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Molecularly imprinted polymer sensor arrays†

Nathaniel T. Greene, Stephen L. Morgan and Ken D. Shimizu*
Department of Chemistry and Biochemistry, University of South Carolina, Columbia, USA.
E-mail: shimizu@mail.chem.sc.edu; Fax: 803 777 9521; Tel: 803 777 6523

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An eight channel molecularly imprinted polymer sensor array was prepared that was able to differentiate six different aryl amine analytes, including diastereomers with 94% accuracy.

Sensor arrays have been shown to be a very practical and effective strategy for engineering sensors that possess high levels of selectivity and discrimination.[1] Examples include natural systems such the human tongue and synthetic systems such as "electronic noses". A limitation of this method, however, is the requirement of synthesizing a large number of sensing elements each possessing differential selectivity patterns. Presented is the extension of the sensor array strategy to molecularly imprinted polymers (MIPs).[2] The use of MIPs in this format has the potential to rapidly and rationally generate sensor arrays based on MIP sensing elements (Fig. 1).

MIPs have been shown to be easily tailored with selectivity for a wide range of analytes and demonstrate high thermal and chemical stabilities.[3] MIPs are highly crosslinked polymer matrices formed in the presence of a template molecule. The removal of the template then leaves a cavity with shape and functional group complementary to the template molecule. Due to their attractive characteristics MIPs have found application in sensing, chromatographic separations, and catalysis. They are also notable for being quickly and inexpensively generated from a common polymer matrix, and thus MIPs appear to be well-suited for use in a sensor array format. The sensor array format also can enhance the utility of MIPs as sensors. Combining multiple sensors together can compensate for many of the limitations in binding of MIPs such as high levels of cross-reactivity and low overall affinities.[4] In the array, individual sensors may show high levels of cross reactivity and poor selectivity but as long as the signal for one or more sensors in the array is different then a unique pattern or 'fingerprint' will be generated for each analyte.[5]

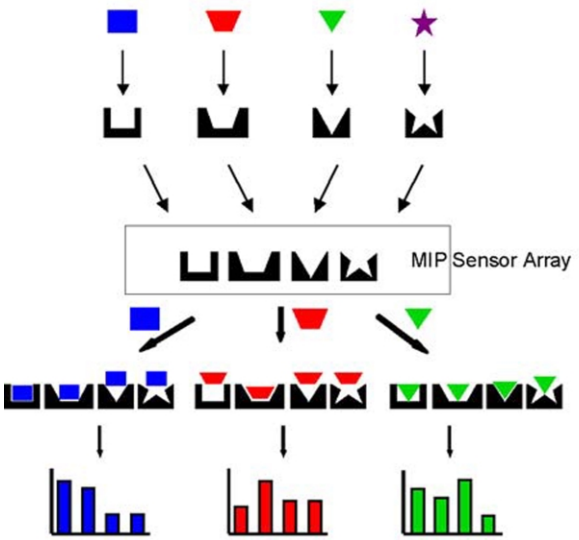


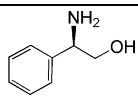
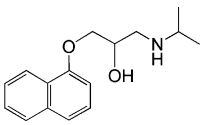
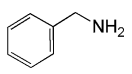
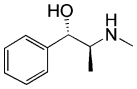
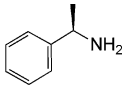
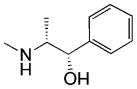
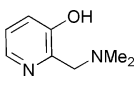
Fig. 1 Four imprinted polymers tested in an array against three different analytes to produce unique patterns for recognition.

To demonstrate the utility and advantages of MIP-based sensor arrays, an eight-channel sensor array was synthesized and tested for its ability to differentiate six different aryl amines. First, eight different polymers (**P0**–**P7**) were synthesized using standard MIP preparation methods (Table 1).[6] A methacrylic acid/ethylene glycol dimethacrylate 20 : 80 mixture was polymerized in toluene (AIBN, UV, 0–15 °C) in the absence (**P0**) and presence (**P1**–**P7**) of seven different aryl amines template molecules (**A1**–**A7**). These template molecules included amines with biological activity including propranolol (**A1**) and diastereomers such as ephedrine (**A3**) and pseudoephedrine (**A2**). The polymer monoliths were individually ground to a fine powder and were then extensively washed by soxhlet extraction to remove template and unreacted monomers.

The affinity of each polymer for six different analytes (**A1**–**A6**) was tested by shaking a constant weight of polymer with 3 mM solutions of each analyte in acetonitrile.‡ The response was measured as the ratio of absorbances $((A_0 - A_i)/A_0)$ at 258 nm of the solutions before and after equilibration with each of the eight individual polymers. Thus, for each analyte, eight different values were measured which are shown as lines on the plot in Fig. 2. Each analyte was tested five separate times against the eight-channel sensor array.

Polymers (**P1**–**P7**) made in the presence of template molecule appear to be imprinted. This is seen by the higher $((A_0 - A_i)/A_0)$ values for the imprinted polymers **P1** through **P7** in comparison to the non-imprinted polymer **P0**. However, the individual imprinted polymers show poor overall selectivity as the imprinted polymers show higher affinity not only for their template molecules but also for the other structurally similar analytes. This cross reactivity combined with the volume of data (eight measurements for each analyte) generated complex patterns with no easily distinguishing features for each analyte.

