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Single-Crystalline EuF_3 Hollow Hexagonal Microdisks: Synthesis and Application as a Background-Free Matrix for MALDI-TOF-MS Analysis of Small Molecules and Polyethylene Glycols

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Single-crystalline EuF_3 hexagonal microdisks with hollow interior were fabricated to serve as a background-free matrix for analysis of small molecules and polyethylene glycols (PEGs) by matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF-MS). The long-lived excited state of europium ions can transfer energy to high-energy vibrations of organic molecules, which provides the potential technological application in MALDI-TOF-MS analysis of small molecules and PEGs. The efficiency of the hollow microdisks as a novel matrix of low molecular weight compounds was verified by analysis of small peptide, amino acid, organic compounds, and hydroxypropyl β-cyclodextrin (HP-β-CD). The advantage of this matrix in comparison with α-cyano-4-hydroxycinnamic acid (CHCA) and 2,5-dihydroxybenzoic acid (DHB) was demonstrated by MALDI-TOF-MS analysis of an amino acid mixture and a peptide mixture. This matrix is successfully used for analysis of PEGs (PEG 2000, PEG 4000, PEG 8000, PEG 15000, and PEG 30000), suggesting a potential for monitoring reactions and for synthetic polymer quality control. The upper limit of detectable mass range was ~35 000 Da (PEG 30000). It is believed that this work will not only offer a new technique for high-speed analysis of small molecules and PEGs but also open a new field for applications of rare earth fluorides.

Mass spectrometry (MS) is a well-established and very popular technique for identification of organic compounds and biomolecules. Matrix-assisted laser desorption ionization (MALDI) is a soft ionization technique for MS and has become a mainstream method for the investigation of biomolecules[1−5] and synthetic macromolecules[6,7] due to its high accuracy, rich structural information, low detection limit, rapid analysis time, and tolerance to salts and other contaminants. Despite these advantages, there are some practical problems in the application of this "soft" ionization technique. Chiefly, this popular technique is rarely used for analysis of analytes of low molecular weight (<500 Da) because the conventional matrix generates strong matrix-related background noise in the low-mass region, which interferes with the analysis of the analytes. Second, the suppression effects among analyte molecules frequently arise during MALDI analysis of the mixtures, resulting in the poor reproducibility and resolution of peaks for analytes.[8] To overcome these limitations, several different approaches have been developed, including exploring suitable matrices[9,10] or matrix additives[11] to improve the sensitivity of MALDI detection of target analytes by selectively enhancing the signals of target analytes or minimizing the background. Recent reports, on the use of nanoscale materials (carbon materials,[12−17] porous silicon,[18−24] magnetic nanoparticles,[25,26]

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(1) Chen, Y. Q.; Bi, F.; Wang, S. Q.; Xiao, S. J.; Liu, J. N. *J. Chromatogr., B* **2008**, *875*, 502–508.
(2) Sturiale, L.; Barone, R.; Palmigiano, A.; Ndosimao, C. N.; Briones, P.; Adamowicz, M.; Jaeken, J.; Garozzo, D. *Proteomics* **2008**, *8*, 3822–3832.
(3) Shen, W. W.; Xiong, H. M.; Xu, Y.; Cai, S. J.; Lu, H. J.; Yang, P. Y. *Anal. Chem.* **2008**, *80*, 6758–6763.
(4) Tang, K.; Opalsky, D.; Abel, K.; van den Boom, D.; Yip, P.; Mistro, G. D.; Braun, A.; Cantor, C. R. *Int. J. Mass Spectrom.* **2003**, *226*, 37–54.
(5) Puapaiboon, U.; Jai-nhuknan, J.; Cowan, J. A. *Anal. Chem.* **2000**, *72*, 3338–3341.
(6) Aksenov, A. A.; Bier, M. E. *J. Am. Soc. Mass Spectrom.* **2008**, *19*, 219–230.
(7) Muller, R.; Laschober, C.; Szymanski, W. W.; Allmaier, G. *Macromolecules* **2007**, *40*, 5599–5605.
(8) Pan, C. S.; Xu, S. Y.; Zhou, H. J.; Fu, Y.; Ye, M. L.; Zou, H. F. *Anal. Bioanal. Chem.* **2007**, *387*, 193–204.
(9) Ayorinde, F. O.; Garvin, K.; Saeed, K. *Rapid Commun. Mass Spectrom.* **2000**, *14*, 608–615.
(10) Ayorinde, F. O.; Hambright, P.; Porter, T. N.; Keith, Q. L., Jr. *Rapid Commun. Mass Spectrom.* **1999**, *13*, 2474–2479.
(11) Guo, Z.; Zhang, Q. C.; Zou, H. F.; Guo, B. C.; Ni, J. Y. *Anal. Chem.* **2002**, *74*, 1637–1641.
(12) Pan, C. S.; Xu, S. Y.; Hu, L. G.; Su, X. Y.; Ou, J. J.; Zou, H. F.; Guo, Z.; Zhang, Y.; Guo, B. C. *J. Am. Soc. Mass Spectrom.* **2005**, *16*, 883–892.
(13) Pan, C. S.; Xu, S. Y.; Zou, H. F.; Guo, Z.; Zhang, Y.; Guo, B. C. *J. Am. Soc. Mass Spectrom.* **2005**, *16*, 263–270.
(14) Han, M.; Sunner, J. *J. Am. Soc. Mass Spectrom.* **2000**, *11*, 644–649.
(15) Xu, S. Y.; Li, Y. F.; Zou, H. F.; Qiu, J. S.; Guo, Z.; Guo, B. C. *Anal. Chem.* **2003**, *75*, 6191–6195.
(16) Sunner, J.; Dratr, E.; Chen, Y. C. *Anal. Chem.* **1995**, *67*, 4335–4342.
(17) Wei, L. M.; Xue, Y.; Zhou, X. W.; Jin, H.; Shi, Q.; Lu, H. J.; Yang, P. Y. *Talanta* **2008**, *74*, 1363–1370.
(18) Dattelbaum, A. M.; Hicks, R. K.; Shelley, J.; Koppisch, A. T.; Iyer, S. *Microporous Mesoporous Mater.* **2008**, *114*, 193–200.
(19) Wei, J.; Buriak, J. M.; Siuzdak, G. *Nature* **1999**, *399*, 243–246.

