Supporting Information

for

Benzoboroxole-functionalized monolithic column for the selective enrichment and separation of cis-diol containing biomolecules

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Experimental sections

1. Materials and Instrumentation

1.1 Materials.

o-Benzotriazol-1-yl-*N,N,N′,N′*-tetramethyluronium hexafluorophosphate (HBTU), *N,N*-diisopropylethylamine (DIPEA), methyl 3-bromo-4-methylbenzoate, dodecanol, catechol, adenosine, 1-hydroxy-7-azabenzotriazole (HOAt), *N,N*-methylenebisacylamide (MBAA) and 2'-deoxyadenosine were purchased from Alfa Aesar (Tianjin, China). Ribonuclease B (RNase B), horseradish peroxidase (HRP), γ-methacryloxypropyltrimethoxysiliane (γ-MAPS), ribonuclease A (RNase A), cytochrome C, β-lactoglubin, glycidyl methacrylate (GMA), HPLC-grade acetonitrile, cetyltrimethylammonium bromide (CTAB) and all standard nucleosides, except adenosine and 2'-deoxyadenosine, were purchased from Sigma (St. Louis, MO). Anti-alpha fetoprotein (anti-AFP) monoclonal antibody, anti-carcinoembryonic antigen (CEA) polyclonal antibody and anti-prostate specific antigen (anti-PSA) monoclonal antibody were purchased from Shuangliu Zhenglong Biochemical Products Laboratory (Chengdu, China). Fused-silica capillary with 150 μm i.d was from the Reafine Chromatography (Hebei, China). Water used in all experiments was purified by a Milli-Q system (Millipore, Milford, MA). All other reagents were of analytical grade. Before use, 2,2-azobisisobutyronitrile (AIBN) was recrystallized from methanol and *N,N*-dimethylformamide (DMF) was dried through CaH_2, followed by distillation under vacuum.

1.2 Instrumentation.

All capillary liquid chromatography (CLC) were performed on an UltiMate 3000 high pressure LC system (Dionex, Sunnyvale, CA) equipped with an LPG-3x00 micropump system, an FLM-3100 microflow manager (1:100 split ratio) which guarantees a constant flow rate, an VWD-3400 variable-wavelength UV-vis absorbance detector with a 3 nL flow cell for on-column detection and an WPS-3000 automatic sampler. Chromeleon software from Dionex was used for system operation and data acquisition and processing. The flow rate was set at 1

μL/min and the column temperature was set at 25 ^{o}C in all experiments. The effective lengths of monolithic columns used in all CLC experiments were 25 cm. The injection volume was 50 nL in all CLC experiments.

A Beckman Coulter P/ACE MDQ instrument (Beckman Instruments, Fullerton, CA) was used for the capillary electrophoresis (CE) experiments. UV-absorbance detection was performed at 254 nm. A fused silica capillary of 56.5 cm (effective length 50 cm) × 50 μm I.D was used as the separation column. The conditions for capillary pretreatment and separation were the same as reported previously.[1]

Scanning electron microscopy (SEM) analyses were performed on a Hitachi FE-SEM S-4800 (Tokyo, Japan). Nitrogen adsorption-desorption measurements were conducted at 77 K on an ASAP2020 instrument (Micromeritics, Norcross, GA, USA). The FT-IR spectrum was acquired on a Thermo Nicolet iS10 FT-IR spectrometer (Waltham, MA). X-ray photoelectron spectra (XPS) were collected with a Thermo Fisher Scientific K-Alpha spectrometer with monochromatic Al Kα radiation (1486.6 eV). A UV-3600 UV spectrophotometer (Shimadzu, Japan) was used for the UV-absorption experiments. ^{1}H NMR, ^{13}C NMR and ^{11}B NMR spectra were recorded on a Bruker Avance DMX 500 MHz instrument (Bruker Biospin GmbH, Rheinstetten, Germany) using the solvent residual as peak as reference (7.26 ppm). $BF_3 \cdot OEt_2$ was used as an external reference for ^{11}B NMR spectra. Mass spectra were recorded on a Micromass GC-TOF mass spectrometer (Manchester, England，or Waters/Micromass UK Ltd., Manchester, UK) with electron ionization (EI) energy set at 70 ev.

