Efficient enrichment and desalting of protein digests using magnetic mesocellular carbon foams in matrix-assisted laser desorption/ionization mass spectrometry

Young-Pil Kim1, Kun Cho2, Dohoon Lee1, Yunxian Piao1, Yeong Hee Ahn3, Jong Shin Yoo3, Taeghwan Hyun4 and Hak-Sung Kim1*

1Department of Biological Sciences, Korea Advanced Institute of Science and Technology (KAIST), Daejeon 305-701, Korea
2Proteomics Team, Korea Basic Science Institute (KBSI), Daejeon 305-333, Korea
3MS Instrumentation Division, Korea Basic Science Institute (KBSI), Daejeon 305-333, Korea
4National Creative Research Initiative Center for Oxide Nanocrystalline Materials and School of Chemical and Biological Engineering, Seoul National University, Seoul 151-744, Korea

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We demonstrate that magnetic mesocellular carbon foams (Mag-MCF-C) can be effectively used for enrichment and desalting of protein digests or peptides in matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS). The large mesocellular pores and surface area of Mag-MCF-C are likely to mainly contribute to high efficiency in enrichment and desalting of protein digests. The magnetic property of Mag-MCF-C enabled easy and simple enrichment and desalting process comprising adsorption, washing, and separation steps by using an external magnet. Following elution from Mag-MCF-C by using a matrix solution (CHCA in 70% ACN/0.1% TFA), the peptides were subjected to MALDI-MS analysis. As a result, MALDI mass spectra of peptides or tryptic protein digests were distinct even at a peptide concentration as low as 50 pM. The use of Mag-MCF-C resulted in significantly improved sequence coverage for protein identification when compared to other conventional methods. Mag-MCF-C will find applications in mass spectrometric analysis of low abundance peptides or protein digests with high sensitivity. Copyright © 2007 John Wiley & Sons, Ltd.
reasoned that Mag-MCF-C has a potential utility for enrichment and desalting of proteins/peptides prior to MALDI analysis.

In this paper, we describe that Mag-MCF-C can be effectively used for enrichment and desalting of protein digests in MALDI-MS. The structural property and magnetism of Mag-MCF-C enabled high loading of proteins/peptides into mesopores and easy separation from a solution by an external magnetic force, resulting in efficient enrichment and desalting of protein samples. Protein digests were concentrated and desalted by using Mag-MCF-C, followed by application to MALDI-TOF analysis. Efficiency of Mag-MCF-C was investigated in terms of concentration factor and detection limit, and compared with currently employed methods. Details are reported herein.

**EXPERIMENTAL**

**Chemicals and materials**

α-Cyano-4-hydroxycinnamic acid (CHCA) and trifluoroacetic acid (TFA) was obtained from Sigma-Aldrich. Acetonitrile (ACN, HPLC grade) was purchased from Merck. Graphite, activated carbon, tetraethyloxsilicate (TEOS, 98%), divinylbenzene (DVB, 98%), 1,3,5-trimethylbenzene (TMB, 98%), and 2,2'-azobisobutyronitrile (AIBN, 98%) were purchased from Aldrich. Bovine serum albumin (BSA) and dithiothreitol (DTT) were purchased from Sigma. Sequencing-grade trypsin was purchased from Promega. TAMRA (5(6)-carboxytetramethylrhodamine)-labeled angiotensin I (TAMRA-DRVYIHPFHL, M₀: 1708.49) was synthesized by Peptron Inc. (Korea). Pluronic P123 (EO₈₅PO₇₀EO₈₅, Mₐ: 5800) was obtained from BASF. All other chemicals were of analytical reagent grade, and were used without further purification. Distilled and deionized water (18 MΩ Milli-Q water system) was used throughout sample preparations.