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fine metal or metal oxide powder,[27] etc.) as matrix substances for the analysis of small molecules, have provided new access to the understanding and manipulation of a background-free matrix. However, these inorganic matrixes have not been widely adopted for their less versatility.

Nowadays, rare-earth compounds attract a great deal of attention due to the potential technological applications in opticals,[28−30] biolabels,[31−34] and catalysts[35] based on their unique properties arising from the transitions of 4f electrons. These transitions are parity forbidden, leading to low absorption cross sections and long luminescence lifetimes. However, the long-lived excited state is highly sensitive to the composition and structures of the rare-earth compounds and can be quenched very efficiently in the presence of the high-energy vibrations of organic solvents, polymers, or ligands.[30] Rare earth fluorides possess a high refractive index, low phonon energy, and adequate thermal and environmental stability and are regarded as excellent host lattices for lanthanide ions.[36] It is widely documented that precise control of morphology, dimensionality, and size of the fluorides leads to enhanced luminescent properties of the crystals.[37,38] During the course of our studies of the luminescent properties of rare earth fluorides, we found that the excited-state lifetime of EuF_3 hollow hexagonal microdisks is in the millisecond range. Moreover, these hollow microdisks have a high UV absorbance and can be used as UV absorbents. We expected that these unique hollow microdisks assist the laser desorption/ionization process of analyte molecules deposited on the microdisk surfaces.

In this article, we report the use of EuF_3 hollow hexagonal microdisks for matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF-MS) characterization of small molecules and PEGs. The hollow hexagonal microdisks were fabricated via a facile aqueous synthetic process, which is virtually a cooperation of an oriented aggregation process[39−41] and Ostwald ripening.[42,43] The excited-state lifetime of EuF_3 hollow hexagonal microdisks is 4.04 ms, which provides the potential application in the MALDI analysis of analytes. It is found that these hollow microdisks have an advantage in the MALDI analysis of small peptides and amino acids in the negative-ion reflection mode. The performance of the EuF_3 hollow microdisk matrix was demonstrated by determining a series of analytes, including amino acids, peptides, organic compounds, hydroxypropyl β-cyclodextrin (HP-β-CD), and PEGs. Mass spectra of analytes were observed without the interference of background ions, revealing the matrix of EuF_3 hollow microdisks to be background free.

EXPERIMENTAL SECTION

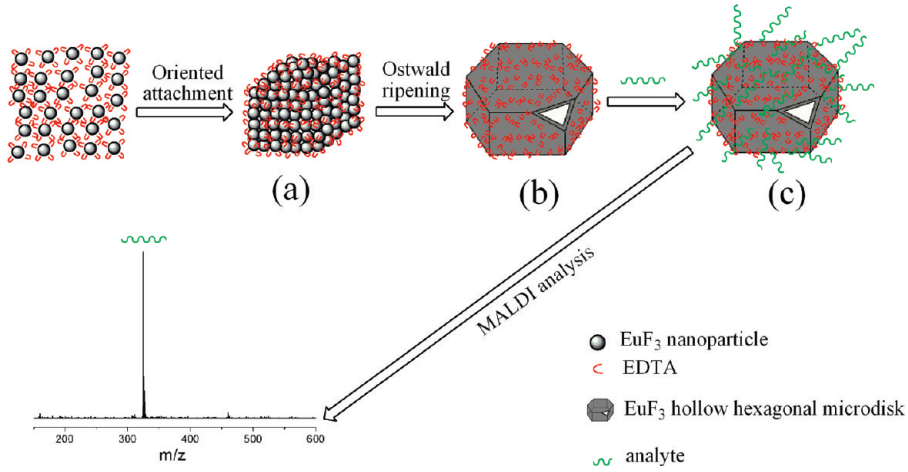
Materials. *N*,*N*′-methylenebisacrylamide, HP-β-CD, acetonitrile (ACN), trifluoroacetic acid (TFA), and the matrixes of α-cyano-4-hydroxycinnamic acid (CHCA) and 2,5-dihydroxybenzoic acid (DHB) were purchased from Sigma (St. Louis, MO). Amino acids of Asn, Glu, His, and Phe were purchased from Fluka (Buchs, Switzerland). Peptides of Val-Gly-Gly, Pro-Glu, Met-Phe, Trp-Leu, and Val-Tyr-Val were obtained from Serva (Feinbiochemica, Heidelberg, Germany). Quinine, propranolol, diantipyrylmethane, Eu_2O_3, ethylenediaminetetraacetic acid (EDTA), and NH_4F were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). PEG 15000 and PEG 30000 were obtained from American Polymer Standards Corp. (Mentor, OH). Other reagents were analytical grade and purchased from Nanjing Chemical Reagent Co., Ltd. (Nanjing, China). The water used throughout the experiment was prepared from a Milli-Q water purification system (Millipore, Milford, MA).

Synthesis of EuF_3 Hollow Hexagonal Microdisks. In a typical synthesis, EDTA solution was obtained by dissolving 500 mg of EDTA in 6.5 mL of 1 M ammonium hydroxide solution. NH_4F solution was obtained by dissolving 200 mg of NH_4F in 7 mL of water/ethanol (2/5, vol/vol). Eu_2O_3 (1 mmol) was completely dissolved in 6.8 mL of 0.74 M HNO_3 to form $Eu(NO)_3$ solution. Then, the EDTA solution and 10 mL of ethanol were added to the obtained $Eu(NO)_3$ solution in sequence. After a 5 min ultrasonic bath, the NH_4F solution was introduced. Subsequently, the mixed system was sonolyzed for 5 min to ensure homogeneous dispersion of all reagents in the solutions and transferred into a Teflon-lined autoclave. After the autoclave was tightly sealed and heated at 110 °C for 12 h, the system was allowed to cool to room temperature naturally. The as-obtained white precipitate was collected, washed with distilled water and absolute ethanol several times, and finally dried at 110 °C in air for 0.5 h. About 177.3 mg of EuF_3 hollow microdisks could be obtained.