2. The synthesis of 3-carboxy-benzoboroxole (3)

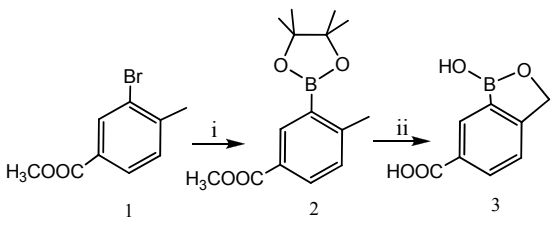

Fig. S1 The synthesis procedure of 3-carboxy-benzoboroxole

2.1 The synthesis of compound 2

To a 100 mL round bottom flask, methyl 3-bromo-4-methylbenzoate (1.00 g, 4.39 mmol), bis(pinacolato)diborane (1.33 g, 5.24 mmol), KOAc (1.28 g, 13.06 mmol), Pd(dppf)Cl_2 (95 mg, 0.13 mmol) were added. The flask was vacuumed and refilled with nitrogen three times. Then anhydrous 1,4-dioxan (30 mL) was injected through a rubber stop. The resulting suspension was stirred and heated at 100 ^oC under nitrogen atmosphere. Five minutes later, triethylamine (0.1 mL) was added into the mixture. The mixture was stirred and heated continually at 100 ^oC for 12 h. After cooling the mixture was diluted with 50 mL of ethyl acetate. The mixture was filtered and the filtrate was washed successively with water twice and brine once. The organic layer was collected and dried with anhydrous $MgSO_4$. The filtered solution was evaporated. The resulting crude product was purified by silica gel column chromatography (Hexane/EtOAc 50:1 v/v). The compound **2** was obtained as a white solid (1.12 g , 92%). ^1H NMR (500 MHz, $CDCl_3$) δ8.42 (1H, d, J = 1.5 Hz), 7.98 (1H, dd, J = 2 and 1.5 Hz), 7.26 (1H, d, J = 7.5Hz), 3.92 (3H, s), 2.61 (3H, s), 1.38 (12H, s). ^{13}C NMR (500 MHz, $CDCl_3$) δ167.3, 150.3, 137.1, 131.8, 129.9, 126.7, 83.7, 51.8, 24.9, 22.4. ^{11}B NMR (500 Hz, $CDCl_3$) δ31.02. MS (EI^+) *m/z*: [276.3 (0.24)] (M^+).

2.2 The synthesis of compound 3

Compound **3** was prepared by the method by Arnab Pal et al[2] with some modification. Compound **2** (1.12 g, 4.06 mmol) was dissolved in CCl_4 (30 mL) and the solution was charged into a three neck round bottom flask equipped with a reflux condenser. Then *N*-bromosuccinimide (0.81 g, 4.58 mmol) and azobisisobutyronitrile (36 mg, 0.02 mmol) was added to the flask resulting into a suspension. The reaction mixture was stirred under reflux for 4 h. After cooling to room temperature the solution was washed successively with water twice and brine once. The organic layer was collected and dried with anhydrous $MgSO_4$. The filtrate was concentrated under reduced pressure. The obtained yellow oil was then dissolved in ether (50 mL). Then the solution was extracted with 15% KOH (30 mL × 3). The organic phase was mixed with 20 mL of 15% KOH with stirring over night. The aqueous phase was collected and cooled

in ice bath. Then the aqueous phase was adjusted to pH 1 with HCl (12 M in water) with stir. The white precipitate was collected by filtration and dried in vacuum at 60°C. The dried white power was then mixed with chloroform (10 mL) and stirred for 4 h. The suspension was filtrated and the white power was dried in vacuum to afford the 3-carboxy-benzoboroxole.(0.45 g, 62 %). ^{1}H NMR (500 MHz, acetone-d_6) δ8.45 (1H, s), 8.14 (1H, d, J = 8 Hz), 7.55 (1H, d, J = 7.5Hz), 5.10 (2H, s). ^{13}C NMR (500 MHz, acetone-d_6) δ167.0, 158.9, 132.0, 131.8, 129.6, 121.5, 70.4. ^{11}B NMR (500 Hz, acetone-d_6) δ32.2. MS (EI$^+$) *m/z*: [177.0(0.41), 178.0(10.55)] (M^+).

3. Determination of the pKa value of 3-carboxy-benzoboroxole

The pKa value of 3-carboxy-benzoboroxole was measured by the method described by Wang et al[3] which based on the changes in the UV (268 nm) absorption following the titration of 1.0 mM of 3-carboxy-benzoboroxole in 100 mM phosphate buffer with 1 M NaOH (Fig. S2). The pKa value of 3-carboxy-benzoboroxole was measured to be approximately 6.92, which is lower than that of its decarboxylated analogue benzoboroxole[4] by 0.28 pH unit. The pKa shift was due to the electron withdrawing effect of the carboxyl group in the structure of 3-carboxy-benzoboroxole. Such a pKa shift is beneficial, because a lower pKa value of boronic acid generally results increased affinity towards cis-diol compounds.

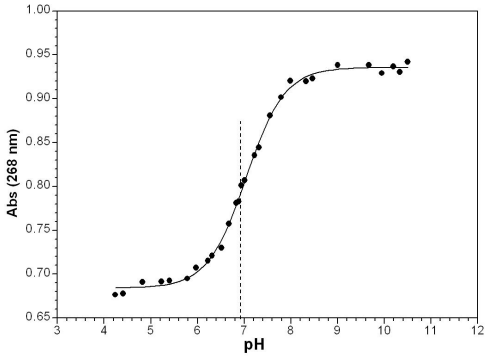

Fig. S2 The pH-dependence of the absorbance at 268 nm of 3-carboxy-benzoboroxole

4. Preparation and post-modification of the poly (GMA-co-MBAA) monolithic capillary

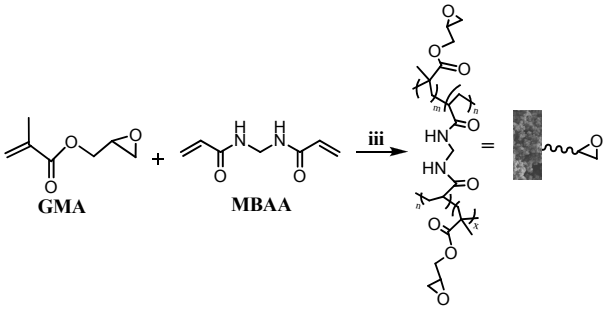

Fig. S3 The preparation of the poly(GMA-co-MBAA) monolith

4.1 Preparation of poly(GMA-co-MBAA) monolithic capillary

The preparation of poly(GMA-co-MBAA) monolithic column is illustrated in Fig. S3. Prior to the preparation of the monolith, vinylization of the inner wall of the capillary was carried out according to the activation procedure reported previously.[5] The base monolith was synthesized by thermo-initiated free radical copolymerization. Briefly, a mixture containing GMA (30 mg), MBAA (60 mg), DMSO (200 mg), dodecanol (174 mg) and AIBN (1 mg) was vortexed for 5 min and sonicated for 30 min to obtain a homogeneous solution. The vinylized capillary was filled with the polymerization mixture, then sealed with rubber at both ends and submerged into a water bath at 75 ^{o}C for 12 h. After the polymerization reaction, the resulting monolithic column was washed with methanol and acetonitrile successively. This capillary monolithic column is designated as "column M-1" in the following text.

