Atrazine-selective Polymer Prepared by Molecular Imprinting Technique

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A synthetic atrazine-selective polymer was prepared by molecular imprinting technique. Liquid-chromatographic (LC) tests were performed using both the polymer imprinted against atrazine and a reference non-imprinted polymer as LC stationary phases. Atrazine was retained 13-fold longer in the imprinted polymer than in the non-imprinted polymer.

Recently the pollution of water resources by herbicides and pesticides has drawn renewed attention, due to the extensive use of such chemicals in agriculture but also for the maintenance of recreational facilities such as golf links and gardens. Currently pollutants are routinely detected using gas- or liquid-chromatography[1] and in only a few cases using special chemical or biochemical assays (*e.g.* enzyme linked immunosorbent assays).[2] The available routine chromatographic methods are limited to laboratory analysis, due to the size of equipment. For rapid on-site field screening, assay-type analytical procedures can be designed to use small sometimes even sensor-like equipment. Furthermore these methods mostly show high selectivity for different pollutants, due to the highly specific binding characteristics of antibodies. The stability, price and availability of antibodies, however, can be serious disadvantages of these systems.

Artificial antibody mimics[3] using molecular imprinting technique,[4] as presented by Mosbach's group some years ago, have the potential to overcome these restrictions. For this reason we decided to test the applicability of these stable artificial antibodies as novel affinity ligand for rapid and convenient pesticide analysis. As model system of practical importance we used the herbicide atrazine (2-chloro-4-ethylamino-6-isopropylamino-s-triazine) **1** for the preparation and chromatographic test of the molecularly imprinted atrazine-selective polymer.

For the preparation of atrazine-selective polymer (AtP), 0.360g of atrazine **1**, the template was dissolved in 25 ml chloroform. The functional monomer methacrylic acid (0.575g), the crosslinking monomer ethyleneglycol dimethacrylate (9.35g) and the initiator 2,2'-azobisisobutyronitrile (0.12 g) were added. For the reference non-imprinted polymer (RfP) the same recipe was used, without the addition of the template. Polymerization was carried out with UV light irradiation in a water bath (approx. 0°C). The obtained polymer was crushed, ground and sieved (25 μm) to obtain regularly sized particles. The resulting particles were packed in LC columns (150 mm x 4.6 mm i.d.) and then washed with methanol-acetic acid (8:2, v/v) to remove the template. Isocratic LC analysis was performed with acetonitrile as the eluent at a flow rate of 1.0 ml/min, and detection at 260 nm. The sample size was 20 μl and the concentration 0.2 mM. Acetic acid as a void marker was used to estimate affinities of the polymers.

As shown in Figure 1, atrazine **1** was retained 14.2 min

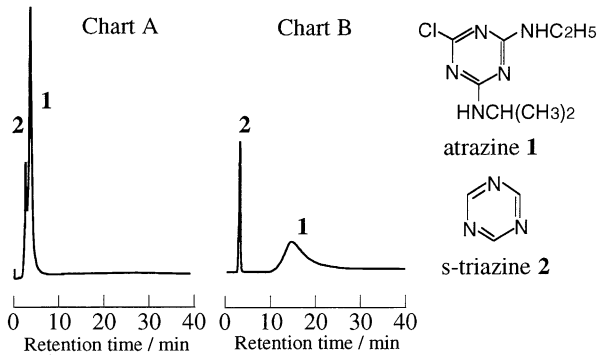

Figure 1. Elution profiles of atrazine **1** and s-triazine **2** in the reference non-imprinted polymer RfP (chart A) and the atrazine-imprinted polymer AtP (chart B).

(k'=4.6) in AtP, while it eluted at 3.4 min (k'=0.35) in RfP. As the both polymers were made of the same chemical composition, it would be reasonable to conclude that carboxylic residues and three dimensional polymer network of the imprinted polymer were arranged suitably to interact with the template molecule as reported in the prior works.[4] Poor retention was observed for s-triazine **2** in both AtP and RfP. This result suggests that the two amino-groups of **1** are important groups for molecular recognition. Triazine-based herbicides, simazine and ametryn, were also retained about 70 % and 60 % in AtP, respectively, compared to atrazine, while almost no retention (< 5 %) was observed for other herbicides of unrelated structures.

Molecular imprinting technique enabled a simple preparation of the stable herbicide-selective material. Furthermore the analytical procedure is performed using organic solvents, as optimal binding of the herbicide to the imprinted polymer is achieved in organic solvents. This is a major advantage compared to enzyme-immuno assays run in aqueous solution, as most of the widely used herbicides exhibit much higher solubility in organic solvents. Therefore we believe that this system may be used for various herbicide-detecting systems instead of antibodies or conventional non-specific stationary phases. In our laboratory detailed polymer characterization and development of a herbicide-sensing system with this polymer are currently underway.

References and Notes

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