Preparation of Analyte Solution. Amino acids of Asn, Glu, His, and Phe and peptides of Val-Gly-Gly, Met-Phe, Trp-Leu, and Val-Tyr-Val were all dissolved in water (0.1% TFA) at the

(20) Lewis, W. G.; Shen, Z. X.; Finn, M. G.; Siuzdak, G. *Int. J. Mass Spectrom.* **2003**, *226*, 107–116.
(21) Zhang, Q. C.; Zou, H. F.; Guo, Z.; Zhang, Q.; Chen, X. M.; Ni, J. Y. *Rapid Commun. Mass Spectrom.* **2001**, *15*, 217–223.
(22) Zou, H. F.; Zhang, Q. C.; Guo, Z.; Guo, B. C.; Zhang, Q.; Chen, X. M. *Angew. Chem., Int. Ed.* **2002**, *114*, 668–670.
(23) Najam-ul-Haq, M.; Rainer, M.; Huck, C. W.; Hausberger, P.; Kraushaar, H.; Bonn, G. K. *Anal. Chem.* **2008**, *80*, 7467–7472.
(24) Kinumi, T.; Shimomae, Y.; Arakawa, R.; Tatsu, Y.; Shigeri, Y.; Yumoto, N.; Niki, E. *J. Mass Spectrom.* **2006**, *41*, 103–112.
(25) Chou, P. H.; Chen, S. H.; Liao, H. K.; Lin, P. C.; Her, G. R.; Lai, A. C. Y.; Chen, J. H.; Lin, C. C.; Chen, Y. *J. Anal. Chem.* **2005**, *77*, 5990–5997.
(26) Lin, P. C.; Tseng, M. C.; Su, A. K.; Chen, Y. J.; Lin, C. C. *Anal. Chem.* **2007**, *79*, 3401–3408.
(27) Kinumi, T.; Saisu, T.; Takayama, M.; Niwa, H. *J. Mass Spectrom.* **2000**, *35*, 417–422.
(28) Wang, X.; Zhuang, J.; Peng, Q.; Li, Y. D. *Inorg. Chem.* **2006**, *45*, 6661–6665.
(29) Heer, S.; Lehmann, O.; Haase, M.; Gudel, H. U. *Angew. Chem., Int. Ed.* **2003**, *42*, 3179–3182.
(30) Stouwdam, J. W.; van Veggel, F. C. J. M. *Nano Lett.* **2002**, *2*, 733–737.
(31) Yi, G. S.; Lu, H. C.; Zhao, S. Y.; Ge, Y.; Yang, W. J.; Chen, D. P.; Guo, L. H. *Nano Lett.* **2004**, *4*, 2191–2196.
(32) Lu, H. C.; Yi, G. S.; Zhao, S. Y.; Chen, D. P.; Guo, L. H.; Cheng, J. *J. Mater. Chem.* **2004**, *14*, 1336–1341.
(33) Sivakumar, S.; Diamente, P. R.; van Veggel, F. C. J. M. *Chem.—Eur. J.* **2006**, *12*, 5878–5884.
(34) Wang, L. Y.; Yan, R. X.; Huo, Z. Y.; Wang, L.; Zeng, J. H.; Bao, J.; Wang, X.; Peng, Q.; Li, Y. D. *Angew. Chem., Int. Ed.* **2005**, *44*, 6054–6057.
(35) Zhou, K. B.; Wang, X.; Sun, X. M.; Peng, Q.; Li, Y. D. *J. Catal.* **2005**, *229*, 206–212.
(36) Li, C.; Quan, Z.; Yang, J.; Yang, P.; Lin, J. *Inorg. Chem.* **2007**, *46*, 6329–6337.
(37) Wang, M.; Huang, Q. L.; Hong, J. M.; Chen, X. T.; Xue, Z. L. *Cryst. Growth Des.* **2006**, *6*, 2169–2173.
(38) Chen, Z. M.; Geng, Z. R.; Shi, M. L.; Liu, Z. L.; Wang, Z. L. *CrystEngComm* **2009**, *11*, 1591−1596.
(39) Penn, R. L.; Banfield, J. F. *Science* **1998**, *281*, 969–971.
(40) Yang, H. G.; Zeng, H. C. *Angew. Chem., Int. Ed.* **2004**, *43*, 5930–5933.
(41) Liu, B.; Zeng, H. C. *J. Am. Chem. Soc.* **2004**, *126*, 8124–8125.
(42) Li, J.; Zeng, H. C. *Angew. Chem., Int. Ed.* **2005**, *44*, 4342–4345.
(43) Li, J.; Zeng, H. C. *Small* **2005**, *1*, 566–571.

Scheme 1. Fabrication and application of EuF_3 hollow hexagonal microdisks as a background-free matrix for MALDI-TOF-MS analysis of small molecules and PEGs: (a) aggregation of EDTA absorbing nanocrystallites into solid cubelike aggregates; (b) creation of a hexagonal microdisk with a hollow interior via Ostwald ripening; and (c) the use of the hollow microdisks as a background-free matrix for MALDI-TOF-MS analysis of small molecules and PEGs



concentration of 5 mM as storage solutions; separately, the mixture solution of the four amino acids and the mixture solution of the four peptides were obtained by mixing storage solutions and diluting with water (0.1% TFA) to the concentration of 1.0 mM each. The storage solutions for quinine, propranolol, *N*,*N*′-methylenebisacrylamide, diantipyrylmethane, His, and Pro-Glu were prepared by dissolving them in water (0.1% TFA) at the concentration of 0.1 mg/mL, separately. The storage solutions for HP-β-CD, PEG 2000, PEG 4000, and PEG 8000 were obtained by dissolving them in water (0.1% TFA) at the concentration of 1.0 mg/mL, separately. The storage solutions for PEG 15000 and PEG 30000 were obtained by dissolving them in water (0.5% TFA) at the concentration of 2.0 mg/mL, separately. All storage solutions were refrigerated at around 4 °C for future use.