4.2 Post-modification of poly (GMA-co-MBAA) monolithic capillary

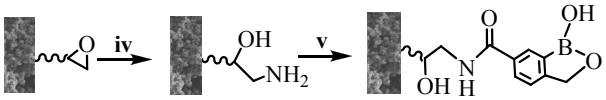

Fig. S4 Post-modification of poly(GMA-co-MBAA) monolithic capillary

The post-modification procedure is schematically shown in Fig. S4. First, column M-1 was amino-modified according to the method reported by Ye and co-workers[6] with some

modifications. A solution of ammonia (28%)/acetonitrile (1:1, v/v) was continuously passed through column M-1 to react with the epoxy for 12 h at 60°C. After reaction, the monolith was rinsed successively with acetonitrile for 6 h and anhydrous DMF for 6 h to completely remove the residual water on the surface of the monolith. The resulting monolithic column is referred as "column M-2" in the following text. Then, compound 3 was immobilized on the surface of column M-2 by the method by Hall et al[2] with some modifications. Briefly, HBTU (118 mg), HOAt, (45 mg), compound 3 (64 mg) and DIPEA (110 μL) was dissolved in 2 mL of anhydrous DMF. Then, the solution was sonicated for 5 min and vortexed for 5 h successively. The resulting bright yellow-green solution was continuously passed thought the monolith for 17 h at 40 °C. Finally, the monolith was washed with anhydrous DMF and acetonitrile for 2 h respectively to completely remove the un-reacted residues. This resulting monolithic column was referred as "column M-3" in the following text. Column M-3 was used for all the CLC experiments. Each column M-3 was activated by 100 mM HOAc and then conditioned by the loading buffer until the baseline was satisfactory.

4.3 Characterization of the modified poly (GMA-co-MBAA) monolithic capillary

The modified monolithic capillary was characterized in terms of scanning electron microscopy (SEM). As shown in Fig. S5, the monolith attached tightly to the inner wall of the capillary and exhibited well-controlled skeleton and a well-distributed macroporous open-channel network.

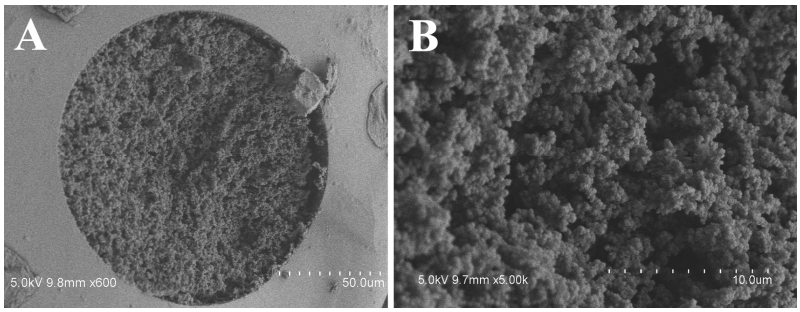

Fig. S5 The SEM images of cross-section of a benzoboroxole-modified monolithic capillary. A) 600×; B) 5,000×.

5. The preparation and post-modification of large-sized monolith for characterization.

5.1 The preparation and post-modification of large-sized monolith

First, the monolith prepared in regular HPLC column was pushed out and cut into thin pieces. Then the pieces were rinsed with methanol using a Soxhlet extractor for 24 h and dried under vacuum at 60 ^{o}C for 12 h. This monolithic material was named M-1.

A portion of the pieces of M-1 (500 mg) was mixed with a solution of ammonia/ acetonitrile (1:1, v/v) and the mixture was headed at 60^{o}C for 12 h. Then the mixture was filtrated and the solid was washed with methanol through a Soxhlet extractor for 24 h, followed by drying under vacuum at 60 ^{o}C for 12 h. The resulting monolithic material was named M-2.

A portion of M-2 (250 mg) was immerged into 2 mL of solution which was prepared by dissolved HBTU (118 mg), HOAt (45 mg), 3-carboxy-benzoboroxole (64 mg) and DIPEA (110 μL) in anhydrous DMF.[2] Then the mixture was shacked while heated at 40 ^{o}C for 17 h. After filtration, the solid was washed through a Soxhlet extractor for 24 h and dried under vacuum at 60 ^{o}C for 12 h. This obtained material was named M-3.

5.2. The characterization of large-sized monoliths

First, the monoliths were characterized by FT-IR spectrum to determine the success of the modification reactions.

