Ultrasound assisted combined molecularly imprinted polymer for selective extraction of nicotinamide in human urine and milk samples: Spectrophotometric determination and optimization study

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A B S T R A C T

Ultrasound-assisted dispersive solid phase microextraction followed by UV–vis spectrophotometer (UA-DSPME-UV–vis) was designed for extraction and preconcentration of nicotinamide (vitamin B_3) by HKUST-1 metal organic framework (MOF) based molecularly imprinted polymer (MIP). This new material was characterized by FTIR and FE-SEM techniques. The preliminary Plackett–Burman design was used for screening and subsequently the central composite design justifies significant terms and possible construction of mathematical equation which give the individual and cooperative contribution of variables like HKUST-1-MOF-NA-MIP mass, sonication time, temperature, eluent volume, pH and vortex time. Accordingly the optimum condition was set as: 2.0 mg HKUST-1-MOF-NA-MIP, 200 μL eluent and 5.0 min sonication time in center points other variables were determined as the best conditions to reach the maximum recovery of the analyte. The UA-DSPME-UV–vis method performances like excellent linearity (LR), limits of detection (LOD), limits of quantification of 10–5000 μg L^{-1} with R^2 of 0.99, LOD (1.96 ng mL^{-1}), LOQ (6.53 μg L^{-1}), respectively show successful and accurate applicability of the present method for monitoring analytes with within- and between-day precision of 0.96–3.38%. The average absolute recoveries of the nicotinamide extracted from the urine, milk and water samples were 95.85–101.27%.

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1. Introduction

Human health strongly depends on the presence and level of different vitamins in their diet [1]. These active organic compounds (essential micronutrients) with serious physiological functions [2] may be water soluble and/or fat-soluble vitamins [3]. Water-soluble vitamins viz. thiamine (B_1), riboflavin (B_2), nicotinamide (B_3), pantothenic acid (B_5), pyridoxine (B_6), biotin (B_7), folic acid (B_9), cyanocobalamin (B_{12}) and ascorbic acid (C) [4,5] have a distinguish role in the best operation of metabolism, muscle tone, immune and bone health, nervous system, synthesis of DNA and RNA, and cell growth [6,7]. Nicotinamide (vitamin B_3) slows refuse from skin aging by elimination of wrinkles, improvement of the skin appearance and elasticity, while is an anti-inflammatory agent and improve the skin appearance by reducing the activity of leukocyte per-oxidase [8–10]. Vitamin level monitoring and assessment in food samples has great interest in the health and nutritional fields and food industry. In this regard, various

protocols like microbiological assays [11], spectrofluorimetry [12], spectrophotometry [9], thin layer chromatography (TLC) [4], high-performance liquid chromatography (HPLC) [13–15], ultrahigh-pressure liquid chromatography (UHPLC) [1], hydrophilic-interaction liquid chromatography (HILIC) [16,17], capillary electrophoresis (CE) [18], electrochemical [19] and Fourier transform infrared (FT-IR) [20] have been used for such a purpose. Some of these methods require expensive instrumentation, tedious preliminary pretreatment and high cost of maintenance and high operational cost and/or consumption of large amounts of materials, while spectrophotometric methods with low-cost and more available instruments are useful for simple and accurate monitoring of analytes although suffer from lower sensitivity and overlapped peaks [21,22]. In most cases, the low concentration of analyte and their complex matrix strongly limit and lower the characteristic performance of most analytical methods and encourage the researchers to supply preliminary separation and/or preconcentration techniques like solid-phase extraction (SPE) [15], dispersive liquid-liquid microextraction (DLLME) [6], electrochemical [19] and ion-pair cloud-point extraction (IP-CPE) [23] to overcome such limitations. These methods despite their unique advantages and

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merits may suffer from tedious stages and more expensive instruments, which extensively can be reduced or replaced by ultrasonic-assisted solid phase microextraction combined with UV–vis spectrophotometry (UA-DSPME-UV–vis). This case is highly recommended in terms of unique possible advantages like simplicity, low-cost, and ease of miniaturization for small-volume samples [24–29]. Application of sonication leads to appearance of very fine emulsions from immiscible liquids and leads to significant improvement in the interfacial contact areas between the liquids and a dramatic increase in the mass transfer among them. The cavitation which appears following ultrasound exposure leads to more available surface area and facilitates the release of interesting extractable compounds and improves the procedure extraction efficiency and efficiency can be increased according to the best selection of adsorbent and method [28,30,31]. Molecularly imprinted polymers (MIPs) are superior to previous adsorbents like anthracite particles [32], C18 cartridges and [33] in terms of selectivity [34,35]. MIP as a versatile and easy pattern leads to building selective recognition sites in a stable polymer matrix which subsequently leads to more successful and accurate monitoring of analyte especially in complicated matrix [36,37] and recent MIP preparation via surface molecular imprinting polymers causes improvement in figures of merit like selectivity [38–40] and reducing the extraction time by improvement in mass transfer, while the stability and regenerability of a sorbent can be improved following imprinting at the surface of a support and closeness of the reactive center to the bulk to make possible more and efficient accumulation of analyte into the MIP [41]. Therefore, HKUST-1 MOF as a porous structure material is possible for versatile controllable modification which emerged from flexible, porous and strong structure of MOFs which lead to high adsorption properties of the surface bonded and cause good recovery, high efficiency and easy phase separation.