Sample Preparation for MALDI-TOF-MS. The matrix of CHCA was prepared as a saturated solution in TA (0.1% TFA in water/acetonitrile, 2/1, vol/vol). The matrix of DHB was obtained by dissolving DHB in TA at 20 mg/mL. The matrix solution of EuF_3 hollow hexagonal microdisks was prepared by dispersing them in acetonitrile with sonication for 2 min at the concentration of 0.5 mg/mL. The solution of analyte (1 μL) was pipetted onto the sample target and left in air at room temperature for 5 to 10 min to form a thin layer of analyte by evaporating water. Then, a 1 μL solution of the matrix was pipetted onto the layer of analyte and left in air for 5−10 min for the evaporation of the solvent and for further analysis by MALDI-TOF-MS.

Characterization. X-ray powder diffraction (XRD) patterns of the products were recorded on a Shimadzu XRD-6000 X-ray diffractometer with Cu Kα radiation (λ = 0.15406 nm) at a scanning rate of 0.1° s^{-1} in the 2θ range from 20 to 80°. Transmission electron microscopy (TEM) images were obtained on a JEM-200CX transmission electron microscope, with an accelerating voltage of 200 kV. Scanning electron microscopy (SEM) images were acquired from a Hitachi S-4800 scanning electron microscope. High-resolution transmission electron microscopy (HRTEM) images were taken on a FEI Tecnai G2 20 S-Twin instrument with a field emission gun operating at 200 kV. A luminescent spectrum was carried out on an Aminco Bowman luminescence spectrometer equipped with a 150 W Xe arc lamp at room temperature. The luminescence lifetime of Eu^{3+} was measured using a Cary-Eclipse spectrofluorimeter with a 15 W Xe pulse lamp as the excitation

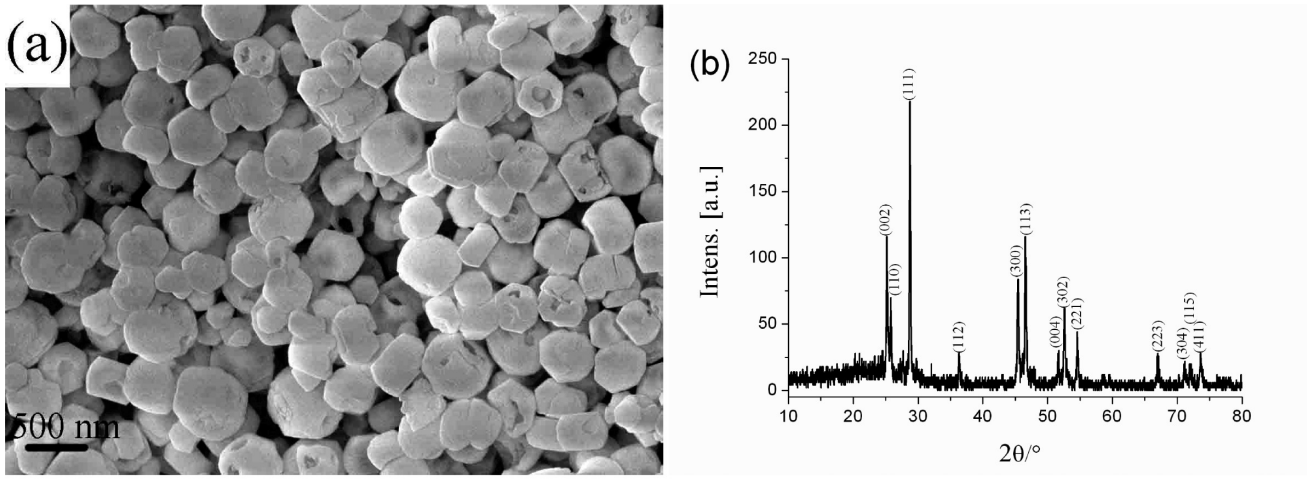


Figure 1. (a) SEM image and (b) XRD pattern for EuF_3 hollow hexagonal microdisks.

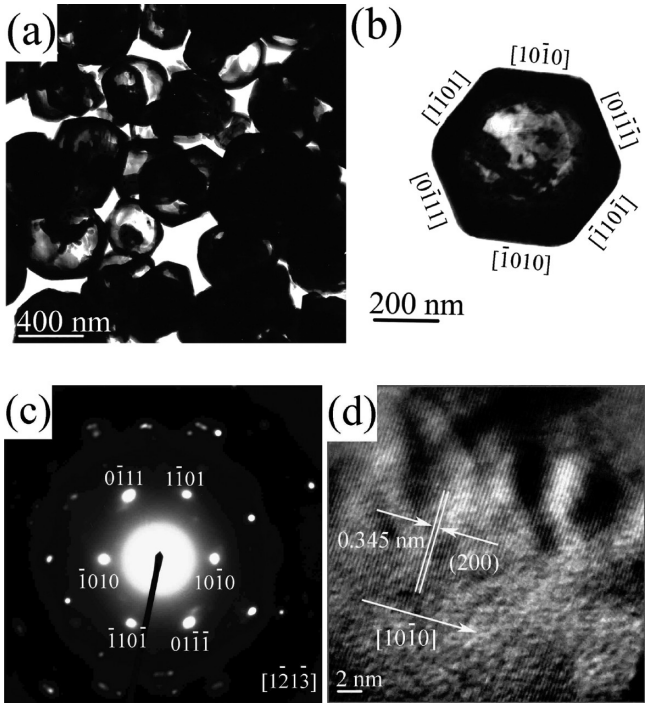


Figure 2. (a, b) Low-magnification TEM images of EuF_3 hollow hexagonal microdisks; (c) SAED patterns for an individual hollow hexagonal microdisk oriented along [$1\bar{2}1\bar{3}$]; (d) HRTEM image of the EuF_3 hollow hexagonal microdisk oriented along [$10\bar{1}0$].

source. An FT-IR spectrum was obtained on a Vectortm 22. Mass spectra were performed on Bruker Autoflex with a nitrogen laser (λ = 337 nm) at −20 − +20 kV. The MALDI used a ground-steel sample target with 384 spots. The analytical range of laser energy was adjusted to slightly above the threshold to obtain good resolution and signal-to-noise ratio (S/N).