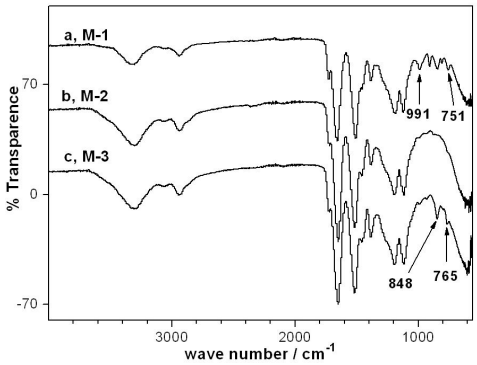

Fig. S6 FT-IR spectra of the bare (a), amino-modified (b) and compound 3-modified (3) poly(GMA-co-MBAA) monolith

From the contrast of trace a and b, the disappearance of the peak between 992 cm^{-1} and 752 cm^{-1} in b indicates the disappearance of epoxy group in M-2, because of the reaction between ammonia and epoxy group. Further, from the contrast of trace b and c, the new peaks in 848 cm^{-1} and 765 cm^{-1} can be attributed to the skeleton vibration of benzene ring, which suggests the successful attachment of compound 3 onto the surface of the monolith.

Second, the boron element loading on the surface of the modified monolith was quantified by X-ray photoelectron spectroscopy (XPS).

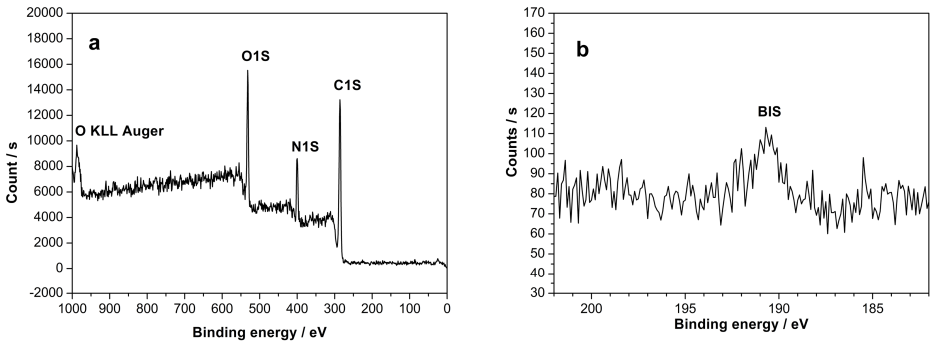

Fig. S7 The X-ray photoelectron spectroscopy survey scan (a) and the boron element of the modified monolith M-3.

In Fig. S7, no peak for boron element is observed and this might because of the low atom concentration of boron element in the modified monolith. From Figure S4b, the peak of boron atom can be observed in spite of the low intensity of the peak. In addition, the atom concentration ratio of C, O, N and B elements in the modified monolith was 67.56:18.64:13.01:0.80.

Finally, the large-sized monolith was characterized by Brunauer-Emmett-Teller (BET) method and the average pore size and specific surface area were measured to be 6.12 nm and 11.40 m^2/g, respectively

6. Further investigation of the boronate affinity property under acidic conditions

Quinol (6) and catechol (7) were additionally used as analytes to test the boronate affinity property of modified monolithic column at acidic conditions. Modified monolithic column with

an effective length of 25 cm was used in this section of experiments. The flow rate was set at 1 μL/min. Because of the strong affinity between the modified monolithic column and the retained catechol, 100 mM HOAc could not elute the retained catechol, so a stronger acidic solution, 1% formic acid, was used to effectively elute the retained catechol.

From Fig. S8 we can see that quinol showed no retention on the modified monolithic column at all tested conditions. In contrast, catechol was completely captured and enriched by the modified monolithic column at pH 5.0 (trace b in Fig. S8). When the pH was gradually reduced from 4.5 to 3.0, catechol could not be captured be the modified monolithic column but could still separated from quinol (trace c-e in Fig. S8). When the loading buffer was changed to 30:70 (v/v) ACN/1% formic acid, the same experimental result could be observe (trace f in Fig. S8).

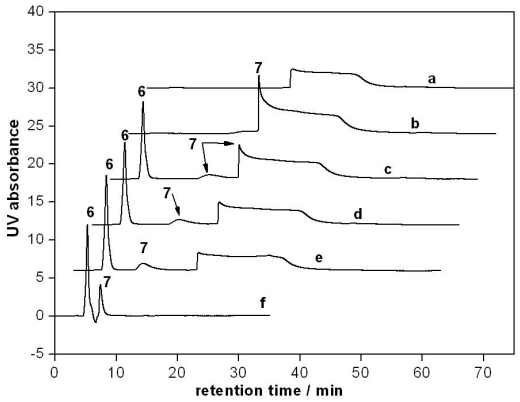

Fig. S8 The chromatographic retention of quinol (6) and catechol (7) on the modified monolithic column. Mobile phase: (a-e) 30:70 (v/v) ACN/30 mM sodium phosphate buffer at pH 5.0, 5.0, 4.5, 4.0, 3.0, respectively, switched to at 30:70 (v/v) ACN/1% formic acid at 15 min, (f) 30:70 (v/v) ACN/1% formic acid; Samples: (a) loading buffer, (b) 1 mg/mL catechol dissolved in responding loading buffer; (c-f) quinol and catechol (1 mg/mL each) dissolved in responding loading buffer; detection at 256 nm.

7. The repeatability of the benzoboroxole-immobilized monolithic column

The repeatability of the benzoboroxole-immobilized monolithic column was evaluated in terms

of the relative standard deviation of the retention time of adenosine. For each column, 5 times of parallel injection was conducted continuously. The sample was adenosine (0.4 mg/mL). The relative standard deviation (RSD) for run-to-run, column-to-column and batch-to-batch repeatability was 0.28% (n = 5), 2.35% (n = 3) and 4.87% (n = 3), respectively. From the results, it can be seen that the functionalized monolithic column showed good repeatability.

