, E. V. Anslyn, *Angew. Chem.* **2001**, *113*, 3212–3225; *Angew. Chem. Int. Ed.* **2001**, *40*, 3119–3130; e) K. J. Albert, N. S. Lewis, C. L. Schauer, G. A. Sotzing, S. E. Stitzel, T. P. Vaid, D. R. Walt, *Chem. Rev.* **2000**, *100*, 2595–2626.

[24] For recent publications concerning optical sensor arrays, see: a) C. Zhang, K. S. Suslick, *J. Am. Chem. Soc.* **2005**, *127*, 11548–11549; b) N. A. Rakow, A. Sen, M. C. Janzen, J. B. Ponder, K. S. Suslick, *Angew. Chem.* **2005**, *117*, 4604–4608; *Angew. Chem. Int. Ed.* **2005**, *44*, 4528–4532; c) K. S. Suslick, N. A. Rakow, A. Sen, *Tetrahedron* **2004**, *60*, 11133–11138; d) L. Baldini, A. J. Wilson, J. Hong, A. D. Hamilton, *J. Am. Chem. Soc.* **2004**, *126*, 5656–5657; e) E. A. Baldauff, J. M. Buriak, *Chem. Commun.* **2004**, 2028–2029; f) M. N. Stojanović, E. G. Green, S. Semova, D. B. Nikić, D. W. Landry, *J. Am. Chem. Soc.* **2003**, *125*, 6085–6089; g) A. P. Goodey, J. T. McDevitt, *J. Am. Chem. Soc.* **2003**, *125*, 2870–2871; h) A. Goodey, J. J. Lavigne, S. M. Savoy, M. D. Rodriguez, T. Curey, A. Tsao, G. Simmons, J. Wright, S.-J. Yoo, Y. Sohn, E. V. Anslyn, J. B. Shear, D. P. Neikirk, J. T. McDevitt, *J. Am. Chem. Soc.* **2001**, *123*, 2559–2570; i) N. A. Rakow, K. S. Suslick, *Nature* **2000**, *406*, 710–713.

[25] a) A. T. Wright, M. J. Griffin, Z. Zhong, S. C. McCleskey, E. V. Anslyn, J. T. McDevitt, *Angew. Chem.* **2005**, *117*, 6533–6536; *Angew. Chem. Int. Ed.* **2005**, *44*, 6375–6378; b) N. T. Greene, K. D. Shimizu, *J. Am. Chem. Soc.* **2005**, *127*, 5695–5700; c) S. C. McCleskey, M. J. Griffin, S. E. Schneider, J. T. McDevitt, E. V. Anslyn, *J. Am. Chem. Soc.* **2003**, *125*, 1114–1115.

[26] For the acid-base properties of aminoglycosides, see: H. Kozłowski, T. Kowalik-Jankowska, M. Jeżowska-Bojczuk, *Coord. Chem. Rev.* **2005**, *249*, 2323–2334.

[27] P. C. Jurs, G. A. Bakken, H. E. McClelland, *Chem. Rev.* **2000**, *100*, 2649–2678.

[28] For concentrations of more than 120 μM, a simultaneous identification and quantification was no longer possible.

[29] a) G. Jaouen, *Bioorganometallics*, Wiley-VCH, Weinheim, **2005**; b) D. R. van Staveren, N. Metzler-Nolte, *Chem. Rev.* **2004**, *104*, 5931–5985; c) R. H. Fish, G. Jaouen, *Organometallics* **2003**, *22*, 2166–2177; d) K. Severin, *Coord. Chem. Rev.* **2003**, *245*, 3–10; e) M. Salmain, G. Jaouen, *C. R. Chimie* **2003**, *6*, 249–258; f) W. Beck, K. Severin, *Chem. Unserer Zeit* **2002**, *36*, 356–365; g) N. Metzler-Nolte, *Angew. Chem.* **2001**, *113*, 1072–1076; *Angew. Chem. Int. Ed.* **2001**, *40*, 1040–1043.

[30] C. White, A. Yates, P. M. Maitlis, *Inorg. Synth.* **1992**, *29*, 228–234.

[31] a) P. Mendes, *Comput. Appl. Biosci.* **1993**, *9*, 563–571; b) P. Mendes, *Trends Biochem. Sci.* **1997**, *22*, 361–363.

Received: November 12, 2005
Published online: March 7, 2006