RESULTS AND DISCUSSION

As described in Scheme 1, EuF_3 hexagonal microdisks with hollow interior were fabricated for use as a matrix in direct identification of small molecules and PEGs. Figure 1a shows field-emission SEM images of the product and reveals that the sample is in the form of hexagonal microdisks with an average width of 440 ± 60 nm and length of 300 ± 60 nm. The corresponding XRD result of the product is indexed to EuF_3 with a hexagonal structure (Figure 1b). All the positions of the peaks are in good agreement with values in the Joint Committee on Powder Diffraction Standards (JCPDS) card file no. 32-0373.

TEM images clearly indicate that these hexagonal microdisks exhibit an obvious hollow structure, and the shell walls are approximately 40−60 nm thick (Figure 2). Figure 2b shows a free-standing single microdisk. The regular hexagonal cross section is the projection view of the microdisk standing on the bottom face. The corresponding selected area electron diffraction (SAED) pattern (Figure 2c), indicating that the microdisk is a single crystal, is indexed to the [$1\bar{2}1\bar{3}$] zone of hexagonal EuF_3. The HRTEM image of the edge area of the hollow microdisk in Figure 2b shows that the microdisk is a single crystalline with a periodic fringe spacing of 0.345 nm (Figure 2d), which corresponds to the interplanar spacing between the {200} planes of the hexagonal EuF_3. The time-dependent crystal morphologies of

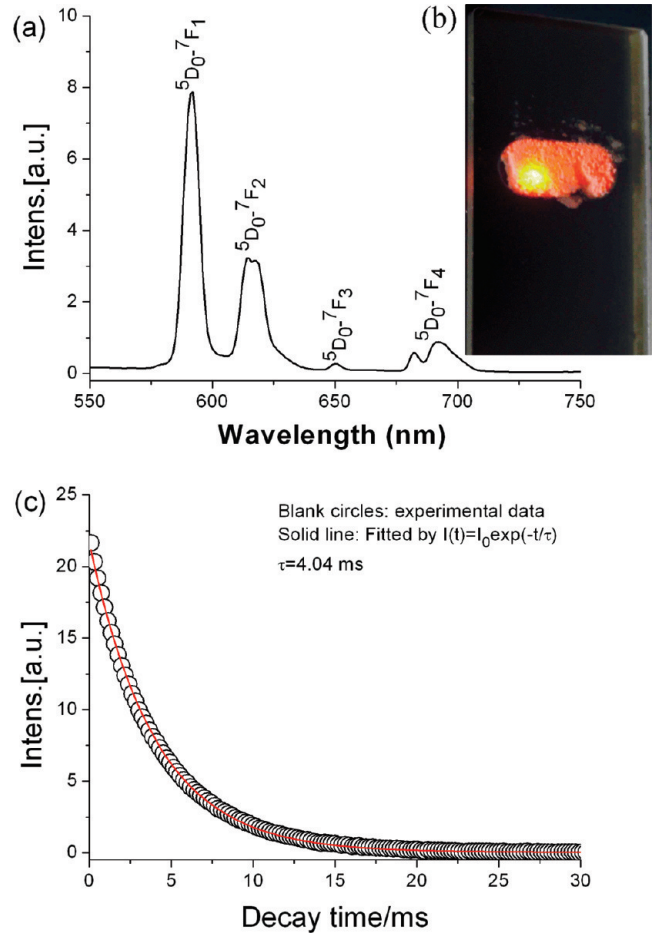


Figure 3. (a) Luminescence spectrum of EuF_3 hollow hexagonal microdisks upon 394 nm UV excitation; (b) the red luminescence observed from the microdisks upon excitation with a 325 nm laser; (c) decay curves of Eu^{3+} luminescence (592 nm) upon 394 nm UV excitation.

the samples indicate that the formation of EuF_3 hollow hexagonal microdisks is virtually a cooperation of an oriented aggregation process and Ostwald ripening (see Supporting Information, Figure S1).

These hexagonal microdisks emit strong and red fluorescence with a symmetric and narrow emission spectrum upon 310−500 nm UV excitation, which makes them suitable as the energy receptacle for UV radiation. The strong red luminescence from the solid sample of the microdisks upon excitation with a 325 nm laser is easily visible to the naked eye (Figure 3b). Figure 3a shows that these hexagonal microdisks have characteristic emission peaks of Eu^{3+} within the wavelength range from 550 to 720 nm, corresponding to the transitions of the excited levels of $5D_0 \rightarrow 7F_{(1-4)}$. The intensity of the $5D_0 \rightarrow 7F_1$ transition (592 nm) greatly exceeds the intensity of the $5D_0 \rightarrow 7F_2$ transition (614−617 nm), indicating that the europium ions in the hollow microdisks are the centrosymmetrical structure.[44] Figure 3c displays the typical luminescent dynamics of the $5D_0 \rightarrow 7F_1$ transition of Eu^{3+}. These curves can be well fitted by a single-exponential function as $I(t) = I_0 \exp(-t/\tau)$ (I_0 is the initial emission intensity at $t = 0$, and τ is the 1/e lifetime of the emission center). Therefore, the luminescence lifetimes of the $5D_0 \rightarrow 7F_1$ transition of Eu^{3+} is 4.04 ms.