Table 1 Copolymers **P0** to **P7** of ethylene glycol dimethacrylate and methacrylic acid (80 : 20) formed in the presence of template molecules **A1** to **A7**[a]

Polymers	Analyte/Template		Polymers	Analyte/Template	
P0	**None**		**P4**		**A4**
P1		**A1**[a]	**P5**		**A5**
P2		**A2**	**P6**		**A6**
P3		**A3**	**P7**		**A7**

[a] **A1** through **A7** are (±)-propranolol, (+)-pseudoephedrine, (−)-ephedrine, *R*-(−)-2-phenylglycinol, benzylamine, α-methylbenzylamine, and 2-(dimethylaminomethyl)-3-hydroxypyridine, respectively.

† Electronic supplementary information (ESI) available: experimental details. See http://www.rsc.org/suppdata/cc/b4/b401677g/

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To deconvolute similar and overlapping patterns as well as to filter out random noise, multivariate analysis was applied. This reduced the eight dimensional data set into a more manageable two dimensional data set which still contained as much of the distinguishing signals of the original data set as possible.[7] Specifically, linear discriminant (LDA) was used to transform the data set into a more visually manageable two dimensional plot (Fig 3). LDA was chosen over the more common principal component analysis (PCA) as it produced greater differentiation and less overlap between groups. Each axis of the LDA plot contains linear combinations of the original eight-dimensional data set weighted by coefficients that produce the greatest differentiation between the different analytes. Each point in the LDA plot, therefore, represents the response of the entire eight-channel MIP sensor array for a single analyte.

This analysis demonstrates that the MIP sensor array is generating unique binding patterns for all six analytes. The replicate data points for each analysis are clustered together and equally importantly, these groupings are separate from one another. This initial data can be treated as a training set and the corresponding LDA plot as the calibration matrix. An unknown would be tested against the MIP sensor array and the eight-channel data processed using the previously derived LDA coefficients or loadings. This data point is then plotted on the LDA plot and its identity is selected based on proximity to the previously measured analytes.

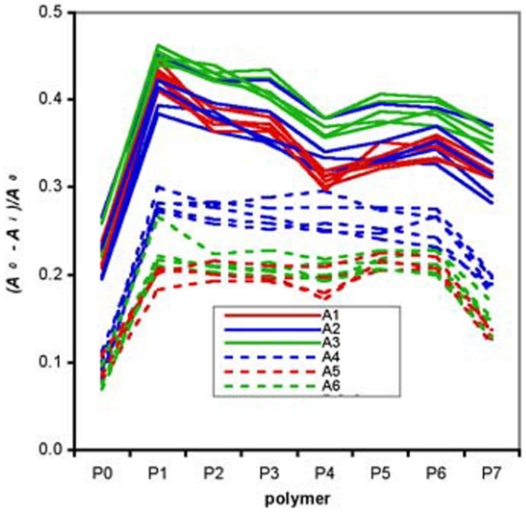

Fig. 2 Plot of the response of the six analytes (3 mM solutions in CH_3CN) tested in replicate (5 times) against the eight-channel MIP array.

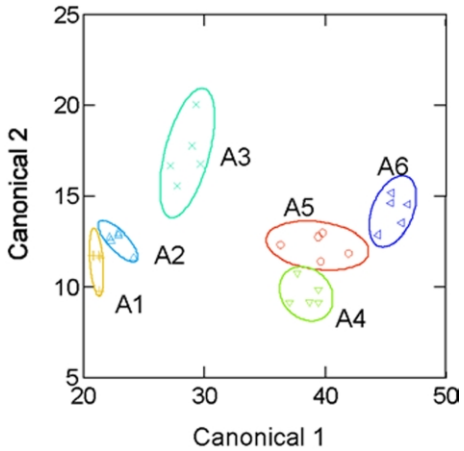

Fig. 3 Two-dimensional LDA plot of the six analytes tested against the MIP array.

To assess the accuracy of MIP sensor array and of the LDA analysis the existing data set was treated as if one of the measurements was an unknown. This data was excluded from training set and a new LDA plot generated. The excluded analyte was then replotted on the abbreviated LDA plot and classified. Using this 'jack-knife' analysis, the MIP array was able to correctly classify 34 out of 36 measured samples, which is an accuracy rate of 94%.

This work demonstrates the potential of template based synthesis methods such as molecularly imprinted polymers to rapidly prepare recognition elements for the sensor array format. Using a molecular imprinting strategy also has the advantage that the individual recognition elements can be rationally designed to have the requisite differential selectivity and can be specifically tailored to the specific analytes being measured. A limitation of this study is that it requires the analytes to have a spectroscopic handle (absorbance at 258 nm).[8] We are in the process of removing this limitation by using dye displacement from the array to measure binding. This allows the MIP array to assay analytes lacking a chromophore and gives a common spectroscopic signal for all analytes. The progress on this work will be reported in due course.

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Notes and references

‡ Analyte **A7** was not tested because of its poor overall affinity and extremely high extinction coefficient, which made analysis of **A7** under similar conditions to the other analytes impossible.

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