The Plackett–Burman designs preliminarily applied for detection of significant factors and also to discriminate among variable influence on method efficiency and subsequently lead to the best optimization of operational parameter at a low cost and in a reasonable time [28,42].

Response surface methodology (RSM) is a good method for optimizing multifactor and evaluating the effects of several factors as individual and interaction part which possible achievement of real and reliable optimum point which enable best prediction of behavior of understudy system [43,44]. Among RSM designs, central composite design (CCD) due to its simple structure and efficiency is a good pathway which efficiently correlates the response to significant term as a consequence of main and combination part [45]. To the best of our knowledge there is no report on application of UA-DSPME-UV–vis for analysis of nicotinamide, which encourages our activity for preconcentration and subsequent determination of nicotinamide (vitamin B_3) content following response surface methodology (RSM) optimization. An advantage of central composite design is to reduce the number of experimental works, while the experimental design includes a wider region of parameter space and replicated center point provides excellent prediction capability near the center of the design space [46,47].

In this study, the extraction efficiency of UA-DSPME-UV–vis method for the analysis of nicotinamide (vitamin B_3) in milk and urine samples following optimization of the effect of variables by the design of experiments (DOE) has been investigated. Firstly, a Plackett–Burman design (2^{6-3}) was used for selection of main effective parameters and subsequently the numerical contribution to the response was examined by CCD. Finally, the performance of the method for the analysis of water and wastewater samples, milk and human urine was evaluated.

2. Experimental

2.1. Chemicals and reagents

NaOH and HCl with the highest purity available were purchased from Merck Co. (Darmstadt, Germany). Copper (II) nitrate hemipentahydrate, nicotinamide (vitamin B_3), benzene-1,3,5-tricarboxylic acid (trimeric acid), acetonitrile, acetone, methanol, ethanol, tetrahydrofuran and N,N-dimethyl form amide (DMF) were purchased from Sigma–Aldrich Company (American). Urine sample was collected from a healthy person (6 years old). Milk sample from a woman (27 year old) was collected at the start of the lactation. Stock solutions of nicotinamide were prepared as 100 mg L^{-1} in double distilled water. Then, the required working standard solutions were freshly prepared by appropriate dilution of the stock solutions.

2.2. Apparatus

The absorbance was measured with a Perkin Elmer Lambda 25 spectrophotometer at a wavelength of 259 nm using a quartz microcell with an optical path of 1 cm. The morphology of the nanoparticles were observed by field emission scanning electron microscopy (FE-SEM: Hitachi S4160, Japan) under an acceleration voltage of 15 kV. To investigate the purity as well as the presence of organic and/or other compounds in the prepared nanoparticles, a Fourier transform infrared (FT-IR) spectrum was recorded using a Perkin Elmer-Spectrum spectrometer (RX-IFTIR, USA) in the range of 300–4000 cm^{-1}. Ultrasonic device (TECNO-GAZ, 60 Hz, 130 W, Parma, Italy) is equipped with digital timer and temperature controller. A Ino Lab pH 730 digital pH meter (Germany) was used for pH adjustments. A HERMLE bench centrifuge (2206A, Germany) was used to accelerate the phase separation. A 10–100 µL Hamilton syringe (Hamilton Company, Nevada) was used for phase separation of collected sediments.

2.3. Preparation of HKUST-1-MOF-NA-MIP

HKUST-1 MOF was prepared by ultrasound-assisted solvothermal method as follow: benzene-1,3,5-tricarboxylic acid (3.6 mmol) and copper (II) nitrate hemipentahydrate (10 mmol) were sonicated for 1.0 h in 100 mL of solvent consist of 80 mL DMF and 20 mL ethanol. Subsequently, the mixture was transferred into a Teflon-lined autoclave, sealed and heated at 130 °C for 24 h to yield octahedral crystals. Finally, the solvent was removed and heated in an oven at 100 °C for 12 h to yield a porous material. The HKUST-1-MOF-NA-MIP was prepared by surface imprinting of the HKUST-1 as follows: a mixture of 0.5 g of HKUST-1 and 80 mL of acetonitrile/methanol (60/40 V:V) was prepared and sonicated in a round bottom flask. Then, 0.2 mmol of NA as the template, 1.0 mmol of MAA as functional monomer, 4.0 mmol of EGDMA and 40 mg of AIBN were added to the flask. The deoxygenation of the solution was undertaken by purging the mixture with N_2 for 15 min. Then, the flask was well-sealed and stirred in an oil bath at 70 °C for 24 h. After surface polymerization product was collected using an external magnetic field and washed several times with acetonitrile/methanol under irradiation of ultrasonic waves in a sonication bath to remove the unreacted materials, and then dried in an oven at 70 °C 12 h. Then, for leaching the nicotinamide trapped in the polymer, methanol/acetic acid (9:1, v/v) was used until achieving a nicotinamide-free infiltrated solution. The final powder product, as HKUST-1-MOF-NA-MIP, was dried at 50 °C under vacuum for 12 h. For comparison, non-imprinted polymer (NIP) was also prepared as a blank in parallel but without the addition of nicotinamide.