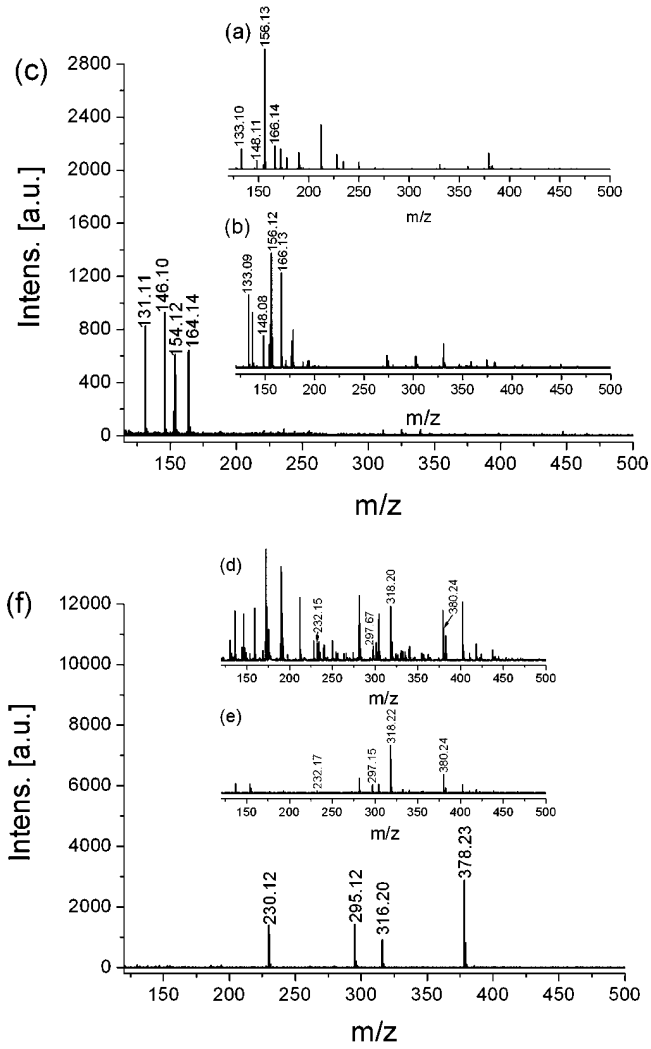


Figure 4. Comparison of (a) CHCA, (b) DHB, and (c) EuF_3 hollow hexagonal microdisks as the matrix for the detection of an amino acid mixture solution containing Asn, Glu, His, and Phe; comparison of (d) CHCA, (e) DHB, and (f) EuF_3 hollow hexagonal microdisks as the matrix for the detection of a peptide mixture containing Val-Gly-Gly, Met-Phe, Trp-Leu, and Val-Tyr-Val. (a, b, d, e) Detected in the positive-ion reflection mode; (c, f) detected in the negative-ion reflection mode.

After structural characterization, EuF_3 hollow microdisks were employed as the matrix for MALDI-TOF-MS analysis of small molecules. In order to evaluate the performance of EuF_3 hollow microdisks as a background-free ionization element, an amino acid mixture containing Asn, Glu, His, and Phe, and a peptide mixture containing Val-Gly-Gly, Met-Phe, Trp-Leu, and Val-Tyr-Val, were chosen as models. We compared the MALDI-TOF mass spectra obtained from the conventional matrixes CHCA, DHB, and EuF_3 hollow microdisks. As shown in Figure 4, all the analytes were detected as $[M + H]^+$ (Asn, 133; Glu, 148; His, 156; Phe, 166; Val-Gly-Gly, 232; Met-Phe, 297; Trp-Lea, 318; Val-Tyr-Val, 380), but strong interference of the backgrounds caused by the matrixes of CHCA (Figure 4a,d) and DHB (Figure 4b,e) makes it difficult to identify analytes. Moreover, the suppression effects among analyte molecules result in the poor

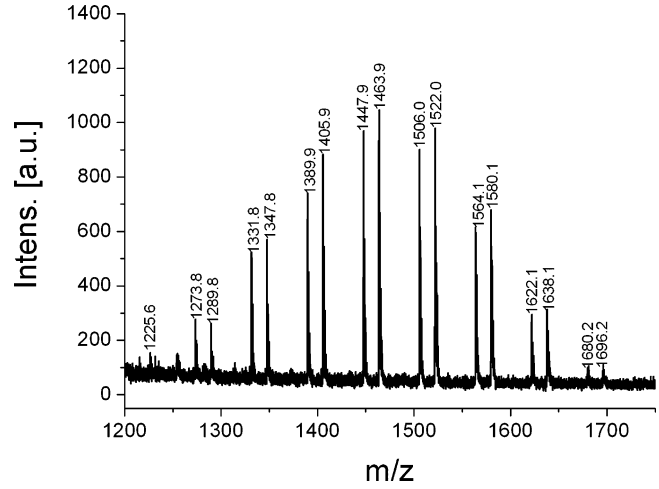


Figure 5. Mass spectrum of HP-β-CD with the matrix of EuF_3 hollow hexagonal microdisks. Peaks of *m*/*z* at 1226, 1274, 1332, 1390, 1448, 1506, 1564, 1622, and 1680 are indexed to the Na^+ adduct ions for one molecule of β-CD reacted with n ($n = 1$, ..., 9) molecules of epoxy propane, and peaks of *m*/*z* at 1290, 1348, 1406, 1464, 1522, 1580, 1638, and 1696 are attributed to the K^+ adduct ions for one molecule of β-CD reacted with n ($n = 2$, ..., 9) molecules of epoxy propane.

Table 1. Intensity, the Ratio of Signal to Noise Ratio (S/N), and the Resolution of Peaks for Analytes by MALDI-TOF-MS with Matrix of EuF_3 Hollow Hexagonal Microdisks[a]

	m/*z*	intensity	S/N	resolution
quinine	325, $[M + H]^+$	780.7	291.2	3358
propranolol	260, $[M + H]^+$	2388.8	298.5	2098
	282, $[M + Na]^+$	413.3	51.8	1971
	298, $[M + K]^+$	81.3	9.9	4070
diantipyrylmethane	389, $[M + H]^+$	895	149.6	2360
	411, $[M + Na]^+$	2509.3	342.9	2720
	427, $[M + K]^+$	2516.6	465.1	2760
N,*N*′-methylenebisacrylamide	155, $[M + H]^+$	71.2	8.9	1317
	177, $[M + Na]^+$	3318.6	361.4	1709
	193, $[M + K]^+$	1179.2	113.7	1709
histidine	154, $[M - H]^-$	2221.5	398.9	1643
Pro-Glu	243, $[M - H]^-$	5349	402.1	3393

[a] All analytes were at a concentration of 0.1 mg/mL.

reproducibility and resolution of peaks for analytes (Figure 4a,e). It is clear that the mass spectrum of EuF_3 hollow microdisks with the peptide mixture and the amino acid mixture were obtained in the negative-ion reflection mode. Four prominent peaks without the interference of background ions for Asn (131, $[M - H]^-$), Glu (146, $[M - H]^-$), His (154, $[M - H]^-$), and Phe (164, $[M - H]^-$) were observed as displayed in Figure 4c, and all the peaks with excellent signal-to-noise ratio for Val-Gly-Gly (230, $[M - H]^-$), Met-Phe (295, $[M - H]^-$), Trp-Lea (316, $[M - H]^-$), and Val-Tyr-Val (378, $[M - H]^-$) were well detected as shown in Figure 4f, suggesting that EuF_3 hollow microdisks should be a "negative-ion" matrix for MALDI-TOF-MS analysis of amino acids and small peptides.