**Synthesis of Mag-MCF-C**

The MCF silica template was first synthesized according to the method previously described.²¹ Briefly, 4 g of P123 was completely dissolved in a solution composed of 130 mL of deionized water and 21 mL of hydrochloric acid (36 wt. %). Then, 4.3 mL of TMB was added into the P123 polymer solution. Following addition of 9.2 mL of TEOS, the resulting solution was aged at 313 K for 20 h and another 24 h at 373 K. The resulting white precipitate was filtered, dried at room temperature, and finally calcined at 550°C in air to obtain the MCF silica template. For the synthesis of Mag-MCF-C, 1 g of dried MCF silica was soaked into the polymer precursor solution composed of 1 mL DVB and free radical initiator, AIBN, with a molar ratio of 1:51. Polymerization was performed by heating at 85°C for 12 h under an atmosphere of argon followed by incorporation of 0.71 g of Fe(NO₃)₃·9H₂O dissolved in ethanol into the pores of the MCF/poly(DVB) composite using the impregnation method. The composite was heated at 800°C for 1 h under an atmosphere of nitrogen (heating rate: 2°C min⁻¹). Dissolution of the MCF template using 1 M NaOH (ethanol/water 1:1, v/v) at 100°C yielded Mag-MCF-C. The surface morphology of the Mag-MCF-C was obtained by using transmission electron microscopy (TEM) and the mesopore size distributions were determined from the analysis of the adsorption branch of Ar (or N₂) isotherms using BJH formalisms. The Mag-MCF-C was found to have a similar overall carbon structure to MCF-C, characterized by co-existence of three types of substructures.

**Preparation of tryptic protein digests**

BSA was denatured in 100 mM Tris buffer (pH 8.0) containing 5 mM DTT, and subjected to digestion in 100 μL with trypsin at an enzyme-to-protein ratio of 1:20 (w/w) overnight at 37°C. The sequencing-grade trypsin was used in a reaction buffer according to the manufacturer’s instructions. The resulting digest was concentrated in a low-temperature vacuum drier, and then diluted with distilled water for MALDI-MS analysis. The observed masses of peaks in the mass spectra were searched against the theoretical digests for identification of peptides. Theoretical digests of BSA were obtained from PeptideMass.²² The experimentally observed masses for the assigned peaks were used to calculate sequence coverage by MS-Fit.²³

**Sample preparation for MALDI-TOF MS**

Unless otherwise stated, enrichment of standard peptides or tryptic digests (sample volume: 100 μL) was achieved by adding 10 μL of Mag-MCF-C (9 mg mL⁻¹) suspended in phosphate buffer (10 mM, pH 7.4). In order to test the effect of the concentration of the peptide or protein digest, the sample solutions were diluted in the range of 100 pM to 10 μM with appropriate buffer, and used for enrichment. The sample solution was mixed with Mag-MCF-C followed by agitation at 250 rpm for 30 min at room temperature. By applying a magnet, Mag-MCF-C pellets were separated, and washed three times with 500 μL of 0.1% TFA. For elution, the resulting Mag-MCF-C pellets were resuspended in 10 μL of freshly prepared matrix solution (10 mg mL⁻¹ of CHCA dissolved in 70% ACN/0.1% TFA). Following incubation for 10 min at room temperature, 0.7 μL of the supernatant solution was taken by using a micropipette under the magnetic field, and then directly spotted onto the MALDI target plate.

To test the performance of Mag-MCF-C in enrichment, angiotensin I (DRVYIHPFHL) with a monoisotopic mass of 1295.68 was conjugated with TAMRA at the N-terminus and used. The dye-labeled peptide was also applied to graphite carbon and activated carbon for comparison with Mag-MCF-C. Enrichment efficiency was determined by measuring the concentration of dye-labeled peptide before and after enrichment from the extinction coefficient of the TAMRA dye (65 000 M⁻¹ cm⁻¹ at 555 nm). The recovery yield was calculated by analyzing the amounts of eluted and adsorbed peptides using a similar procedure to that described above.

To check the desalting effect, angiotensin I was dissolved in 10 mM phosphate-buffered saline (pH 7.4) containing 1 M NaCl followed by incubation with a predetermined amount of Mag-MCF-C for 30 min. After washing three times with 0.1% TFA solution, the peptide was eluted, subjected to MS analysis, and compared with a salt-free solution of the peptide. As a control, the same concentration of sample was directly applied to a MALDI plate without enrichment. Typically, 1 μL of a solution of standard peptide (or a tryptic
digest) was mixed with an equal volume of the matrix solution (10 mg/mL of CHCA dissolved in 70% ACN/0.1% TFA), and then 0.7 μL of the mixture was spotted onto the MALDI target plate for the MS analysis. For comparison with a currently used method, a C18 ZipTip (Millipore) was tested according to the manufacturer’s specifications. A 10 μL volume of the sample was adsorbed onto the ZipTip, and rinsed in 0.1% TFA. Elution of target peptide from the ZipTip was conducted directly onto a standard stainless steel MALDI target by dispensing about 0.7 μL of a matrix solution.