8. The separation of standard mixtures of glycoproteins and non-glycoproteins

To further demonstrate the selectivity and affinity of the benzoboroxole-immobilized monolithic column towards target glycoproteins, three samples of the target glycoproteins mixed respectively with standard non-glycoprotein were separated on the benzoboroxole-immobilized monolithic column.

As shown in Fig. S9, the tested glycoproteins were specifically captures by the monolithic column while the interfering non-glycoprotein was not retained by the monolithic column. These results further demonstrated the selectivity and affinity of the benzoboroxole-immobilized monolithic column towards glycoproteins.

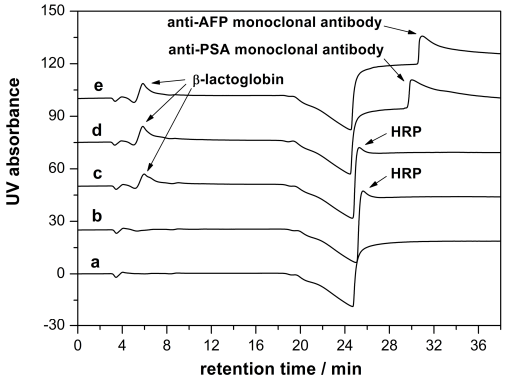

Fig. S9 The separation of the mixtures of glycoproteins and non-glycoprotein. Mobile phase, 20 mM HEPES containing 100 mM $MgCl_2$ (pH 7.0), switching to 100 mM HOAc at 15 min; samples, (a) the loading buffer as blank control; detection at 214 nm; (b) 0.5 mg/mL HRP; (c) 0.5 mg/ mL HRP mixed with 0.5 mg/ mL β-lactoglobin; (d) 0.5 mg/mL anti-PSA monoclonal antibody mixed with 0.5 mg/ mL β-lactoglobin; (e) 0.5 mg/mL anti-PSA monoclonal antibody mixed with 0.5 mg/ mL β-lactoglobin.

9. The binding capacity

9.1 The binding capacity for small molecule

The binding capacities of the benzoboroxole-immobilized monolithic column for small molecule at different pH values were measured using frontal chromatography using adenosine as the test compound with deoxyadenosine as the dead time marker. The column was first equilibrated with responding loading buffer, then a sample solution containing 1 mg/mL adenosine and 0.01 mg/mL deoxyadenosine dissolved in loading buffer was pumped through the column. Adenosine was captured by the column and eluted until the column was saturated with adenosine. The binding capacity values in the pH range from 5.0 to 8.5 are listed in Table S1.

Table S1 Binding capacities of the modified monolithic column at different pH values.

pH	capacity (μmol/mL)
5.0	5.30
6.0	12.16
7.0	17.58
7.4	19.07
8.0	20.76
8.5	22.33

As could be seen in Table S1, the modified monolithic column showed high binding capacity. The binding capacity of this boronate affinity monolithic column at a given pH condition is the highest as compared with those of the boronate affinity monolithic columns reported previously.[5,7-10] A comparison can be made between the current monolithic column and

the Wulff-type boronic acid functionalized monolithic column (referred as "Wulff-type column") previously reported by our group,[5] taking the specific surface area and surface boron atom concentration (B%) into consideration. The data for the column M-3 were 11.40 m^2/g (surface area) and 0.8 (B%) while those for the Wulff-type boronic acid-functionalized column were 5.34 m^2/g (surface area) and 1.01 (B%). The product of the two parameters for the column M-3 is only 1.69-fold as high as that for the Wulff-type column. However, the binding capacity at pH 7.0 was 17.86 and 6.82 μmol/mL for the benzoboroxole-functionalized and the Wulff-type boronic acid-functionalized column, respectively; the data for the former is 2.58-fold as high as that for the latter. Clearly, the improved boronate affinity was also a major contribution of the increased binding capacity of the benzoboroxole-type monolithic column.

9.2 The binding capacity for macromolecule

The binding capacity of the benzoboroxole-immobilized monolithic column for macromolecule (glycoprotein) at neutral pH condition was measured by frontal chromatography using HRP as the test compound with RNase A as the dead time marker. The column was first equilibrated with responding loading buffer (20 mM HEPES containing 100 mM $MgCl_2$, pH 7.0), then a sample solution containing 0.5 mg/mL HRP and 0.1 mg/mL RNase A dissolved in loading buffer was pumped through the column. HRP was captured by the column and eluted until the column was saturated with HRP. The dead time was 7.88 min and the column was saturated by HRP at 20.47 min. The benzoboroxole-immobilized monolithic column used here was 25cm long with a diameter of 150μm. The flow rate was set at 1 μL/min and the detection wavelength was set at 214nm. The molecular weight of HRP was 44,000 Da. Thus, the binding capacity for HRP was calculated to be 0.033 μmol/mL.

10. Analysis of urinary samples

10.1 Pretreatment of urine samples

Two urine samples were spontaneously collected from a healthy male and a healthy female respectively. The collected urinary samples were frozen immediately and stored at -20 ^oC. Prior

to analysis, the samples were thawed at room temperature. A 10 mL aliquot of urine was centrifuged for 30 min at 15000 r/min. The supernatant were collected and the pH values were adjusted to 7.4 by ammonia (28%).