To demonstrate the general applicability of the matrix of EuF_3 hollow microdisks, we analyzed six small-molecule compounds with distinct chemical structures, including quinine, propranolol, *N*,*N*′-methylenebisacrylamide, diantipyrylmethane, histidine, and Pro-Glu (0.1 mg/mL). The matrix of EuF_3 hollow microdisks generated very clean spectra that show the pres-

(44) Zhu, L.; Liu, X. M.; Meng, J.; Cao, X. Q. *Cryst. Growth Des.* **2007**, *7*, 2505–2511.

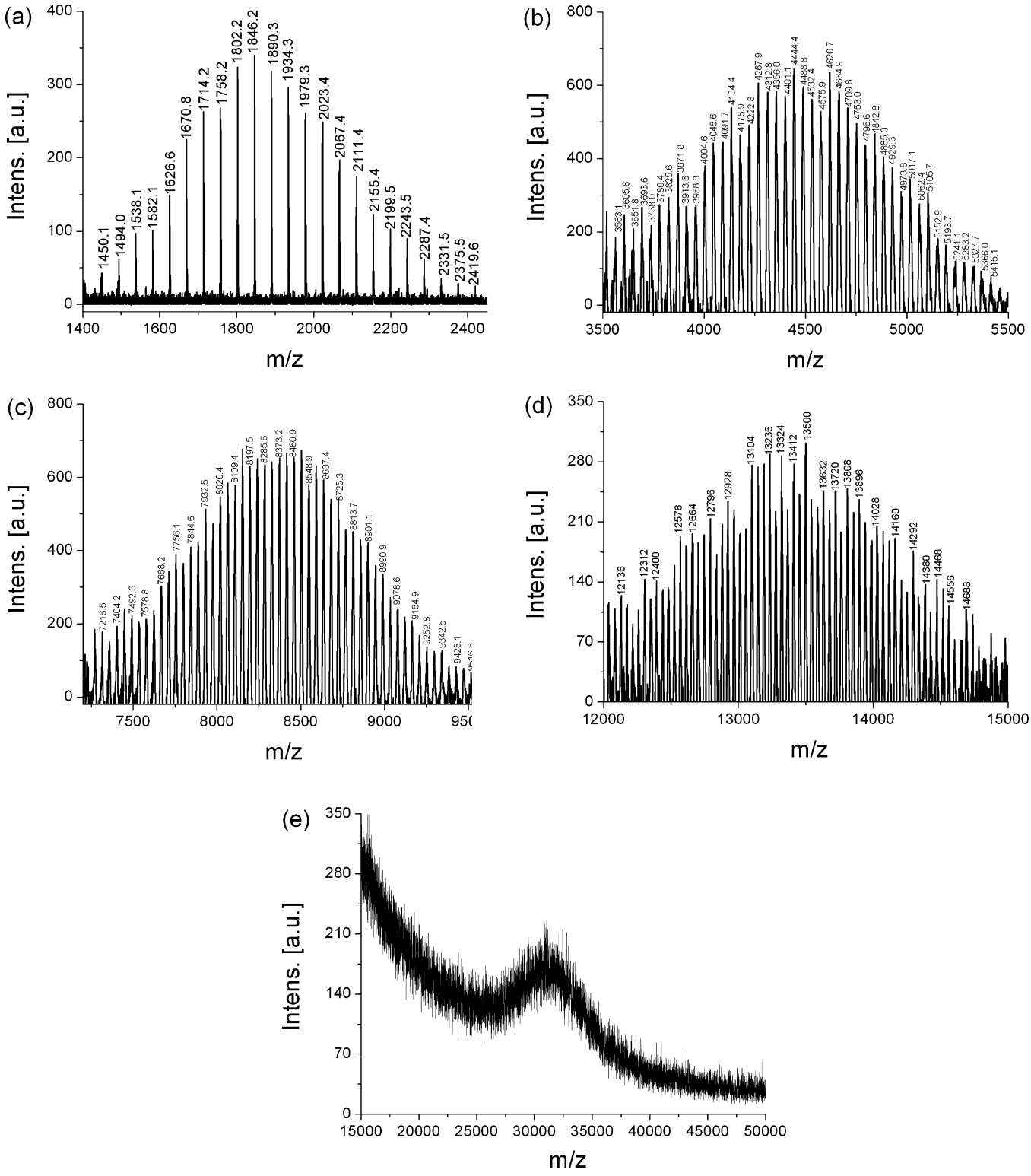


Figure 6. Mass spectra of (a) PEG 2000, (b)PEG 4000, (c) PEG 8000, (d) PEG 15000, and (e) PEG 30000 with the matrix of EuF_3 hollow hexagonal microdisks.

ence of only the small-molecule compound-related peaks for each analyte, including the cationic parent, sodium adduct, potassium adduct, and anion parent (see Supporting Information, Figure S2). The intensity, the ratio of signal-to-noise (S/N), and the resolution of peaks for the six small molecule compounds are listed in Table 1. As shown in Table 1, all parameters for mass spectra demonstrated the suitability of the matrix of EuF_3 hollow microdisks. The intensity and S/N of peaks for analytes are high, suggesting that the efficiency for the desorption/ionization of the analytes on the surface of EuF_3 hollow microdisk is prominent, which might be explained by the following factors: First, the special unpaired 4f electrons of Eu^{3+} could make EuF_3 hollow microdisk function as an energy receptacle for UV laser radiation. Subsequently, the long-lived

excited states of europium ions transfer energy to analyte molecules. Also, the carboxylic groups (originated form EDTA, see Supporting Information, Figure S3) on the surfaces of the hollow microdisks could provide analytes with plenty of H^+, Na^+, K^+, or $-COO^-$ to form $[M + H]^+$, $[M + Na]^+$, $[M + K]^+$, or $[M - H]^-$ during the process of desorption/ionization.