MALDI analysis
Mass spectrometric analyses of peptides or protein digests were performed using a 4700 proteomics analyzer (Applied Biosystems, USA) equipped with delayed extraction and a 200-Hz repetition rate ND:YAG laser (355 nm). The instrument was operated in the reflection positive mode and run in an automated (batch-mode) fashion after an external calibration procedure using a mixture of standard peptides. Mass spectra were obtained from each sample spot using 1000 shots per spectrum, and the data were then processed by using the Data Explorer (Applied Biosystems, version 4.5) software. Mean intensities and standard deviations of mass peaks in the present study were obtained from at least duplicate experiments. For identification, the data were submitted to Swiss-Prot database with mass error set below 100 ppm. Standard 192-well stainless steel MALDI sample plates were used in this study.

RESULTS AND DISCUSSION
Characteristics of Mag-MCF-C
We previously found that magnetic mesoporous cellular carbon foams (Mag-MCF-C) possess high surface area (865 m² g⁻¹) and large pore volume (1.43 cm³ g⁻¹) from TEM images and BET analysis.¹¹ TEM images revealed that Mag-MCF-C has three kinds of substructures (Fig. 1): interconnected large mesopores (diameter ca. 16.6 nm), small mesopores surrounding the cells (width ca. 3 nm), which are empty spaces produced by the removal of silica template, and micropores (width ca. 0.7 nm) present throughout the carbon framework. Magnetic nanoparticles (MNPs) were well distributed mainly on the exterior surface of MCF-C due to the larger size (~30 nm) than the cellular diameter of MCF-C. Considering the pore structures and magnetic property, Mag-MCF-C is expected to adsorb large amounts of peptides or protein digests. More importantly, Mag-MCF-C can be easily separated from a solution for enrichment of target peptides or protein digests by applying a magnet.

Enrichment of target peptides using Mag-MCF-C
We first tested the performance of Mag-MCF-C in enrichment by using TAMRA-conjugated angiotensin I as a target peptide. The schematic procedure and changes in absorbance of TAMRA-angiotensin 1 at each step are shown in Fig. 2. The peptide concentration in each step was determined from the extinction coefficient (65 000 M⁻¹ cm⁻¹ at 555 nm) of the TAMRA dye. The red-colored peptide solution (1 mL in 10 μM) in the initial microcentrifuge tube turned clear immediately after the addition of Mag-MCF-C (5.3 μL in 9 mg mL⁻¹) to the solution (Mag-MCF-C/ dye = 1:1, w/w). No color change was observed during the washing step with 0.1% TFA, but the red color returned when eluted with a matrix solution (10 mg/mL CHCA dissolved in 70% ACN/0.1% TFA) through magnetic separation. This indicates that Mag-MCF-C can be effectively used for enrichment of peptides or protein digests. Conjugation of TAMRA dye onto peptides might increase the hydrophobicity of the peptides, leading to enhanced enrichment yield. However, when dye-free peptides (DRVYIHPFHL or AGCKNFWKTFTSC) were tested by measuring the absorption at 205 nm (for DRVYIHPFHL) or 280 nm (for AGCKNFWKTFTSC), a similar result was also observed in terms of enrichment and recovery (data not shown), supporting that carbon materials can accommodate the peptides into the mesopores. It is noteworthy that a porous carbon column was involved in the successful MS identification of hydrophobic peptides.⁵,¹⁴

We determined the enrichment efficiencies of other carbon materials including graphite and activated carbon to compare with that of Mag-MCF-C by using a similar experimental procedure. As shown in Fig. 3, Mag-MCF-C showed an enrichment efficiency higher than 99%, whereas graphite and activated carbon exhibited efficiencies of around 56% and 91%, respectively. This result indicates