A piece of the column M-3 with an effective length of 25 cm was used for boronate affinity extraction of urinary nucleosides. The urine samples were treated as the following procedure: the column M-3 was first conditioned with the loading buffer (30 mM sodium phosphate buffer, pH 7.4) for 40 min. After 200 μL of the supernatant was passed thought the modified monolithic column at a flow rate of 1 μL/min, the column was washed with the loading buffer for 15 min in order to completely flush out the un-retained components. Finally, the retained components were eluted by 100 mM HOAc and collected to tubes. For each tube, 5 μL eluate was collected and subsequently analyzed by the micellar electrokinetic chromatography (MEKC) method described by Jiang et al[1] with slight modification that a longer separation capillary (effective length 50 cm, 50 μm I.D) was used.

10.2 The enrichment and separation of nucleosides in urine samples

The applicability of the benzoboroxole-functionalized monolithic column to complex samples was demonstrated by the enrichment of urinary nucleosides from two urine samples (from a male and a female healthy adult respectively). As the urine samples had big difference in the pH values, the pH of the two samples were adjusted to physiological pH (7.4) prior to enrichment to eliminate the influence of pH on the extraction efficiency. The prepared samples were separated by the MEKC method described by Jiang et al.[1] This method was chosen because of its rapid separation speed and high performance. In addition, there was a system peak in the electropherograms, which could serve as an internal standard.

The electropherograms of the untreated and treated urine samples are shown in Fig. S10 and Fig. S10. The identities of some peaks were determined by spiking the urine sample with pure single nucleoside standards. As shown in Fig. S10A, the contrast between the un-extracted (trace a) and extracted (trace a') female urine samples, the components in the dotted square ii were removed while most of the peaks in the dotted square i were apparently enriched. Such

enrichment effect could also be seen in the male urine sample (Fig. S10B). These results indicate the high capacity and high enrichment efficiency of the benzoboroxole-functionalized monolithic column towards different urine samples.

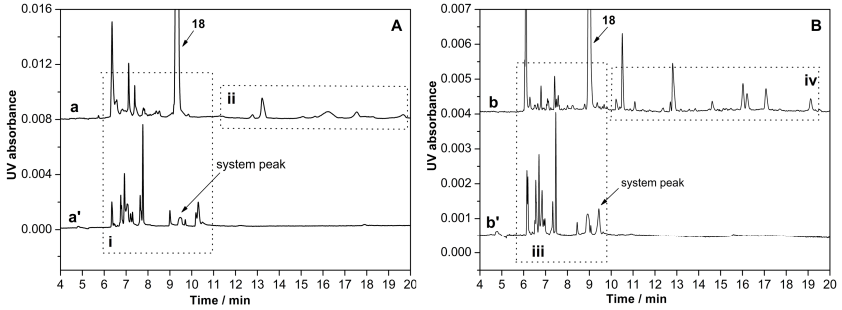

Fig. S10 The electropherograms of un-extracted (female: trace a; male: trace b) and extracted (female: trace a'; male: trace b'). Separation conditions: running buffer, 25 mM $Na_2B_4O_7$ +25 mM NaH_2PO4 +25 mM CTAB (pH 9.50); separation voltage, -15 kV ; UV detection, 254 nm; injection, 10 s × 0.5 psi; capillary: untreated fused silica, 56.5 cm (50 cm to detector, 50 μm I.D).

From the contrast between the un-extracted urine samples (Fig. S11A), it can be seen that the male urine sample had much lower component concentrations as compared with the female urine sample; especially in the migration time range from 5 to 10 min. In addition, the two traces show very different peak profiles. However, after extracted by the benzoboroxole-functionalized monolithic column, the differences in component concentration and peak profile were dramatically reduced, as shown in Fig. S11B. This allows for convenient sample-to-sample comparison according to the profiles of urinary nucleosides. Thus the usefulness of boronate affinity-based extraction in metabolomic analysis of urinary nucleosides is clearly demonstrated, which can specifically extract the targets and meanwhile remove the interfering species in the sample matrixes.

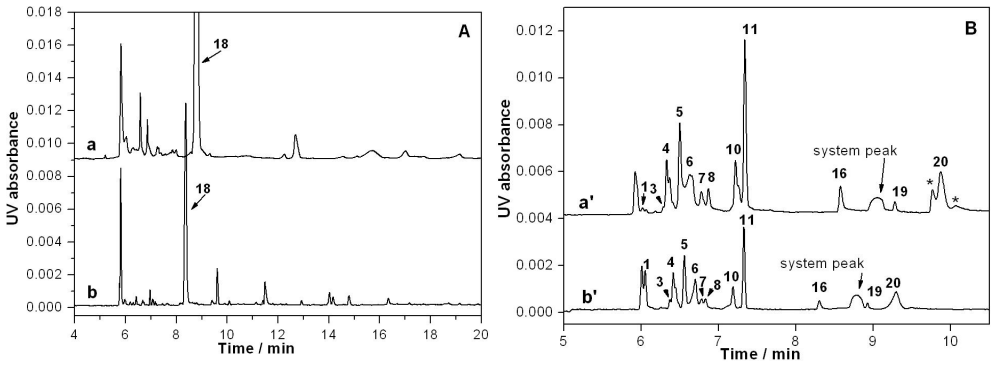

Fig. S11 The comparison of the electropherograms of female and male urine samples. A) the comparison of un-extract female (trace a) and male (trace b) urine samples; B) the comparison of extracted female (trace a') and male (trace b') urine samples. Peak identity: 1, uridine; 3, cytidine; 4, xanthosine; 5, adenosine or guanosine; 7, inosine; others, unknown. Separation conditions: running buffer, 25 mM $Na_2B_4O_7$ +25 mM NaH_2PO4 +25 mM CTAB (pH 9.50); separation voltage, -15 kV ; UV detection, 254 nm; injection, 10 s × 0.5 psi; capillary: untreated fused silica, 56.5 cm (50 cm to detector, 50 μm I.D).