MALDI-TOF-MS has an advantage in the high-speed determination of structural information, molecular weight averages, and purity of macromolecules. We extended the application of the EuF_3 hollow microdisk matrix to the detection of HP-β-CD, to evaluate the availability of this matrix for analysis of synthetic compounds. Figure 5 shows the mass spectrum of HP-β-CD with the matrix of EuF_3 hollow hexagonal microdisks, in which a series of compounds of HP-β-CD with an interval of 58 Da were examined in positive-ion reflection mode. Peaks of m/z at 1226, 1274, 1332, 1390, 1448, 1506, 1564, 1622, and 1680 could be easily indexed to the Na^+ adduct ions for one molecule of β-CD reacted with n (n = 1, ..., 9) molecules of epoxy propane, and peaks of m/z at 1290, 1348, 1406, 1464, 1522, 1580, 1638, and 1696 could be attributed to the K^+ adduct ions for one molecule of β-CD reacted with n (n = 2, ..., 9) molecules of epoxy propane, suggesting that there are 1−9 hydroxypropyl groups in this kind of HP-β-CD.

One of the most frequently used polymers in MALDI-TOF-MS is PEG, which is a simple molecule of the form $HO-(CH_2CH_2O)_n-H$, and is widely used in pharmaceutics, cosmetics, and the textile and leather industries. We characterized PEG 2000, PEG 4000, PEG 8000, PEG 15000, and PEG 30000 with the matrix of EuF_3 hollow microdisks to explore the application of this matrix in the detection of synthetic polymers. Figure 6 shows the mass spectra of PEG 2000, PEG 4000, PEG 8000, PEG 15000, and PEG 30000 with the matrix of EuF_3 hollow microdisks, indicating that the upper limit of the detectable mass range was ~35000 Da. It is noted that the mass ranges of PEG 2000, PEG 4000, PEG 8000, PEG 15000, and PEG 30000 are at 1404−2420 Da, 3519−5327 Da, 7129−9561 Da, 12000−15000 Da, and 25000−35000 Da, respectively, with the matrix of EuF_3 microdisks, but with the matrix of DHB they are at 1405−2773 Da, 4407−5996 Da, 7185−9560 Da, 13000−15000 Da, and 18000−33000 Da (see Supporting Information, Figure S4), respectively. The number average molecular weight (M_n), mass average molecular weight (M_w), and polydispersity (pd) of PEG 2000, PEG 4000, PEG 8000, and PEG 15000 were analyzed and listed in Table 2. The M_n of a polymer is often found to vary when different matrixes are used.[45] The M_n of the PEGs analyzed in the EuF_3 microdisk matrix are lower than the same PEG samples analyzed in the DHB matrix, suggesting that it requires a higher laser energy to ablate the PEGs into the gas phase in the matrix of EuF_3 microdisks. The pd of each PEG is ≤0.013 in the EuF_3 microdisk matrix, indicating that EuF_3 microdisks have an advantage as a matrix of PEGs.

(45) Wetzel, S. J.; Guttman, C. M.; Girard, J. E. *Int. J. Mass Spectrom.* **2004**, *238*, 215–225.

Table 2. Number Average Molecular Weight (M_n), the Mass Average Molecular Weight (M_w), and the Polydispersity (pd) of PEG 2000, PEG 4000, PEG 8000, and PEG 15000 in the Matrixes of EuF_3 Hollow Hexagonal Microdisks and DHB

polymer	M_n (Da)	M_w (Da)	pd
PEG 2000[a]	1903.15	1928.30	1.013
PEG 2000[b]	2083.22	2109.68	1.013
PEG 4000[a]	4453.63	4482.16	1.006
PEG 4000[b]	5015.35	5027.55	1.002
PEG 8000[a]	8365.34	8386.42	1.003
PEG 8000[b]	8421.89	8443.56	1.003
PEG 15000[a]	13410.8	13430.0	1.001

[a] PEGs were analyzed in a EuF_3 microdisk matrix. [b] PEGs were analyzed in a DHB matrix.

CONCLUSION

In summary, single-crystalline EuF_3 hollow hexagonal microdisks were fabricated via a straightforward method. These hollow microdisks were employed as an ionization platform, in which the hollow microdisk matrix functions as the energy receptacle for UV laser radiation and the energy transporter for the desorption/ionization of analytes with the minimization of interference signals caused by the matrix ion. The suitability of the hollow microdisk matrix was demonstrated by MALDI-TOF-MS analysis of a series of analytes, including small peptides, amino acids, organic compounds, HP-β-CD, an amino acid mixture, and a peptide mixture. The practical application in determining low molecular weight analytes shows that this matrix has an advantage in the analysis of small peptides and amino acids in the negative-ion reflection mode. This matrix is successfully used for analysis of PEGs, suggesting a potential for monitoring reactions and for synthetic polymer quality control. Though further development is still needed, EuF_3 hollow microdisks can now be considered as a novel background-free matrix for MALDI-TOF-MS analysis of small molecules and PEGs.

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SUPPORTING INFORMATION AVAILABLE

TEM images for EuF_3 products at different preparation stages; mass spectra of quinine, propranolol, diantipyrylmethane, *N*,*N*′-methylenebisacrylamide, histidine, and Pro-Glu with matrix of EuF_3 hollow hexagonal microdisks; FT-IR spectrum of EuF_3 hollow hexagonal microdisks; mass spectra of PEG 15 000 and PEG 30 000 with matrix of DHB. This material is available free of charge via the Internet at http://pubs.acs.org.

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