![Figure 1. TEM image (left) of synthesized Mag-MCF-C and schematic illustration (right) of the pore structures with magnetic nanoparticles (MNPs).](image-url)
that Mag-MCF-C was more effective for enrichment of peptides than other carbon materials tested. The surface areas of graphite carbon and activated carbon were estimated to be 16 m² g⁻¹ and 500 m² g⁻¹, respectively. On the other hand, the surface area of Mag-MCF-C was determined to be about 865 m² g⁻¹. MCF-C without MNPs also showed a similar enrichment efficiency to Mag-MCF-C (data not shown), which implies that the high enrichment efficiency of Mag-MCF-C is mainly attributed to the high surface area of MCF-C, rather than the existence of MNPs. It was observed that even large glucose oxidase (GOx) was well incorporated into the pores of MCF-C.7

To determine the optimal ratio of Mag-MCF-C to peptide, enrichment was carried out at varying concentrations of mesoporous carbons (Fig. 4(a)). Enrichment efficiency was calculated from the maximum absorbance of the TAMRA-angiotensin I before and after enrichment. Within 60 min, the enrichment efficiency reached above 90% in the given range of Mag-MCF-C/peptide (0.2:1 to 3.5:1, w/w).

As the ratio of Mag-MCF-C to peptide decreased, the enrichment efficiency was reduced with the decrease in the incubation time. A maximum loading of GOx in Mag-MCF-C was previously reported to be ~53 wt %.11 Meanwhile, a loading of the peptide approached about 500 wt % (0.2:1, w/w), resulting in ten-fold higher loading than that of GOx, which is likely to be due to the much smaller molecular mass of the peptide. Following enrichment and extensive washing, the dye-labeled peptides were eluted from the Mag-MCF-C using the matrix solution (CHCA in 70% ACN/0.1% TFA), which has been used as an elution solvent for a graphite powder.5 The presence of hydrophobic matrix (CHCA) in the elution solvent provides a competitive reaction for hydrophobic interaction between the peptides and Mag-MCF-C, resulting in increased recovery yield from the mesoporous carbon.5 The recovery yield (the ratio of eluted amount to adsorbed one) was estimated to be 82% (Fig. 4(b)), which indicates easy elution of the adsorbed peptides by using the matrix solution.

![Figure 2](image1.png)

**Figure 2.** Schematic illustrations of overall procedure (top) and the changes in absorbance at each step (bottom). Mag-MCF-C (9 mg mL⁻¹) was used for enrichment and desalting of TAMRA-labeled angiotensin I.

![Figure 3](image2.png)

**Figure 3.** TAMRA-angiotensin I solution (left) and absorbance spectra (right) after incubation for 60 min with various materials: (a) before enrichment, (b) graphite carbon, (c) activated carbon, and (d) Mag-MCF-C. The ratios of carbon to peptide were at 1:1 (w/w). Maximum absorbance of the respective supernatant was measured at 555 nm with a UV-Vis spectrophotometer. The percentage means enrichment efficiency of the TAMRA-peptide and was estimated as described in the Experimental section.
Enrichment effect of Mag-MCF-C in MS analysis

The enrichment effect of Mag-MCF-C on peptides was directly assessed by MS analysis of angiotensin I. For comparison, different sample preparation methods were used (Fig. 5). In contrast to direct deposition from the mixture of the peptide and matrix solution (1:1, v/v; Fig. 5(a)), enrichment using Mag-MCF-C yielded a strong peak intensity in the region of the monoisotopic molecular ion mass (m/z 1296.68) of the peptide (Fig. 5(b)), and the peak intensity was distinctly enhanced as the volumetric capacity increased to 1 mL (Fig. 5(c)). In addition, the maximum peak intensities ([M + H]+) of peptides after enrichment at a sample volume of 100 μL showed a much higher profile than those of direct deposition without enrichment (Fig. 6). By employing Mag-MCF-C for enrichment, the peptide signal was distinctly detectable even at a concentration as low as 50 pM (inset graph). This detection sensitivity of peptides in MALDI analysis was about one order of magnitude higher than previously used methods employing nanoparticles.15,16

Figure 4. (a) Effect of the ratios (0.2:1–3.5:1, w/w) of Mag-MCF-C to peptide on enrichment with respect to the time course of incubation (5, 30, and 60 min). Enrichment efficiency was calculated from maximum absorbance of the TAMRA-labeled peptide before and after enrichment. (b) UV-Vis spectra of TAMRA-peptide solutions showing elution of the analyte from Mag-MCF-C. Recovery efficiency was estimated to be ~82% from maximum absorbance of the TAMRA dye before (solid line) and after (dotted line) elution of TAMRA-labeled angiotensin I.