11. ^{1}H NMR, ^{13}C NMR and ^{11}B NMR spectra of compounds 2 and 3

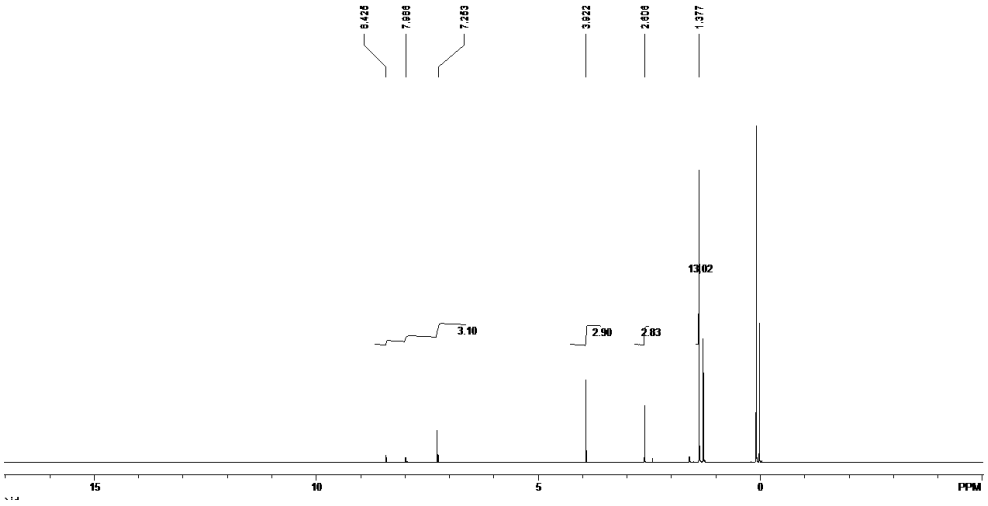

Fig. S12 ^{1}H NMR spectrum of compound 2 in $CDCl_3$

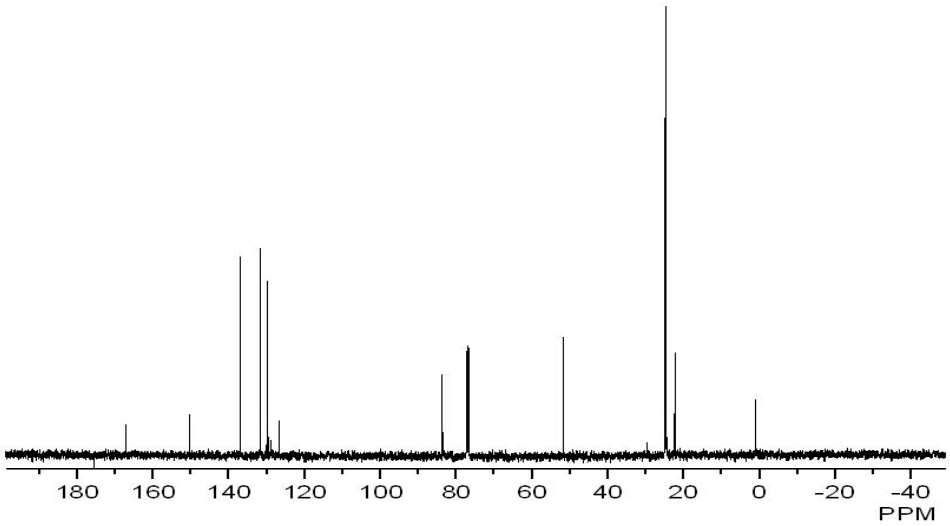

Fig. S13 ^{13}C NMR spectrum of compound 2 in $CDCl_3$

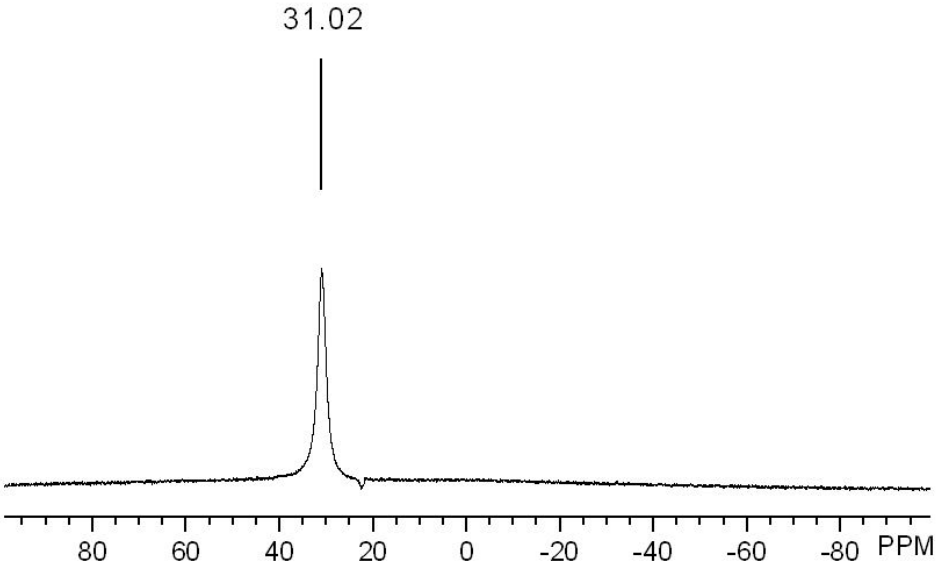

Fig. S14 ^{11}B spectrum of compound 2 in $CDCl_3$.