Figure 5. MALDI mass spectra of angiotensin I with different preparation methods: (a) direct mixing of matrix and analyte, and (b) and (c) enrichment by Mag-MCF-C. Sample volumes at the same concentration were different: (a) 1 μL, (b) 100 μL, and (c) 1 mL.

Figure 6. Enrichment effect of Mag-MCF-C on the peak intensity of molecular mass ([M + H]+) from angiotensin I. Concentrations of angiotensin I varied from 50 pM to 10 μM through enrichment and elution by Mag-MCF-C (black circles or bars), or direct mixing of the analyte and matrix without enrichment (white circles or bars). Sample volume at each concentration was 100 μL for Mag-MCF-C and 1 μL for direct mixing, respectively. Inset shows magnified peak intensities at low concentrations of angiotensin I, namely 50 and 100 pM.
and conventional alkyl-derivatized magnetic beads, where minimal detection limits were reported to be about 500 pM.

Desalting effect of Mag-MCF-C in MS analysis
To evaluate the desalting effect of Mag-MCF-C, angiotensin I was dissolved in phosphate buffer containing a high salt concentration (1 M NaCl), and followed by MALDI-TOF analysis (Fig. 7). When the peptide solution (100 nM) was enriched with Mag-MCF-C and rinsed with 0.1% TFA, the molecular ion peak of the peptide was clearly observed in the mass spectrum, and there was little change in the peak intensity before and after the addition of 1 M NaCl (Figs. 7(a) and 7(a')). On the other hand, direct deposition without desalting resulted in drastically reduced peak intensity of the peptide in the presence of 1 M NaCl (Figs. 7(b) and 7(b')). In addition, in the case of direct deposition, the sodium adduct peak ([M+Na]+) increased after the addition of a high salt concentration. This result indicates that Mag-MCF-C can also be effective for desalting of peptides or protein digests for MS analysis. Since many kinds of salts suppress the mass ion peaks, increasing background noise, simple removal of salts using Mag-MCF-C can enhance the sensitivity of MS analysis of peptides or protein digests. It has been reported that surfactants such as SDS, Triton X, or Tween-20 interfere with MS analysis by reducing analyte ion signals and degrading mass resolution. When a non-ionic surfactant, Tween-20, was mixed in a sample solution for MS analysis, the Mag-MCF-C was found to be also effective in removing the surfactant at a final concentration of 0.05–1% (data not shown).

Enrichment of protein digests by Mag-MCF-C
To demonstrate the utility of Mag-MCF-C for enrichment of protein digests, a tryptic digest of bovine serum albumin (BSA) was enriched with Mag-MCF-C, analyzed by using MALDI-TOFMS, and compared with direct deposition and a conventional silica material (ZipTip) (Fig. 8). At 50 pM of protein digest, which is equal to 50 amol μL⁻¹, the mass fingerprints of BSA were not detected in both cases of direct deposition with matrix solution (Fig. 8(a)) and the use of a ZipTip (Fig. 8(b)). On the other hand, enrichment by using Mag-MCF-C resulted in distinct peptide peaks (Fig. 8(c)), similar to the case of the standard peptide (Fig. 5). Seven peptides were found to match corresponding fragments of BSA through MS-Fit search, representing 11.4% sequence coverage (69 identified sequences out of 607 total amino acids). In the case of enrichment with Mag-MCF-C, the sequence coverage increased with the increasing concentration of protein digests, showing relatively higher values than those by direct deposition or the ZipTip method (Table 1). In a 4700 MALDI TOF/TOF™ analyzer, the minimal detection limit in general MALDI-MS analyses has been reported to be in the range of nM (~fmol μL⁻¹). Thus, the use of Mag-MCF-C seems to be very effective for enrichment and MS analysis of low abundance peptides or proteins that would not be detected by conventional methods. In addition, the pore size (16.6 nm diameter) of the used Mag-MCF-C was sufficient to enrich a broad mass range (m/z 800–4000) of digested BSA regardless of their digested size. However, given the Mag-MCF-C with different pore sizes, it may be possible to selectively concentrate peptides with different sizes.

The detection sensitivity of MALDI-MS might be influenced by several factors including sample volume with respect to enrichment capacity or amount of adsorbent. In contrast to Mag-MCF-C, the ZipTip method might not afford sufficient enrichment of sample due to its volume capacity as low as 10 μL. To effectively compare the concentrating effect for different sample volumes, we introduced a concentration factor (CF) as follows:

$$CF = \frac{CF_E}{CF_D}$$

where $CF_E$ is the elution concentration factor (i.e. sample volume/elution volume) and $CF_D$ is the deposition concentration factor (i.e. deposition volume per spot in a MALDI
plate/elution volume). As for the ZipTip approach, 10 μL of sample was loaded, followed by elution of 2 μL, and 0.7 μL was deposited for MALDI analysis. On the other hand, in the case of Mag-MCF-C, 1 mL of sample was employed, followed by elution of 5 μL, and 0.7 μL was subjected to MALDI analysis. Accordingly, the CF of Mag-MCF-C was estimated to be 28.0, which corresponds to 16-fold higher than that of ZipTip (CF: 1.75). However, when considering the amounts of the respective material (0.6 μL bed volume for C18 silica of ZipTip and 10 μL volume for Mag-MCF-C), the two different methods showed a similar CF per unit volume of adsorbent (2.9 CF μL⁻¹ for ZipTip; 2.8 CF μL⁻¹ for Mag-MCF-C). This comparison clearly demonstrates that Mag-MCF-C provides higher adsorption capacity and recovery yield with small sample loss, resulting in enhanced detection sensitivity compared with that by silica-based carbon materials including ZipTip. Commercially available ZipTip has been proposed for sample clean-up, rather than for the enrichment.20 In this regard, Mag-MCF-C would be a primary choice because it can be used for enrichment as well as desalting of digested samples. It was reported that the use of other C4 or C18 silica-based columns or cartridges usually resulted in a detection sensitivity higher than 500 pM (500 amol μL⁻¹) even for a sample volume of 1 mL.21,22 The superior performance of Mag-MCF-C is likely to be mainly attributed to its high binding capacity and large surface area, indicating the potential of Mag-MCF-C for enrichment of low abundance peptides or protein digests prior to MALDI-MS analysis. By employing gentle magnetic separation, sample can remain intact without damage.

Table 1. Comparison of sequence coverage from BSA digests with different sample preparation methods. Varying concentrations of sample were tested in MALDI-MS analysis for the protein identification

<table>
<thead>
<tr>
<th>Sample preparation method</th>
<th>Sample concentration</th>
<th>No. of matched peptides</th>
<th>Sequence coverage (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Direct deposition</td>
<td>10 nM</td>
<td>2 ± 0</td>
<td>2.3 ± 0.0</td>
</tr>
<tr>
<td></td>
<td>1 nM</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>ZipTipa</td>
<td>10 nM</td>
<td>31 ± 4</td>
<td>46.5 ± 2.1</td>
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<td></td>
<td>1 nM</td>
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<tr>
<td></td>
<td>50 pM</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Mag-MCF-Cb</td>
<td>10 nM</td>
<td>38 ± 5</td>
<td>48.8 ± 3.2</td>
</tr>
<tr>
<td></td>
<td>1 nM</td>
<td>37 ± 3</td>
<td>45.5 ± 1.4</td>
</tr>
<tr>
<td></td>
<td>50 pM</td>
<td>7 ± 0</td>
<td>11.4 ± 0.0</td>
</tr>
</tbody>
</table>

a,bSample enrichment and desalting were performed with different sample volumes (a: 10 μL, b: 1 mL).

c Sequence coverage for the assigned peaks was calculated by MS-Fit.13
CONCLUSIONS

We have demonstrated that Mag-MCF-C is very effective for enrichment and desalting of peptides or protein digests compared to other carbon and conventional C18-derivatized silica materials. Due to high adsorption capacity and large surface area of Mag-MCF-C, high enrichment efficiency was observed in mass spectrometric analysis of peptides by using MALDI-TOF, resulting in the sensitivity one order of magnitude higher than conventional methods. In addition, the magnetic property of Mag-MCF-C enabled a rapid and easy enrichment and desalting process in a single micro-centrifuge tube through magnetic separation. Mag-MCF-C will find applications in mass spectrometric analysis of low abundance peptides or protein digests with high sensitivity.

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REFERENCES