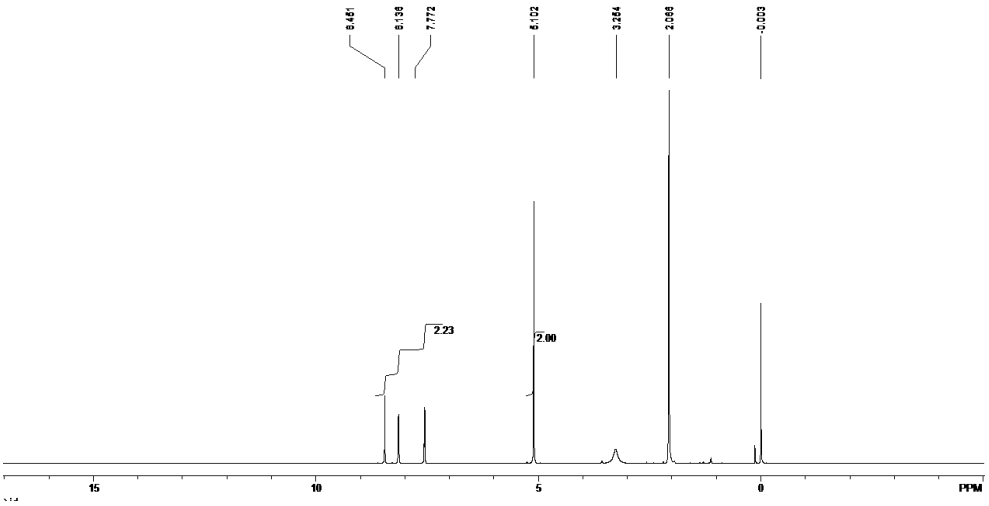

Fig. S15 ^{1}H NMR spectrum of compound 3 in acetone-d_6

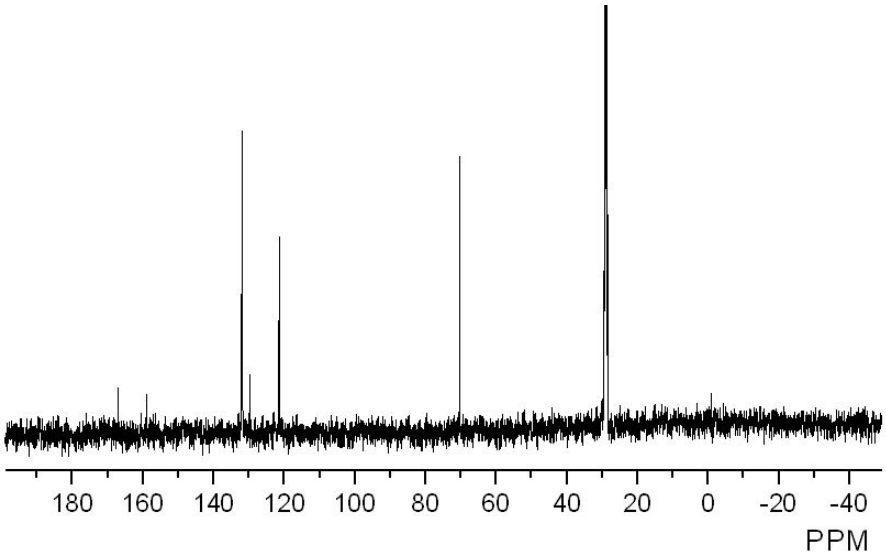

Fig. S16 ^{13}C NMR spectrum of compound 3 in acetone-d_6

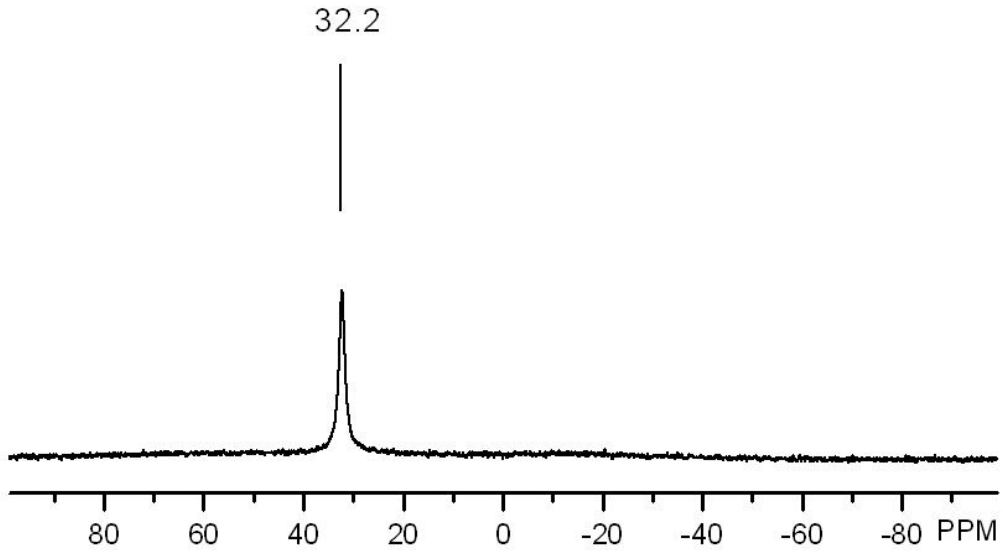

Fig. S17 ^{11}B spectrum of compound 3 in acetone-d_6.

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